RESEARCH PROTOCOL and STATISTICAL ANALYSIS PLAN

December 7, 2021

and

INFORMED CONSENT FORM

December 3, 2019

Pharmacokinetics of Benzo[a]Pyrene: Impact of Diet

Principal Investigator: David E. Williams, PhD

NCT03802721

APPLICATION & PROTOCOL

This new form reflects changes in federal and local policies and procedures and will serve as a bridge between the previous forms and the eventual web-based system. Your feedback on the content of this form is vital during this transition. Please email your questions, suggestions, and training requests to irb@oregonstate.edu

Thank you for collaborating with the Human Research Protection Program to continuously improve the submission process.

Contents – *to link directly to a specific section, **hold 'ctrl' and click** the section in the table of contents*

APPLICATION & PROTOCOL

APPLICATION & PROTOCOL

 Additional information or instructions

 Link to information or additional forms

 Only type into shaded rows that include this pencil icon

 External document may need to be submitted

SECTION 1 - Submission Type		Check ONE
	Track all changes after the initial submission	
1	New submission, not previously reviewed or approved by OSU	☒
2	Re-submission of an expired protocol	☐
3	Previously approved protocol with proposed changes (project revision or minor change)	☐
SECTION 2 - Study Title and Team		
1	Study Title	
	Pharmacokinetics of benzo[*a*]pyrene: impact of diet	
2	Name of Principal Investigator	
	David E. Williams, PhD	
	Submissions will only be reviewed when received *directly* from the PI.	
	FAQ Who can be a Principal Investigator (PI)?	
	FAQ Who must be listed as study team members? This FAQ will also assist you in determining whether external collaborators must be listed here or whether some collaborators may be certified by the PI after approval.	
	Do not list individuals who will receive IRB approval at their own external institution or whose institution has determined that they are not engaged	
3	Name of additional study team member(s) to be copied on correspondence. *If external to OSU, provide email address.*	
	Sandra Uesugi	

4	Name of additional study team member(s) who will not be copied on correspondence. If external to OSU, provide email address.			
	Lisbeth Siddens,			
	If this is a project revision and you are adding or removing study team members, confirm that the **track change** feature of MS Word is on before you make these changes.			
SECTION 3 - Study Summary				
1	Using lay language, briefly describe the study purpose or primary research question:			
	We will study how our bodies absorb and eliminate a common pollutant called benzo[a]pyrene (BaP). We will study how the body handles low levels of BaP and how this might be affected by cruciferous vegetables like Brussels sprouts. BaP is in the family of PAH compounds, which come from burning material like cigarettes or coal.			
	You will be asked for aims, background justification, and specific methods and procedures in later sections.			
SECTION 4 - Determination of Whether the Project Requires IRB Review		Yes	No	N/A
	What types of projects require IRB review?			
SECTION 4.1 - Research		Yes	No	N/A
1	"Research" is defined as a systematic investigation, including research development, testing and evaluation, designed to develop or contribute to generalizable knowledge. Does the project involve research?	☒	☐	
	**Systematic Investigation** Typically predetermined method for studying a specific topic, answering a specific question(s), testing a specific hypothesis, or developing theory. A scientific or scholarly activity involving qualitative or quantitative data collection and/or data analysis that sets forth an objective(s) and a set of procedures intended to reach the objective(s), i.e., to acquire knowledge, develop a theory, or to answer a question. **Includes**: Observational studies, interview or survey studies, group comparison studies, pilot studies, test development and interventional research. **Generalizable Knowledge** The intent or purpose of the systematic investigation is dissemination of findings (publication or presentation) outside of OSU. Intended to have an impact (theoretical or practical) on others within one's			

	discipline. Dissemination with the intent to influence behavior, practice, theory, future research designs, and the like, are contributing to generalizable knowledge. **Research does NOT include**: Class projects, some program evaluation, or an examination of just one person. Scholarly and journalistic activities (e.g., oral history, journalism, biography, literary criticism, legal research, and historical scholarship), including the collection and use of information, that focus directly on the specific individuals about whom the information is collected.			
2	If you think the project does NOT involve research, please explain why and provide the relevant project details:			☒
				
SECTION 4.2 – Human Subjects		Yes	No	N/A
1	"Human subject" is defined by obtaining data **about**, or specimens from, one or more living individuals through: • intervention, OR • interaction, OR • the collection of identifiable private information Does the project involve human subjects?	☒	☐	
	**Human Subject** A living individual about whom an investigator conducting research obtains: • data through intervention or interaction with the individual; **or** • identifiable private information or identifiable biospecimens **Intervention**: Includes physical procedures by which data are gathered and manipulation of the subjects or the subjects' environment that are performed for research purposes. **Interaction**: Includes communication or interpersonal contact between investigator and subjects. The interaction may be as remote as an anonymous, online survey. **Private information:** Private information includes information about behavior that occurs in a context in which an individual can reasonably expect that no observation or recording is taking place, and information which has been provided for specific purposes by an individual and which the individual can			

	reasonably expect will not be made public (for example, a medical record). Private information must be individually identifiable (i.e., the identity of the subject is or may readily be ascertained by the investigator or associated with the information) in order for obtaining the information to constitute research involving human subjects. **Identifiable private information:** Identifiable private information is private information for which the identity of the subject is or may readily be ascertained by the investigator or associated with the information. **Identifiable biospecimen:** An identifiable biospecimen is a biospecimen for which the identity of the subject is or may readily be ascertained by the investigator or associated with the biospecimen. **NOT** considered to include human subjects: Projects limited to pre-existing data or samples that were (1) not collected for the current project, **and** (2) not collected by the team members on this project, **and** (3) de-identified by someone who is not associated with the current project.			
2	If you think the project does NOT involve human subjects, please explain why and provide the relevant project details:			☒
				
SECTION 4.3 – OSU Engagement		Yes	No	N/A
1	Are any of the following true? • OSU is the only institution participating in this study • OSU is the primary awardee on the funding • OSU employees or students are obtaining consent from participants • OSU employees or students will have access to individually identifiable data or samples	☒	☐	
	If “no” to all four, OSU is not engaged in this research.			
2	If you think the project does NOT engage OSU in research, please provide the details of OSU’s role in this project:			☒
				
	If “no” to research, human subjects, **OR** engagement (4.1-4.3 above), proceed by completing **ONLY THE FUNDING SECTION**, then submit this form to the HRPP office for a formal determination regarding the requirement for IRB oversight. Please also attach any test			

	instruments, including survey or interview questions, if applicable.			
SECTION 5 - Extent of the review required by OSU		Yes	No	N/A
	FAQ If I am collaborating with researchers at external institutions, which IRB reviews my study?			
1	Are OSU-affiliated individuals the only people conducting study activities; including recruitment, obtaining consent, data collection, data analysis, data or sample sharing or storage?	☐ *Skip to section 7*	☒	
	If any of the study team members have an appointment at OSU as well as an external institution, check "no" here and identify the additional institution as the "external site".			
2	Name of the external site(s)			☐
	Lawrence Livermore National Laboratory (LLNL) and Pacific Northwest National Laboratory (PNNL)			
3	Name of non-OSU researcher(s)			
	Dr. Kenneth Turteltaub at LLNL will only receive coded, de-identified samples for analysis. Dr. Jordan Smith at PNNL will only receive coded, de-identified data for analysis.			
	If this is a project revision and you are adding or removing sites, confirm that the **track change** feature of MS Word is on before you make these changes.			
	While it need not be described for the IRB, researchers should have a plan for maintaining communication between research sites that includes a method for assuring all participating sites: • have the most current version of the protocol • are made aware of any adverse events and unanticipated problems involving risks to participants or others			
4	What are the procedures for transferring and storing data or samples between research sites?			☐
	A portion of the de-identified coded study samples will be forwarded to LLNL for analysis with no personal identifying information contained within the shipment, so there is no risk of loss of confidentiality with the mailing of samples to LLNL. Samples sent to LLNL will be processed at OSU before shipment and are no longer considered biohazardous materials after processing. The radioactivity level in collected blood and urine are well below what is considered a background level when we conduct routine laboratory swipe surveys (below the limit of detection by liquid scintillation counting) and thus, these samples are not considered radioactive material. OSU personnel processing the samples are not exposed to above background levels			

	of radioisotopes and/or carcinogen. This material does not pose a biohazard, carcinogen hazard, or radioactivity health risk to postal carriers or LLNL employees.			
	**Examples**: All data will be stored in, and accessed via, [insert name of approved cloud server]. All mobile computer systems will be encrypted with at least the 256-bit. All samples are coded and the linked list of identifiers will be stored on a separate local server at the external institution only.			
	Information on data security guidance			
SECTION 6 - OSU will be the RESPONSIBLE institution		Yes	No	N/A
1	Will OSU be asked to provide IRB review for non-OSU researchers?	☐	☒ *Skip to section 7*	
2	Will one or more of these researchers be affiliated with an institution that has an IRB?	☐	☐	
2a	If yes, provide any details you have about the status of IRB review at the external institution(s):			☐
				
3	Will one or more of these researchers *not* be affiliated with an institution that has an IRB?	☐	☐	
	Complete and attach an Individual Investigator Agreement form for *each* collaborator who is not affiliated with an institution that has an IRB.			
4	Will anyone from the other research sites perform the following activities:			
4a	Recruit participants	☐	☐	
4b	Obtain informed consent from participants	☐	☐	
4c	Collect data or samples	☐	☐	
4d	Receive individually identifiable data or samples for analysis	☐	☐	
SECTION 7 - OSU will be the RESPONSIBLE institution but Review External Documents		Yes	No	N/A
1	Will OSU be asked to approve this study based on review of documents that have already been approved by another IRB?	☐	☒ *Skip to section 8*	
	**Example**: The Washington State University (WSU) IRB has reviewed and approved this study but OSU will also review this study and issue a separate approval notice. In this case, the PI can submit copies of the			

	documents that have been approved by WSU and skip many sections of this form.			
2	If yes, are the OSU research activities described accurately and completely in that protocol? If no, please explain:	☐	☐	
				
	If OSU will be asked to base this review on documents already approved by another IRB, proceed by completing **ONLY THE CONFLICT OF INTEREST, FUNDING, AND ASSURANCE** sections of this form, then submit this document, and all relevant external documents, to the HRPP office for review.			
SECTION 8 - OSU will be the RELYING institution		Yes	No	N/A
1	Will OSU be asked to rely on an IRB at another institution for review of this study?	☐	☒ *Skip to section 9*	
	**Example**: The Washington State University (WSU) IRB has reviewed and approved this study and the OSU will cede oversight to them, but retain a mirror copy of the study file, including all documents approved by WSU.			
2	The external document(s) should describe the overall study. Please briefly describe just OSU's involvement in this study:			
				
3	Will anyone from OSU perform the following activities:			
3a	Recruit participants	☐	☐	
3b	Obtain informed consent from participants	☐	☐	
3c	Collect data or samples	☐	☐	
3d	Receive individually identifiable data or samples for analysis	☐	☐	
	If OSU will be asked to rely on IRB review at another institution, proceed by completing **ONLY THE CONFLICT OF INTEREST, FUNDING, AND ASSURANCE SECTIONS** of this form, then submit this document, the external IRB approval letter, and documents approved by that IRB to the HRPP office for review. OSU will contact the external institution(s) for review of this request.			
SECTION 9 - Regulatory Flexibility		Yes	No	N/A
	The requirement to comply with some regulations and policies can be waived for eligible studies. To assist			

	the HRPP in determining whether this study is eligible for a flexible application of the regulations, answer the questions in this section. If "yes" to any item in this section, you may stop and skip to the next section.			
1	Does the study involve more than minimal risk to participants?	☒	☐	
	Risk is minimal when the probability and magnitude of harm or discomfort anticipated in the proposed research are not greater, in and of themselves, than those ordinarily encountered in daily life or during the performance of routine physical or psychological examinations or tests.			
2	Is there federal funding or a plan for future federal sponsorship for this study?	☒	☐	
	Research funded or otherwise regulated by a federal agency that has signed on to the Common Rule, including all agencies within the Department of Health and Human Services. Included are proof of concept studies for federal RFPs, pilot studies intended to support a federal grant application, training and program project grants, no-cost extensions.			
3 	Are there contractual obligations or restrictions triggered by a non-federal award that require the application of the federal regulations or which require that annual review be conducted by an IRB?	☒	☐	
4	Does the study involve federally classified research procedures and/or results that are legally knowable only by individuals with US government security clearance?	☐	☒	
5	Is there an NIH-issued or pending Certificate of Confidentiality?	☒	☐	
	Certificates of Confidentiality protect the privacy of research subjects by prohibiting disclosure of identifiable research information to anyone not connected to the research except when the subject consents or in a few other specific situations. NIH-funded researchers are automatically issued a Certificate through their award if the information collected could be individually identifiable.			
6	Does the study involve any FDA-regulated components?	☒	☐	
7	Does the study include any clinical interventions?	☒	☐	
	For the purposes of this policy, **clinical intervention** is defined as one that is intended to change or assess a health-related processes and/or endpoint. Examples include the use of drugs, dietary supplements, devices, blood draws, imaging (e.g., DXA, x-ray), delivery systems (e.g., telemedicine, face-to-face), diet, cognitive therapy, exercise, and any intervention that includes treatment, prevention, or diagnostic strategies.			
8	Does this study need to be registered with ClinicalTrials.gov?	☒	☐	
9	Will any of the participants be prisoners or parolees? This refers to the target population, not incidental enrollment.	☐	☒	

	If "no" to all of the above, the study may be eligible for **flexibility** in the application of the regulations. When applicable, subsequent sections will contain special instructions related to these studies.			
	Information on Regulatory flexibility			
SECTION 10 - Review Category		Yes	No	N/A
	If the study involves more than minimal risk to participants, it cannot be reviewed as exempt or expedited, and is not eligible for regulatory flexibility. Skip to section 11. If "no" to all of the questions in Section 9, the study may be eligible for regulatory **flexibility.** For these studies, the exempt and expedited categories that follow will serve as *examples* of research that may be deemed minimal risk. However, an actual category will not be assigned and you may opt to skip to section 11. The Human Research Protection Program staff will make the final determination of review level based on information that you provide throughout the form. However, the selection that you make here will assist both you and the HRPP in making an initial determination of the minimum level of review that the proposed project requires.			
SECTION 10.1 - Exempt Categories		Yes	No	N/A
	Limitations on exemptions: ➢ All of the exemptions can be applied to research involving pregnant women, human fetuses, and neonates ➢ None of the exempt categories can be applied to research involving prisoners as subjects *unless* the research is aimed at a broader subject population and only incidentally includes prisoners ➢ Exempt categories 1, 4, 5, 6, can be applied to research involving children ➢ Exempt category 1 may not be applied to research involving deception ➢ Exempt category 2 may not be applied to research involving children, with one exception: Exempt category 2 can only be applied to research involving educational tests or the observation of public behavior when the investigator(s) do not participate in the activities being observed ➢ Exempt category 6 may not be applied to research involving the ingestion of alcohol.			
	Children are defined as persons who have not attained the legal age for consent under the applicable law of the jurisdiction in which the research will be conducted. In Oregon, legal age for consent is 18, but that is not the case for all states or countries.			
1	Do ALL of the study activities fall within one or more of the federally defined exempt categories?	☐ *Skip to*	☒ *Skip to*	

APPLICATION & PROTOCOL

	Check all that apply.	*section 11*	*section 10.2*	
1a	**Category 1:** Research on educational practices, instructional techniques, and curricula.	☐		
	Regulation: Research conducted in established or commonly accepted educational settings, involving normal educational practices, such as (i) research on regular and special education instructional strategies, or (ii) research on the effectiveness of or the comparison among instructional techniques, curricula, or classroom management methods.			
1b	**Category 2:** Anonymous or non-sensitive research using educational tests, surveys, questionnaires, interviews, focus groups, or observation of public behavior.	☐		
	Regulation: Research involving the use of educational tests (cognitive, diagnostic, aptitude, achievement), survey procedures, interview procedures or observation of public behavior, unless: (i) information obtained is recorded in such a manner that human subjects can be identified, directly or through identifiers linked to the subjects; and (ii) any disclosure of the human subjects' responses outside the research could reasonably place the subjects at risk of criminal or civil liability or be damaging to the subjects' financial standing, employability, or reputation.			
1c	**Category 3:** Research using educational tests, surveys, questionnaires, interviews, focus groups, or observation of public behavior, not exempt under category 2 above.	☐		
	Regulation: Research involving the use of educational tests (cognitive, diagnostic, aptitude, achievement), survey procedures, interview procedures, or observation of public behavior that is not exempt under [category 2 above], if: (i) the human subjects are elected or appointed public officials or candidates for public office; or (ii) federal statute(s) require(s) without exception that the confidentiality of the personally identifiable information will be maintained throughout the research and thereafter.			
1d	**Category 4:** Secondary analysis of publicly available or de-identified information.	☐		
	Regulation: Research involving the collection or study of existing data, documents, records, pathological specimens, or diagnostic specimens, if these sources are publicly available or if the information is recorded by the investigator in such a manner that subjects cannot be identified, directly or through identifiers linked to the subjects.			
1e	**Category 5:** Federally supported research designed to examine public benefit or service programs.	☐		
	Regulation: Research and demonstration projects which are conducted by or subject to the approval of department or agency heads, and which are designed to study, evaluate, or otherwise examine: (i) public benefit or service programs;			

	(ii) procedures for obtaining benefits or services under those programs; (iii) possible changes in or alternatives to those programs or procedures; or (iv) possible changes in methods or levels of payment for benefits or services under those programs.			
1f	**Category 6:** Taste and food quality evaluation and consumer acceptance studies.	☐		
	Regulation: Taste and food quality evaluation and consumer acceptance studies: (i) If wholesome foods without additives are consumed, or (ii) If a food is consumed that contains a food ingredient at or below the level and for a use found to be safe, or agricultural chemical or environmental contaminant at or below the level found to be safe, by the Food and Drug Administration or approved by the Environmental Protection Agency or the Food Safety and Inspection Service of the U.S. Department of Agriculture.			
SECTION 10.2 - Expedited Categories		Yes	No	N/A
1	If any of the activities do not fit into the exempt categories, do ALL of the study activities fall within the federally defined categories for expedited review? Check all that apply.	☐	☒ *Skip to section 11*	☐
1a	**Category 1:** Research involving drugs or devices that do not require an FDA-issued IND or IDE.	☐		
	Regulation: Clinical studies of drugs and medical devices only when condition (a) or (b) is met. (a) Research on drugs for which an investigational new drug application (21 CFR Part 312) is not required. (b) Research on medical devices for which (i) an investigational device exemption application (21 CFR Part 812) is not required; or (ii) the medical device is cleared/approved for marketing and the medical device is being used in accordance with its cleared/approved labeling. Research on marketed drugs that significantly increases the risks or decreases the acceptability of the risks associated with the use of the product is not eligible for expedited review.			
1b	**Category 2:** Low volume blood draws.	☐		
	Regulation: Collection of blood samples by finger stick, heel stick, ear stick, or venipuncture as follows: (a) from healthy, nonpregnant adults who weigh at least 110 pounds. For these subjects, the			

	amounts drawn may not exceed 550 ml in an 8 week period and collection may not occur more frequently than 2 times per week; or (b) from other adults and children, considering the age, weight, and health of the subjects, the collection procedure, the amount of blood to be collected, and the frequency with which it will be collected. For these subjects, the amount drawn may not exceed the lesser of 50 ml or 3 ml per kg in an 8 week period and collection may not occur more frequently than 2 times per week.			
1c	**Category 3:** Non-invasive biospecimen collection (e.g., buccal swabs).	☐		
	Regulation: Prospective collection of biological specimens for research purposes by noninvasive means.			
1d	**Category 4:** Non-invasive data collection using routine clinical procedures (e.g., physical sensors applied to skin, moderate exercise).	☐		
	Regulation: Collection of data through noninvasive procedures (not involving general anesthesia or sedation) routinely employed in clinical practice, excluding procedures involving x-rays or microwaves. Where medical devices are employed, they must be cleared/approved for marketing. (Studies intended to evaluate the safety and effectiveness of the medical device are not generally eligible for expedited review, including studies of cleared medical devices for new indications.)			
1e	**Category 5:** Research on data or specimens that that have or will be collected for non-research purposes.	☐		
	Regulation: Research involving materials (data, documents, records, or specimens) that have been collected, or will be collected, solely for nonresearch purposes (such as medical treatment or diagnosis, student or employment records).			
1f	**Category 6:** Collection of data from voice, video, digital, or image recordings made for research purposes.	☐		
1g	**Category 7:** Collection of data using surveys, interviews, or focus groups.	☐		
	Regulation: Research on individual or group characteristics or behavior (including, but not limited to, research on perception, cognition, motivation, identity, language, communication, cultural beliefs or practices, and social behavior) or research employing survey, interview, oral history, focus group, program evaluation, human factors evaluation, or quality assurance methodologies.			
1h	**Category 8:** Renewal of research previously approved by the full board that has not yet enrolled subjects; is now limited to data analysis; or is closed to enrollment, subjects have completed the interventions, and	☐		

	activities are limited to long-term follow up of subjects.			
	Regulation: Continuing review of research previously approved by the convened IRB as follows: (a) the research is permanently closed to the enrollment of new subjects; all subjects have completed all research-related interventions; AND the research remains active only for long-term follow-up of subjects; OR (b) where no subjects have been enrolled and no additional risks have been identified; OR (c) where the remaining research activities are limited to data analysis			
1i	**Category 9:** Renewal of research not involving an FDA-issued IND or IDE that is re-classified at a full board meeting because it involves no more than minimal risk and no new risks were identified at the meeting.	☐		
	Regulation: Continuing review of research, not conducted under an investigational new drug application or investigational device exemption where categories two (2) through eight (8) do not apply but the IRB has determined and documented at a convened meeting that the research involves no greater than minimal risk and no additional risks have been identified.			
SECTION 11 - Conflicts of Interest and Competing Relationships		Yes	No	N/A
1	Does a researcher or family member have a financial or other business interest in an entity that is supplying funding, materials, products, equipment, research participants, or the site of data collection for the current research project?	☐	☒	
	**Family member** is defined as anyone having a relationship to a person as a spouse or domestic partner; the parent, child, or sibling of the individual or domestic partner; or any person for whom the individual has a legal support obligation **Examples** of conflicting or competing relationships: • A researcher or family member participates in research on a technology, process or product owned by a business in which the faculty member holds a financial interest. • A researcher participates in research on a technology, process, or product developed by that researcher. • A researcher or family member has a financial or other business interest in an entity that is supplying funding, materials, products, equipment, research participants, or the site of data collection for the current research project. • A researcher or family member is employed by or otherwise affiliated with the organization under study. • A researcher has an existing relationship with potential research participants recruited for this project. • A researcher or family member serves on the Board of Directors of a business that is supplying funding, materials, products, equipment, research participants, or the site of data collection for the current			

	research project. • A researcher receives consulting income from an entity that is funding the current research project.			
2	If yes, please provide details:			☐
				
	Conflicts of interest and competing relationships must be disclosed to research participants as part of the consent process.			
SECTION 12 - Sources of Funding and Support for this project		Yes	No	N/A
	Source(s) of support (check all that apply)			
1	Is funding for the project pending/awarded?	☒	☐ *Skip to section 13*	
2	Is there internal funding? If yes, indicate sources of funding:	☐	☒	
				
3	Is there external funding? Check all that apply	☐	☐	
3a	NSF	☐		
3b	NIH	☒		
3c	USDA	☐		
3d	DoD	☐		
3e	Other federal agency:	☐		
				
3f	OSU Foundation	☐		
3g	OSU Agricultural Research Foundation	☐		
3h	Other non-federal source:	☐		

4	Details of external funding. This information enables OSRAA to match the HRPP/IRB notices to the information in Cayuse.			☐
4a	Cayuse number:			
	17-1040; 17-1713			
	If externally funded and no Cayuse number is provided, the grant or contract must be attached for congruency review.			
4b	Grant or contract number:			
	R01 ES028600			
4c	Name of PI on grant or contract:			
	David E. Williams, PhD			
4d	Grant or contract title:			
	Benzo[*a*]pyrene Micro-dosing of Humans: A New Tool for Exposure, Risk Assessment and Prevention (see IRB Protocol 8233 for this document)			
5	Describe any substantive discrepancies or conflicting information between grant or contract and this protocol:			☐
	This protocol only pertains to Aim 3 of the grant.			
6	Is an external (non-OSU) organization or company providing material, equipment, drugs, supplements, or devices for this study? If yes, describe:	☐	☒	
SECTION 13 - Study Overview		Yes	No	N/A
1	List the study aims or research questions:			
	Benzo[a]pyrene (BaP) is the most intensely studied of the polycyclic aromatic hydrocarbons (PAHs), environmental pollutants formed naturally from forest fires, volcanoes, etc. Anthropogenic sources include coal, tobacco smoke, creosotes, coal tar-based pavement sealants, petroleum products including diesel and gasoline, wood and mixtures from production of coke, aluminum and graphite, among others[1]. BaP is a class 1, known human carcinogen (Internal Agency for Research on Cancer)[2] currently 8th on the ATSDR[3]			

(Agency for Toxic Substances and Disease Registry, a division of the Centers for Disease Control and Prevention) list of agents of concern at high-priority pollutant sites. BaP is strongly associated with lung cancer (number one cause of cancer mortality). Previously, EPA IRIS estimated the oral exposure slope factor for lifetime cancer risk at 7.3 mg/(kg-day) (Linear Extrapolation Model, no threshold) but a recent review of BaP resulted in a lower of that value to 1 mg/(kg-day)[4]. This risk assessment is based on high dose rodent studies. Few studies have been done examining PAHs in human plasma and none following administration of defined doses.

Our hypothesis is that pre-administration of Brussels sprouts or 3,3'-diindolylmethane (DIM) will alter [^{14}C]-BaP metabolism and increase the rate of elimination consistent with predictions based on a previously developed Physiologically-Based Pharmacokinetic (PBPK) model for BaP[5]. Briefly, this hypothesis will be tested by dosing individuals with 50 ng [^{14}C]-BaP alone and, following a 3-week washout period, ingestion of ½ cup (about 44 g) Brussels sprouts or 300 mg of 3,3'-diindolylmethane (DIM) supplement for 1 week prior to the [^{14}C]-BaP micro-dose. The impact of the supplement and the whole food will be assessed with respect to alterations in uptake from the GI tract, metabolism and rate of elimination. To date, our accelerator mass spectrometry (AMS) studies with PAHs have not investigated the impact of diet on [^{14}C]-BaP pharmacokinetics[6,7]. The consumption of cruciferous vegetables will be assessed at the beginning of the study by completion of a dietary questionnaire[8] to examine typical eating patterns in the previous 3 months and by collection and extraction of blood and urine to assay for DIM by LC/ESI-MS/MS-SRM)[9]. In addition, for each phase, urine will be assayed for DIM as an estimate of crucifer or DIM supplement intake[9]. [^{14}C]-BaP and metabolites will be extracted from plasma and urine (with and without β-glucuronidase/sulfatase) and shipped to Lawrence Livermore National Laboratory (LLNL) for UPLC-AMS analysis. The PK parameters will be assessed at PNNL and the impact of Brussels sprout or DIM consumption on uptake, metabolism and elimination of [^{14}C]-BaP determined. In preclinical and clinical studies administration of Brussels sprouts or DIM impacts the activity of the same enzymes responsible for the phase 1 (CYP1A1 and CYP1B1) and phase 2 enzymes (GSTM1, UGT, SULT)[10]. Monitoring changes in β-estradiol metabolites[11] will confirm the mechanism of alteration in the metabolic profile of [^{14}C]-BaP.

The results from this study will be disseminated in peer-reviewed toxicology/environmental health journals, at scientific meetings and provided to the U.S. EPA for incorporation into their risk assessment models. The experimental procedure employs accelerator mass spectrometry (AMS) for analysis. AMS (10^3-10^9 more sensitive for ^{14}C than scintillation counting) is increasingly used to determine pharmacokinetics of drugs under development. A dose given to human volunteers of no more than 1/100 the expected therapeutic dose or less than 100 µg of [^{14}C]-labeled drug (usually 100-200 nCi) is referred to

	as “microdosing” (Phase 0 studies or eIND studies). This approach has been validated for exploratory clinical development by the Consortium for Resourcing and Evaluating AMS Microdosing (CREAM Trial) and the EU Microdosing AMS Partnership Programme (EUMAPP). This early use of human subjects in drug development is consistent with the goals of the FDA Critical Path Initiative and AMS in exploratory IND applications has been addressed by FDA[12]. AMS has been used in toxicology and carcinogenesis studies. AMS sensitivity allows the study of pharmacokinetics and DNA binding of environmental contaminants known to be animal carcinogens and suspected of being human carcinogens including the cooked meat mutagens PhIP and MeIQx, the mycotoxin and human hepatocarcinogen aflatoxin B_1 (AFB_1), in addition to BaP[13]. It is noteworthy that our group has successfully completed AMS studies with AFB_1 and the PAH, dibenzo[*def,p*]chrysene (DBC)[6,7,14]. IARC estimates that a non-smoker, not exposed occupationally, will receive a daily dose of 270-700 ng of BaP; about 95% through the diet[2]. The European Union maximum limit for BaP in fish is 2,000 ng/Kg f.w. and the FDA Limit of Concern (LOC) is 35,000 ng/Kg. The dose of 50 ng would be the equivalent of eating 25 or 1.4 g of fish at the EU, and FDA limits, respectively. The World Health Organization has set an estimated safe daily lifetime (70 years for a 70 Kg individual, cancer endpoint) exposure to BaP of 42-350 ng[15]. With respect to the internal dose of [^{14}C] of 5.4 nCi, this represents 0.6% the dose given in a common diagnostic procedure (^{14}C-urea test for Helicobacter)[16] and 4 to 5 orders of magnitude lower than a recently published paper dosing people with 300 µCi of epicatechin[17]. Therefore, from the standpoint of both chemical and radioisotope dose to the volunteers, this protocol represents de minimus risk and that was the finding of the FDA (IND 117175) in a “study may proceed” determination for our current study (IRB protocol 5644) which uses a dose of 46 ng (5 nCi).			
	Provide survey questions, questionnaires, interview and focus group guides, references/citations, etc., as separate attachments.			
2	Provide details of where data will be collected. Examples: Online, OSU campus, K-12 classrooms, U.S. parks, senior living communities in Denmark.			
	All study visits to collect samples and data from subjects will take place in the Clinical Research Center, LPSC 407, in the Linus Pauling Institute.			
3	Provide background justification:			
	An estimated 95% of daily BaP exposure (270-700 ng, non-occupational; non-tobacco) is dietary[1]. PAHs, including BaP, are especially high in charcoal-broiled or smoked meats and cheeses and almost all foods contain appreciable amounts. Exposure to BaP, is associated with cancer of the lung, skin, stomach, ovary and testis in addition to non-cancer chronic disease such as asthma, cardiovascular disease and diabetes[2]. The fetus and infant are especially susceptible to PAH exposure. Even though PAHs are ubiquitous environmental pollutants of potential concern to human health, there is little or no information on the			

pharmacokinetics of PAHs in humans and certainly no information on the impact of diet. With the utilization of AMS located at LLNL, interfaced with Ultra-High Pressure Liquid Chromatography (UPLC), it is possible to measure [^{14}C]-isotopically-labeled BaP and metabolites over time in human plasma and urine following administration of micro-doses.

The oral exposure slope factor for cancer risk (70 years, 70 Kg adult) for BaP using a linear, non-threshold model (EPA-IRIS) was, until recently[4,18] is 7.3 mg/kg-day^{-1} but a recent review by EPA lowered that number to 1 mg/(kg-day). Cal/EPA uses a safety factor for age sensitivity (2.9 mg/kg-day^{-1}). This risk assessment is based on high dose (1-100 mg/kg-day) exposures in rodents- 5-6 orders of magnitude higher doses than human exposures. This risk assessment may not adequately protect some populations. For comparison, our preliminary results show the plasma levels, as determined by AMS at C_{max} (highest concentration obtained) after dosing individuals with 46 ng [^{14}C]-BaP, were ~ 8 fM (femto(10^{-15}) moles/L), or 10^7 lower than an *in vitro* study (50 nM) showing no adverse impact on hepatocytes. The sensitivity of AMS allows for micro-dosing with [^{14}C]-BaP in humans with *de minimus* risk to determine [^{14}C]-BaP and individual metabolites in the low fM range (2-20 femtograms/mL plasma). Our premise is- the best model for humans is humans. Utilizing PBPK models, we can provide a much more accurate estimate of BaP levels in human tissues following exposure at environmental levels, when dosed alone or subsequent to a week of consuming Brussels sprouts or DIM, as well as individual metabolites.

Cruciferous vegetables (Brussels sprouts, broccoli, cauliflower, turnip, kohlrabi, kale, water cress, radish and bok choy), along with the phytochemical 3,3'-diindolylmethane (DIM) derived from cruciferous vegetables, have been shown to alter the metabolism of high dose environmental chemicals in preclinical animal models[10,19]. There are multiple clinical trials ongoing or completed involving cruciferous vegetables, DIM or its precursor, indole-3-carbinol (I3C) (www.clinicaltrials.gov). The mechanism of action of DIM in alteration of BaP metabolism could occur by inhibition of CYPs involved in bioactivation, induction of CYPs and/or phase 2 enzymes (UGT, NQO, SULT, etc.) involved in detoxication (traditionally known as blocking mechanisms)[10,19]. Other than epidemiology studies, there have been no assessments of the potency and efficacy of DIM or Brussels sprouts impacting the pharmacokinetics (uptake, metabolism, elimination) of BaP or any environmental toxicant at truly environmentally relevant levels. It is important to note that this study is not testing any health benefits or claims of DIM or Brussels sprouts as we are not looking at any health endpoints in this study. The dose of BaP is almost a million fold lower than the dose producing cancer in animals and is in the low range of a safe daily lifetime exposure of 46-350 ng[15] established by the World Health Organization.

Background justification should support the objectives of the research as well as the knowledge that is anticipated from the research results. Explain the need for the study and what gap in knowledge the

	results are expected to fill. Summarize relevant existing data, literature, past and ongoing studies, and how your study ties in with these. Provide specific methods and procedures in a later section.			
4	Is the study student-driven (for the purpose of a thesis, dissertation, or other)?	☐	☒	
SECTION 14 - Target Enrollment		Yes	No	N/A
1	What is the target enrollment number?			☐
	Up to 50 people will be enrolled in this study. Our goal is for 7 subjects to complete the study.			
	A target enrollment number is not applicable for studies limited to chart review or review of large, pre-existing datasets, such as those originating from public health surveillance organizations.			
	If the study is determined to be exempt, expedited, or eligible for **flexibility** in the application of the regulations, an approximate number is sufficient for the evaluation of risk. The PI will not be required to report enrollment numbers to the IRB over the course of the study. However, if the study involves more than minimal risk or is FDA-regulated, an exact number is required and cannot be exceeded without prior approval.			
	FAQ What is the “total target” enrollment number?			
2	Provide scientific justification for the target enrollment number:			☐
	We expect a high percentage of screen failure. If a large number of study enrollees qualify to participate in the study, we will stop enrolling subjects prior to reaching our maximum enrollment figure.			
SECTION 15 - Participant Demographics		Yes	No	N/A
1	Age ranges	*Check all that apply*		
1a	0-7	☐		
1b	8-17	☐		
1c	18-89	☒		
1d	90+	☐		

APPLICATION & PROTOCOL

	If the study is intended to be limited to adults, all enrolled participants must have attained the legal age to consent to research under the applicable law of the jurisdiction in which the research will be conducted. Not all states or countries consider 18 years to be the age of majority; in Oregon it is 18. Describe nuances related to participant age in the inclusion and exclusion criteria sections below. **Sample recruitment or consent language**: "In order to be in this study you must be of legal age to consent, which is 18 in most states."			
2	Indicate all populations permitted to enroll who may need additional safeguards or for whom additional regulations may apply:			
2a	Children	☐	☒	
	Guidance on research with children			
2b	Adults lacking capacity to consent	☐	☒	
2c	Children in foster care or wards of the state	☐	☒	
	Regulations related to enrolling children in foster care or wards of the state			
2d	Pregnant women **AND** the study involves more than minimal risk **OR** a physical intervention	☐	☒	☐
2e	Fetuses and/or neonates	☐	☒	
2f	Prisoners	☐	☒	
	Guidance on research with prisoners. If in Oregon, submit an application to the Oregon Department of Corrections application. If outside of Oregon, contact the state DOC for instructions.			
2g	Economically or educationally disadvantaged persons	☒	☐	
2h	American Indians or Alaska Natives	☒	☐	
	Guidance for research involving Tribal populations			
2i	People in the European Union (regardless of citizenship)	☐	☒	
	Contact the Office of Information Security (IS) if there is a plan to collect data from people who are in the EU. IS will determine the applicability of the EU's General Data Protection Regulation and ensure compliance with this regulation when necessary.			
3	Will any of the following OSU-affiliated groups be permitted to enroll:			

3a	Students currently enrolled in a class or lab instructed by a study team member	☐	☒	
3b	Employees who report to or are otherwise supervised by a study team member	☐	☒	
3c	Any of the study team members	☒	☐	
	Guidance on Students and Employees as Research Subjects and self-experimentation.			
3d	If "yes" to 3a, 3b, or 3c, provide scientific justification for permitting these individuals to enroll and a plan for mitigating the potential for actual or perceived coercion:			☐
	The PI is the only team member permitted to enroll in this study. He will be subject to all eligibility screening and informed consent procedures described above and his participation will strictly adhere to the protocol.			
4	Will people who do not speak or read English be permitted to enroll?	☐	☒	
4a	If not, provide justification and information regarding the impact of the exclusion on the generalizability of the data:			☐
	We do not have the resources to translate documents into other languages or to provide translators over the course of the study. Given the nature of this study and the risks involved, it would not be appropriate to rely on limited English proficiency or informal translation by a friend or family member, nor would we be able to adequately assess comprehension of people with limited English proficiency. This study is not powered to create generalizations with a maximum of 7 subjects will complete this study. Therefore, these exclusions will not impact the generalizability of the data.			
5	Are people of any sex, gender/gender identity eligible to participate?	☒	☐	
5a	If not, who will be included/excluded and why?			☒
				
6	Are people of any race or ethnicity eligible to participate?	☒	☐	
6a	If not, who will be included/excluded and why?			☒
				
7	List any inclusion criteria not addressed above and explain why this is a scientifically appropriate population for the study:			☐
	• Willing to defer blood donation for one month before, throughout, and one month after completion of			

	study activities • Willing to avoid consuming any non-study cruciferous vegetables, I3C or DIM supplements, smoked or cured meat or cheeses, or charcoal-grilled meats for 2 weeks prior to and during each study cycle. (gas grilled foods acceptable) • Health history review and physical assessment showing general good health, as determined by study physician. Acceptable physical exam may have been conducted as part of protocol 8233 or 8554 if subject has not had significant changes in health status.			
8	List any exclusion criteria not addressed above and explain why this is a scientifically appropriate population for the study:			☐
	• Adults aged 66 years or older –may have altered gut absorption and metabolism that differs from the 21-65 year old population • Persons aged 18-20 years old – Prior to January 25, 2016, NIH defined adults as age 21 years and older. Our previous studies with BaP and DBC have used study populations aged 21-65, so we will continue to use this age range for scientific comparison • Women who are not post-menopausal or have not had surgical sterilization - to eliminate any possibility for fetal exposure • Smoker (tobacco or other substances) in past 3 months or living with smoker – excessive PAH exposure • Use of smokeless tobacco in past 3 months- excessive PAH exposure • Regular use of medications that affect gut motility or nutrient absorption (e.g. cholestyramine, sucralfate, orlistat, pro- or anti-motility agents) – altered gut absorption and metabolism • History of gastrointestinal surgery (e.g. bariatric surgery, cholecystectomy) or gastrointestinal disorder (e.g. Crohn's disease, celiac disease, IBS, or colitis) – altered gut absorption and metabolism • Current or history of kidney or liver disease – altered PAH metabolism • Prior high-dose ^{14}C exposure from medical tests. (micro-dose ^{14}C exposure not exclusionary) – excessive ^{14}C exposure and potentially high background levels that would interfere with AMS detection of ^{14}C-BaP and metabolites • Occupational PAH exposure (e.g. roofers, asphalt pavers, fire-fighters, etc.) – excessive PAH exposure • Regular use of indole-3-carbinol or DIM dietary supplements - altered PAH metabolism • Allergy or intolerance to Brussels sprouts or similar foods			

SECTION 16 - Identification and Recruitment of Participants		Yes	No	N/A
1	How will potential participants be identified and recruited?			
	We will recruit subjects through postings in *OSU Today*, Craigslist, local regional and local media, and flyer advertisements placed on the OSU campus and throughout the Corvallis area. We also anticipate recruiting from the LIFE registry maintained by the Center for Healthy Aging Research (CHAR) at OSU. With the exception of contacting individuals on the LIFE registry, only subjects requesting information about the study will be contacted, and no subjects will be prospectively contacted by researchers without contact first being initiated by the individual. In situations that potential subjects respond to an abbreviated version of the full text recruitment posting (e.g. OSU Today's 75-word limit for postings), the nurse coordinator can provide the full text of the recruiting flier via phone or email if more information is requested. Prior to study enrollment, the coordinator may also respond to general inquiries about study logistics (location, number of visits, study activities, the basic study schedule and type of specimen collected).			
	Information on Recruitment of Research Participants			
	Letters of support are generally required for studies that involve vulnerable populations. However, the IRB may request that you provide letters of support or permission under additional circumstances to ensure that appropriate safeguards are in place and/or that the study is feasible. Examples of supporting documents include school district permission forms or letters from local organizations attesting to feasibility or cultural appropriateness of international studies.			
	Attach advertisement or other recruitment material (including content of electronic posts or email).			
2	Confirm that the recruitment materials include the following information:			
2a	Study title	☒	☐	
2b	Name of the Principal Investigator	☒	☐	
2c	A clear statement that this is research	☒	☐	
2d	Contact information for study personnel	☒	☐	
3	If no to any of the above, provide justification for the omission of the item(s):			☐

				
SECTION 17 - Informed Consent		Yes	No	N/A
	*"Respect for persons requires that subjects, to the degree that they are capable, be given the opportunity to choose what shall or shall not happen to them. This opportunity is provided when adequate standards for informed consent are satisfied." Belmont Report, 1979*			
	Attach all consent forms, verbal consent guides, research information sheets, or other documents used for obtaining consent or notifying people of study participation.			
	Information on Resources for the Consent and Assent Process			
SECTION 17.1 - Obtaining Consent		Yes	No	N/A
	Information on Elements of informed consent			
1	Will consent be obtained from all participants?	☒	☐	
2	Are you seeking a waiver of the requirement of one or more elements of consent?	☐	☒	
3	If yes, which of the criteria below does this study meet? Check all that apply.			☒
3a	The research involves no more than minimal risk to the subjects	☐	☐	
3b	The waiver of consent or omission of one or more the elements of consent will not adversely affect the rights and welfare of the subjects	☐	☐	
3c	The research could not practicably be carried out without the waiver or alteration	☐	☐	
3d	Study is exempt or eligible for a **flexible** application of the regulations, and the waiver or alteration will not adversely affect the rights (such as FERPA) and welfare of the subjects.	☐	☐	
4	Whenever appropriate, will the subjects be provided with additional pertinent information after participation?	☐	☒	
5	Does the research involve deception?	☐	☒	
6	If yes, review the guidance on research involving deception and provide all required information, including justification, and describe any debriefing process:			☒
				
	Deception occurs as the result of investigators providing false or incomplete information to participants for the purpose of misleading research participants.			

SECTION 17.2 - Written Consent		Yes	No	N/A
	There is no IRB-related requirement for *signed* consent forms if the study is exempt or eligible for regulatory **flexibility**. However, other laws or regulations, such as FERPA, may require that a signature be obtained.			
1	Will participants be asked to ***sign*** written consent forms?	☒	☒	☐
2	If not, which of the criteria below does this study meet? Check all that apply.			☐
2a	The only record linking the subject and the research would be the informed consent form and the principal risk would be potential harm resulting from a breach of confidentiality. Each subject (or legally authorized individual) will be asked whether the subject wants documentation linking the subject with the research, and the subject's wishes will govern;	☐		
2b	The research presents no more than minimal risk of harm to subjects and involves no procedures for which written consent is normally required outside of the research context; or	☐		
2c	Study is exempt or eligible for a **flexible** application of the regulations.	☐		
2d	Written consent will be obtained for the primary research activities but not for eligibility screening.	☒		
3	Will participants be provided with a copy of their signed consent form or a written notification or explanation regarding the research?	☒	☐	☐
SECTION 17.3 - Consent Process		Yes	No	N/A
1	Indicate where and when consent will be obtained (e.g., in a location that protects the participants' privacy, prior to involvement in any study activities):			☐ *Skip to section 17.4*
	Consent will be obtained in the Clinical Research Center (LPSC 407). The nurse coordinator will verbally review the informed consent document at the screening visit prior to any study activities taking place. We will offer subjects an opportunity to voice any questions or concerns, to take time to consult others (family members, health care providers), or do any other research that would help them understand the study activities before they sign the consent form. Except for telephone screening, no study activities will take place before written consent is obtained. Discussions regarding consent with potential subjects and acquisition of consent will take place in a private location with measures taken to ensure privacy (closed doors, periods of time between appointments with other subjects).			

2	Will consent be obtained in a web-based environment?	☐	☒	
2a	Will there be a mechanism provided for participants to directly and privately communicate questions or concerns to a study team member?	☐	☐	☒
2b	If no, explain why:			☒
				
3	Explain how comprehension of consent information will be assessed and what questions will be asked of the participants to determine comprehension of the study information:			☐
	We will ask subjects to briefly, in their own words, describe the details of the study, required activities, and their understanding of the risks involved.			
	Agreement without understanding is not informed consent. Open-ended questions are one useful tool for assessing comprehension. **Examples**: What questions can I answer for you? To ensure that you understand what the study involves, would you please tell me what you think we are asking you to do? In your own words, can you tell me what the biggest risk to you might be if you enroll in this study?			
SECTION 17.4 - Parental Permission		Yes	No	N/A
1	If **children** may be enrolled in the research, provide a plan for obtaining consent from parents or legal guardians:			☒ *Skip to section 17.5*
				
2	Are you requesting that the requirement for parental permission be waived because (a) the research involves no more than minimal risk AND (b) the target population includes children in education settings (e.g., K-12 settings, 4-H programs, etc.)?	☐	☐	
3	Are you requesting that the requirement for parental permission be waived because (a) the research involves no more than minimal risk AND (b) the target population includes college or university students who are not yet 18 years of age?	☐	☐	
4	If you are asserting that the requirement for obtaining parental permission is not a reasonable requirement to protect children in this study, describe the alternative mechanism for safeguarding the rights and welfare of these participants (e.g., notification to parents with an opt-out period):			☐
				

5	If children the target population is children in foster care or wards of the state, describe the plan and process for addressing the potential for changes in guardianship over time and for ensuring that ongoing consent is in place from the current guardian:			☐
				
SECTION 17.5 - Non-English Speakers		Yes	No	N/A
1	If participants who **do not speak English** may be enrolled, describe the investigator's language proficiency in the participants' native language (conversational, fluent, do not speak the language) and provide information regarding the use of a translator, if one will be utilized:			☒ *Skip to section 17.6*
				
	Attach all documents that participants will see translated into the language they speak and understand.			
	Indicate whether you will use a translator and/or an interpreter and explain their qualifications. Consider issues of confidentiality related to using a translator or interpreter and describe instructions that they will receive with respect to any sensitive information. If the translator is not a native speaker of the language, is not a professional translator, or does not have a master's degree in languages, provide back translations of the documents into English.			
SECTION 17.6 - Education Records		Yes	No	N/A
	Directory information			
1	If the study involves the collection of individually identifiable **education records** (e.g., grades, assignments), beyond directory-level information, confirm that consent will be obtained in writing or in an authenticated environment (valid login credentials required). If the education records will be de-identified before they are accessed by the researchers, check "N/A" and skip to 17.7.	☐		☒ *Skip to section 17.7*
2	If the study involves the collection of **education records** (e.g., grades, assignments), beyond directory-level information, confirm that the following FERPA-required information appears in the consent form:			
2a	Data to be released (e.g., course grades, assignments, GPA, video-recordings of class activities, etc.)	☐	☐	
2b	To whom it will be released (e.g., researchers, funding agency, publications, etc.)	☐	☐	
2c	For what purpose the data is being released	☐	☐	
2d	A field for the student/participant to include the date consent was given	☐	☐	

SECTION 17.7 - Capacity to Consent		Yes	No	N/A
1	If adult participants with **diminished or fluctuating capacity to consent** will be enrolled, address the following areas:			☒ *Skip to section 18*
1a	Describe how capacity for consent will be determined if some or all participants lack capacity to consent:			
				
1b	If a participant's capacity to consent may decline during the study (e.g., beginning stages of dementia), explain the plan for ongoing assessment of the participant's ability to understand that they are participating in a study:			
				
1c	If a participant may regain capacity to consent after being enrolled in the study through a surrogate consent process, describe the plan for assessing capacity to consent and the consent process:			
				
1d	Describe the procedure for identifying a surrogate decision maker for a participant unable to consent:			
				
SECTION 18 - Assent		Yes	No	N/A
	The responses in this section should demonstrate an understanding of the target population and the nature of assent.			
	Check both "yes" *and* "no" for obtaining assent and requesting a waiver of assent when the plan is to obtain assent for at least one study activity but not others. Complete the entire section and provide clarifying details in the description of the process below.			
	Information on Assent and Research with Children			
1	Will children and/or people who cannot provide consent be enrolled (e.g., individuals lacking capacity to consent)?	☐	☒ *Skip to section 19*	
2	Provide a description of the assent process:			☐
				

3	Indicate who will discuss the study with the child and, if not the parent, describe their training in presenting research in a clear and age-appropriate fashion:			☐
				
4	If children enrolled in this study may reach the age of majority (18, in Oregon) before their study participation ends, describe the plan and process for obtaining their consent to continue:			☐
				
5	Will a written assent document or explanation of research be provided to children?	☐	☐	
	Written assent form or verbal assent guide unless requesting a waiver.			
6	Will a written signature be obtained from children prior to enrollment?	☐	☐	
7	Will the assent process be strictly verbal (no written document)?	☐	☐	☐
8	Are you seeking a waiver of the requirement to obtain assent?	☐	☐	
9	If yes, check the appropriate reasons below:			☐ *Skip to section 19*
9a	The ages, maturity, or psychological state of the children to be enrolled make them incapable of providing assent; or	☐	☐	
9b	The intervention or procedure involved in the research holds out a prospect of direct benefit that is important to the health or well-being of the children and is available only in the context of the research; or	☐	☐	
9c	The research involves no more than minimal risk to children, and the research could not practicably be carried out without the waiver of assent.	☐	☐	
SECTION 19 - Eligibility Screening		Yes	No	N/A
1	Describe the eligibility screening process, including whether it will take place before or after written informed consent has been obtained and what will be done with the data if the individual is ineligible to proceed (“screen fails”):			☐
	Participants will be screened by telephone. A *telephone screening guide* with an *eligibility checklist* is attached. An overview of study description and activities will be described to individuals during telephone screening, and then subjects will be asked eligibility screening questions. Participants who qualify by telephone screening will be scheduled for a screening visit for in-depth written consent and health screening. Subjects that are not eligible based on their responses to the telephone screening questions will			

be thanked for their interest and told that they do not qualify to participate in the study. Any personal information gathered will be shredded or deleted immediately.

We seek a waiver for the telephone screening portion of the recruitment to be able to collect screening information over the telephone prior to the screening visit to make the best use of participant and study team time. The *telephone screening guide* describes the process of obtaining verbal consent for the eligibility screening questions. If the subject is eligible for a screening visit, the telephone screening guide is filed in the subject's research record to document the phone screening consent along with the signed informed consent form that is subsequently obtained at the screening visit.

Screening visit (60 minutes)

The nurse coordinator will review the informed consent document including study activities, schedule and diet restrictions, answer questions, and provide further information as requested. After written consent is obtained, the nurse coordinator will collect demographic information, health history, height, weight, blood pressure and heart rate (see *Health Assessment Form* and *Demographic Form*). Female subjects will be asked to provide a spot urine sample for a pregnancy test. The study physician will perform a physical exam.

Evaluation of eligibility

The study physician will review Health Assessment information to assess eligibility to continue with the study. Subjects who qualify for this study will be notified of their eligibility and invited to participate. Subjects that do not qualify based on the screening visit health assessment will be notified of their ineligibility and if appropriate, referred to their primary care physician for follow-up. Their written consent, telephone screening document, screening visit data will be retained and protected for confidentiality.

Subjects who were previously eligible for IRB protocol 8233 (Benzo[a]pyrene Ultralow Dose-Response Study) or IRB protocol 8554 (Ultralow Dose PAH Binary Mixture Study) will be potentially eligible for this study without an additional physical exam. All other screening activities, eligibility criteria, and obtaining written consent apply (e.g. telephone screening, health assessment, etc.). Any changes in health or medication since the previous screening visit will be reviewed by the nurse coordinator, or if significant, by the study physician, to determine continued eligibility. If deemed necessary, subjects will undergo another physical exam. **A subject may not simultaneously participate in this study and 8233 or 8554.** The study team will ensure that any subjects who have completed protocol 8233 or 8544 will have at least 3 weeks'

	washout period between ^{14}C-BaP doses from either study.			
	Please attach screening guide, eligibility checklist, or similar document.			
SECTION 20 - Methods and Procedures		Yes	No	N/A
1	Provide a description of the methods and procedures to be followed during this research project:			
	All study activities will take place in the Clinical Research Center in the Linus Pauling Science Center, room 407. <u>Screening visit (60 minutes)</u> The nurse coordinator will review the informed consent document including study activities, schedule and diet restrictions, answer questions, and provide further information as requested. After written consent is obtained, the nurse coordinator will collect demographic information, health history, height, weight, blood pressure and heart rate (see Health Assessment Form and Demographic Form). Female subjects will be asked to provide a spot urine sample for a pregnancy test. The study physician will perform a physical exam. Fasting is not required for this visit. <u>Cruciferous vegetable intake survey</u> Subjects will be asked to complete the Arizona Cruciferous Vegetable Food Frequency Questionnaire[8] before they begin the first cycle. The time frame of the questionnaire will cover the previous 3 months prior to the first study cycle (excluding diet restriction period) to obtain baseline levels of cruciferous vegetable intake. <u>Diet restrictions</u> Subjects will be asked to avoid consuming any non-study cruciferous vegetables, I3C or DIM supplements, smoked or cured meat or cheeses, or charcoal-grilled meats for 2 weeks prior to and during each study cycle. Gas-grilled foods are allowed. Subjects will be provided a list of cruciferous vegetables and condiments to avoid. Subjects will be asked to confirm they have followed the dietary restrictions before study cycle activities begin. Subjects who indicate that they have not followed the diet restrictions for 2 weeks will be allowed to have up to two dietary lapses with subsequent washout extension before they will be automatically removed from the study due to lack of compliance. <u>Study cycles (3 total):</u> Subjects will be asked to complete a 3-day food diary covering the 3 days prior to each study cycle and			

another diary covering the 48 hours of each study cycle. Administration of this diary in our current study (IRB protocol 5644) showed that, on average, volunteers decreased their dietary BaP exposure by 35-54 ng/day[20] The results will be used to estimate dietary intake of PAHs using the latest data from the Joint FAO/WHO Expert Committee on Food Additives (JECFA, http://www.food.gov.uk/multimedia/pdfs/poly-aromatic-hydrocarbons.pdf, accessed 11-7-2016).

For all 3 cycles: Subjects will fast overnight (no food or drink besides water) before Day 1 of each cycle. Subjects will be asked to provide a spot urine sample for baseline analysis and then to empty their bladders completely. Female subjects' urine will also be used for a urine pregnancy test, and test results must be negative to proceed with study activities. The results of these pregnancy tests will not be revealed to the subject in compliance with the Oregon Health Authority, Laboratory Compliance Section.

Cycle 1: [^{14}C]-BaP capsule only

The IV catheter will be placed in an appropriate vein in the forearm or antecubital region by the nurse coordinator. Subjects will swallow a capsule containing the [^{14}C]-BaP with 100 mL water. Prior to micro-dosing with [^{14}C]-BaP, a time zero blood sample of 20 mL will be collected with 10 mL analyzed for background plasma levels of BaP (and 62 additional PAHs) with GC-MS/MS[19] and the remaining 10 mL analyzed for [^{14}C]-BaP and metabolites as with subsequent time points.

Blood will be sampled at 0, 0.25 0.5, 1.0, 1.5, 2, 3, 4, 8, 24, and 48 hours (11 draws total, 8-9 of which come from the catheter). The catheter will remain in place for blood draws through hour 4. After the 4 hour blood draw, subjects will have the choice of having the catheter removed and returning for the 8 hour blood draw by straight stick, or they can keep the catheter in place and remain on-site until after the 8 hour blood draw. The subjects will be monitored by the nurse coordinator between blood draws. The 8 (possibly), 24, and 48 hour blood draws will be done with straight stick phlebotomy. No more than three (3) skin punctures will be made in an attempt to draw blood at each visit. The first blood draw of this cycle will be 20 mL and then subsequent blood draws will be 10 mL.

Subjects will be instructed to collect all urine during the entire 48-hour study cycle in containers provided by the study team.

Two hours after swallowing the capsule, a standardized breakfast will be provided, after which subjects may resume normal eating and drinking. Snacks or fluids will be provided prior to the standardized breakfast if the study team determines it is necessary.

Cycles 2 and 3: [^{14}C]-BaP capsule with Brussels sprouts and [^{14}C]-BaP capsule with DIM

Visit 0 (20 minutes): Seven days before the capsule is taken, subjects will come to the CRC for a spot urine collection (analyzed for DIM an β-estradiol metabolites) and 25 ml blood sample (analyzed for background BaP and 62 other PAHs, DIM, β-estradiol and metabolites.) They will pick up their study Brussels sprouts or DIM capsules as described below:

> *Brussels sprouts:* Subjects will be provided 7 days' servings of frozen, pre-cooked Brussels sprouts. Each serving will be about 50 grams (about ½ cup) of frozen Brussels sprouts. Study team members will record serving weights before providing to subjects and serving weights will be standardized as much as possible. Subjects will be instructed to consume one serving with dinner (defrost, warm and season, if desired) during the 7 days prior to the study cycle.
>
> *DIM capsules:* Subjects will be provided 7 days' worth of BioResponse DIM® 150 capsules. They will be instructed to take 2 capsules (300 mg total) each evening with dinner during the 7 days prior to the study cycle.

The IV catheter will be placed in an appropriate vein in the forearm or antecubital region by the nurse coordinator. Prior to micro-dosing with [^{14}C]-BaP, a time zero blood sample of 25 mL will be collected with 10 mL analyzed for background plasma levels of BaP (and 62 additional PAHs) with GC-MS/MS[21] and the remaining 15 mL analyzed for DIM as well as β-estradiol and metabolites. Fifteen mL will be taken at the 48 hour time point to determine what changes may have occurred in blood and urine levels of DIM and estrogens. Subjects will swallow a capsule containing the [^{14}C]-BaP with 100 mL water. For the DIM study cycle, subjects will also take 2 capsules (300 mg BioResponse DIM®) with the [^{14}C]-BaP capsule.

Blood will be sampled at 0, 0.25 0.5, 1.0, 1.5, 2, 3, 4, 8, 24, and 48 hours (11 draws total, 8-9 of which come from the catheter). The catheter will remain in place for blood draws through hour 4. After the 4 hour blood draw, subjects will have the choice of having the catheter removed and returning for the 8 hour blood draw by straight stick, or they can keep the catheter in place and remain on-site until after the 8 hour blood draw. The subjects will be monitored by the nurse coordinator between blood draws. The 8 (possibly), 24, and 48 hour blood draws will be done with straight stick phlebotomy. No more than three (3) skin punctures will be made in an attempt to draw blood at each visit.

Subjects will be instructed to collect all urine during the entire 48-hour study cycle in containers provided by the study team.

Two hours after swallowing the capsule, a standardized breakfast will be provided, after which subjects may resume normal eating and drinking. Snacks or fluids will be provided prior to the standardized

breakfast if the study team determines it is necessary.

Summary of samples collected for Brussels Sprouts and DIM cycles

Time point	Total blood collected (mL)	Urine
-7 days	25	Spot
0 (background)	25	Spot
0.25 – 24 hours	90 (9 x 10 mL)	Total urine (0-48 hours)
48 hours	15	

The total amount of blood collected for the BaP-only cycle is 120 mL. The total amount of blood collected for the Brussels sprouts and DIM cycles is 155 mL per cycle.

Subjects who indicate that they have not consumed the Brussels sprouts or DIM capsules as instructed will be allowed to have up to two lapses with subsequent washout and dietary restriction extension before they will be automatically removed from study participation due to lack of compliance.

There will be at least three weeks' washout period between cycles. Previous multiple micro-dosing of the same individual has shown that after 3 weeks there is no detectable [^{14}C] in plasma or urine, validating the choice of 3 weeks as adequate for a "washout period"[6,7]. Extending the washout period will not adversely impact the data collected from the subjects but will minimize the subject's exposure to PAHs.

Subjects who have participated in IRB protocol 8233 (Benzo[a]pyrene Ultralow Dose-Response Study) or IRB protocol 8554 (Ultralow Dose PAH Binary Mixture Study) will participate in only the [^{14}C]-BaP plus DIM and [^{14}C]-BaP plus Brussels sprout cycles if they have complete the 50 ng [^{14}C]-BaP-only cycle within the prior 12 months. There is no set order to the Brussels sprouts or DIM cycles.

Subjects will be asked to defer blood donation for one month before, throughout, and for one month after completion of the study. At any point during the study, subjects will be referred to their physician if the study physician or nurse consider this advisable.

<u>Processing of samples:</u>

Blood samples will be centrifuged to obtain plasma and peripheral blood mononuclear cells (PBMCs). The [^{14}C]-BaP and metabolites will be extracted directly from plasma by liquid/liquid extraction with ethyl acetate. The urine will be pooled over the time periods pre-capsule (baseline), 0 – 12, 12-24, and 24-48

hours. After volumetric quantitation of each pool of urine, 12 mL will be collected, 4 mL will be adjusted to pH 5 and half treated with β-glucuronidase/sulfatase prior to extraction with ethyl acetate. The plasma and urine extracts will be blown down under argon, sealed in amber vials and shipped to LLNL where they will be injected onto an Ultra-Pressure Liquid Chromatograph (UPLC) interfaced via a "moving wire" into the AMS[22]. We will be able to determine femto (10^{-15}) g/mL levels of BaP and BaP metabolites (by co-elution with unlabeled standards available from the OSU Superfund Research Center PAH repository). The time-course of blood and urine levels of parent BaP and metabolites will be used to construct a pharmacokinetic model[5]. In addition, DNA isolated from PBMCs at 0 hr and 48 hr will be counted directly on the AMS to check for levels of covalent DNA binding.

DIM levels in blood and urine will be measured as follows: Triplicate samples from each time point or pooled urine samples will be spiked with [$^2H_2$]DIM internal standard and incubated 20 hours at 37°C with β-glucuronidase/arylsulfatase. Samples will then be extracted twice with t-butyl methyl ether, solvent evaporated with nitrogen stream, and DIM levels determined. We will utilize the recently developed LC/ESI-MS/MS-SRM assay[9] from Dr. Stephen Hecht's (a collaborator on the NIH grant funding this work) group as a biomarker of both DIM and Brussels sprouts consumption.

The levels of β-estradiol and metabolites (2-, 4- and 16α-hydroxylated, conjugated and total) is assessed by a UPLC-tandem mass spectrometry method[11]. Briefly, 2 mL of serum is divided into equal fractions with 1 treated with glucuronidase/arylsulfatase for deconjugation. A liquid-liquid extraction with MTBE is performed and the estrogens derivatized with pyridine-3-sulfonyl chloride, re-extracted with MTBE, evaporated to dryness under nitrogen and then taken up in 20% methanol in water prior to injection onto a UPLC column interfaced to a TSQ mass spectrometer by electro-spray ionization. The identity of each metabolite will be determined by UPLC retention time and SRM-MS[11]. Dr. Richard van Breemen, Director of the Linus Pauling Institute, has expertise with this assay[23] and is available for consultation if needed.

Summary of sample analyses for Brussels sprouts and DIM cycles

Time point	DIM blood	DIM urine	Estrogen blood	Estrogen urine	14C-BaP blood	14C-BaP urine	Background PAH blood
-7 days	x	x	x	x			x
0	x	x	x	x	x	x	x
0.25 – 24 hours	x	x			x	x pooled	
48 hours	x	X 24-48 hr pool	x	X 24-48 hr pool	x	x 24-48 hr pool	

	If the study involves accessing student education records, list all data to be used (e.g. course grades, assignments, GPA, video-recordings of class activities, etc.)			
	Identify any surveys or questionnaires that are being tested or validated instruments that have been modified for the purposes of this study.			
	Identify any novel or modified experimental activities that are being tested the purposes of this study.			
	Specific information related to the use of drugs, devices, biologics, food, biospecimens, and radiation will be requested later in this document.			
2	If any of the activities would be conducted regardless of the research, briefly describe those activities here:			☒
	Example: Grant is funding the expansion of an existing training program. Research will be conducted to compare outcomes between participants from the original program and those participating in the expanded program. In this scenario, the program would be administered regardless of the research question and should be briefly described in this section.			
3	Describe the qualifications that study team members possess to safely and appropriately conduct the study activities:			
	Williams, PhD in Biochemistry, is a Toxicologist with over 40 years of experience working with Polycyclic Aromatic Hydrocarbons (PAHs) and has directed similar human studies involving micro-dosing of PAHs at OSU. Uesugi, RN, the Clinical Research Nurse Coordinator (nurse coordinator) has extensive experience in			

	clinical research coordination, laboratory research, science communication for the lay public, and project management. In addition to being a registered nurse, Ms. Uesugi has completed a certified phlebotomy training course, intravenous therapy training, and a clinical research coordinator (CRC) training course. Siddens has performed carcinogen and radioisotope handling over the course of 32 years, including 15 years with the study PI. Siddens is trained and approved to handle extreme carcinogens and participates in OSU's medical surveillance program. As the person in charge of the LPI extreme carcinogen facility it is her responsibility to dilute extreme carcinogens such as benzo[a]pyrene to concentrations considered safe for handling per the OSU EHS guidelines. Siddens also maintains chemical and ionizing isotope inventories. Siddens is trained in proper shipping and handling of biological and hazardous materials. For this protocol, Siddens will perform handling of extreme carcinogens, dilute carcinogen solutions as well as BaP capsule preparation. Siddens was trained to carry out the capsule preparation protocol by Dr. Erin Madeen, a former graduate student in the Williams lab and person responsible for capsule preparation on previous IRBs. Madeen was a pharmacy technician before entering graduate school. The training first entailed observing Madeen prepare capsules for several cycles. Under Madeen's guidance Siddens prepared a set of capsules. Capsules were evaluated for the proper level of ^{14}C-BaP with liquid scintillation counting. This was repeated over several weeks to ensure consistency over time.			
4	Will participants be audio or video recorded for research purposes?	☐	☒	
5	If "yes", indicate whether being recorded is required in order to participate and explain this in the consent document or guide:			☐
6	Does the study involve conducting research activities online?	☐	☒	
7	Is the study designed to be implemented in phases, where fully describing one phase is dependent upon the outcome of another?	☐	☒	
	IRB approval must be obtained prior to initiating each phase.			
SECTION 21 - Compensation		Yes	No	N/A
1	Describe any compensation or incentives for participants:			☐
	Subjects will receive $125 for the BaP-only cycle and $150 each for the DIM and Brussels sprouts cycles for a total of $425 for the 3 cycles or $300 if only doing the DIM and Brussels sprouts cycles. Subjects who			

	discontinue the study early will be paid an amount pro-rated to the percentage of total blood samples completed. The amount will be determined by multiplying the number of study blood samples completed by $11.36. Subjects also receive up to $10 worth of breakfast foods and beverages on day 1 of each cycle.			
	Include details concerning the conditions under which research participants would receive partial payment or no payment at all (e.g., withdrawing early from the study)			
SECTION 22 - Costs		Yes	No	N/A
1	Describe any costs to participants that are associated with the study (e.g., parking, travel, etc.):			☐
	Subjects are responsible for transportation to and from the study site. A parking space outside of LPSC is reserved for study subjects during their visits.			
	This section should not include costs incurred by the study team or the study.			
SECTION 23 - Education Records		Yes	No	N/A
	**Education Record** is defined by FERPA as a range of information about a student that is maintained in schools in any recorded way, such as handwriting, print, computer media, video or audio tape, film, microfilm, and microfiche.			
1	Does the study involve the use of student education records?	☐	☒ *Skip to section 24*	
1a	If yes, list all data to be used (e.g. course grades, assignments, GPA, video-recordings of class activities, etc.):			
				
2	Are any of the education records related to OSU students?	☐	☐	
3	How will education records will be accessed or obtained (e.g., provided by class instructor; request will be submitted to the Office of the Registrar; etc.)?			
				
	Information on Feasibility Determinations and Data Requests through the Office of the Registrar			

SECTION 24 - Drugs or Biologics		Yes	No	N/A
	Guidance and decision trees on Drugs, Biologics, and Dietary Supplements			
1	Are one or more drugs or biologics being studied as part of this project?	☒	☐ *Skip to section 25*	
2	Drug Name (include generic and trade name, if applicable):			
	[7-^{14}C]-benzo[*a*]pyrene (BaP), specific activity 27 µCi/µmol			
3	Approval Status (e.g., FDA Approved, FDA Approved/Unapproved Use, not FDA approved):			
	not approved for therapeutic use; this protocol is an amendment to FDA IND 117175 (see IRB Protocol 8233 for FDA correspondence)			
4	Has an IND application been submitted to the FDA?	☒	☐	
5	If no, provide justification:			
6	Chemical formula:			
	$C_{20}H_{12}$			
7	Dosage strength(s):			
	50 ng			
8	Rationale for choosing the drug or substance dose:			
	The impact of diet and/or supplements on the pharmacokinetics of a PAH when an individual is exposed to an environmentally relevant defined dose has never been determined. This information will be very useful to regulatory agencies in order to incorporate this important variable into the risk assessment models for PAH mixtures that utilizes the Relative Potency Factor (RPF) approach, derived from animal studies[24]. BaP is the reference compound and has an RPF of 1. The accuracy of the RPF approach is based on the assumption that the interaction between all PAHs is additive (same Mechanism of Action or MOA) and that high-dose animal data (fed semi-synthetic defined diets) can model human exposure at environmentally relevant levels (orders of magnitude lower than the animal studies). If cruciferous vegetables and their			

	phytochemical components can impact the uptake, metabolism and/or elimination of BaP, this will have a significant impact on the validity of the RPF approach. DIM is an OTC commercially available and common supplement which has GRAS status. This study is intended to evaluate the effect on structure or function of the body (rate of metabolism and elimination of micro-doses of [^{14}C]-BaP).			
9	Method/route of administration:			
	oral in a food-grade cellulose capsule			
10	Mechanism of action:			
	BaP is not a therapeutic, so the mechanism of action described involves the current understanding of BaP carcinogenesis. The most well accepted mechanism for BaP carcinogenesis involves metabolic activation[25]. Bioactivation to the ultimate carcinogen is initiated by epoxygenation at the 7,8-position. The epoxide is hydrolyzed by the action of the enzyme epoxide hydrolase and a second epoxygenation produces the ultimate mutagenic and carcinogenic metabolite, the BaP-7,8-dihydrodiol-9,10-epoxide. The (+)-*anti*-BaP-7,8-dihydrodiol-9,10-epoxide metabolite is the most mutagenic and carcinogenic form of BaP. The metabolite is formed by oxidative metabolism by cytochrome P450 enzymes (CYPs) enzymes, primarily in the liver (also to some degree in lung and GI). The epoxide is chemically unstable and has been shown to react with the N^2 position in guanine in DNA which can lead to mutations in genome sequence. To date we have not detected any of this [^{14}C]-BaP metabolite (or tetrol hydrolysis products) by UPLC-AMS in plasma or any covalent DNA adducts in PMBCs.			
11	Known drug interactions:			
	Not applicable at this microdose			
12	Manufacturer/Sponsor:			
	American Radiolabeled Inc. (ARC, custom synthesis)			
13	Manufacturer/Sponsor Location:			
	St. Louis, Missouri			
	If manufactured on site at OSU, provide details of facilities and restricted access.			
14	Name of supplier:			
	Same as Manufacturer (custom synthesis)			

15 Summarize preclinical and early human studies (for studies that are required to have an investigational new drug (IND) number):

BaP is one of the most extensively studied PAH environmental contaminants. BaP is a skin carcinogen in the rodent 2-stage model involving dermal application and promotion by TPA[1,2]. In addition to dermal exposures, BaP has been documented as an animal carcinogen following oral or inhalation exposures. Target tissues include liver, forestomach, esophagus, auditory canal and oral cavity. Occupational exposures in humans are associated with increased incidences of cancers of the lung, skin and bladder[2].

Animal studies in mouse and rat confirm the carcinogenic potential of BaP when administered by the oral route, a mimic of human exposure[2]. Dose-dependent appearance of tumors can be observed in the forestomach, esophagus, tongue and larynx in a somewhat reliable manner. A summary of animal studies in which BaP was administered by the oral route is provided in the table below, the dose information was converted to mg/kg for ease of comparison. The summary only includes studies which evaluated untreated control animals as a negative control.

Animal Studies with Orally Administered Benzo[a]pyrene

Species	Dose (mg/kg)	Route; Regimen	Duration	Tumor Observations	Reference (all in Ref 2)
Mouse,A/J	0 550 3,350	Oral, Diet	260 days	Forestomach tumors 0/21 (0%) 5/25 (20%) 27/27 (100%)	Weyand et al., 1995 Chem Res Toxicol 8(7): 949-954
Mouse, B6C3F1	0 5 25 100	Oral, Diet	2 years	Forestomach tumors 1/48 (2%) 3/47 (6%) 36/46 (78%) 46/47 (98%)	Culp et al., 1998 Carcinogenesis 19(1): 117-124
Mouse,Swiss	0 50	Oral 2X/week, 4 weeks	27 weeks	Forestomach tumors 0/10 (0%) 10/10 (100%)	Badary et al., 1999 Eur J Canc Prev. 85 435-440

Mouse, Muta	0 75 125	Oral, 5 consec. days	41 weeks	Forestomach tumors 0/8 (0%) 10/10 (100%) 10/10 (100%)	Hakura et al., 1998 Regul Toxicol Pharmacol 27(3): 273-279
Rat, Sprague Dawley	0 6 39	Oral, 1x/9 days 5X/week	Lifespan	Combined tumors 3/64 (4.7%) 3/64 (4.7%) 10/64 (15.6%)	Brune et al., 1981 J Canc Res Clin Oncol 102(2): 153-157
Rat Crl:CD	0 63	Oral, 1x/week 8 weeks	49 weeks	Mammary Tumors 1/30 (3%) 11/30 (37%)	El-Bayoumy et al.,1995 Carcinogenesis 16(2):431-434

The mouse study of Culp et al., 1998 (see above Table) utilized the lowest exposure with 5 mg/kg (a dose 100,000 times greater than the dose in our microdose studies) which did not produce a significant increase in tumor incidence. This is similar to the rat study of Brune et al., 1981 (see above Table) in which a dose of 6 mg/kg did not result in an increase in tumors when animals were monitored for their lifetime.

16	Will an FDA-approved drug be administered for an indication, dose, route of administration, or subject population that is different from what has been approved for the marketed product? If "yes", explain and provide justification and safety information:	☐	☒	
17	Provide the plan for the storage, dispensing, handling, inventory control, and disposal of investigational, FDA-approved drugs, and/or biologics:			
	Preparation Location: The lab where the capsules are prepared (LPSC 383) is not a radiopharmacy. Because these capsules are created solely for the purposes of this study and are not to be considered a therapeutic drug, they are not made to GMP standards but will be prepared with the highest quality, consistency and precision possible. Room 383 is a locked, secure laboratory, and no other activities occur in this space besides those related to BaP handling and capsule preparation. Siddens and Williams are the only persons with keys to access this space.			

Capsules intended for human use will be prepared in a clean hood dedicated solely to capsule preparation. Concentrated ^{14}C-BaP stock solution is not diluted in the same area as capsule preparation. The hood where capsule preparation is performed will be surveyed for radioactivity by taking swipes and evaluating with liquid scintillation counting. The capsule preparation hood will be sterilized with bleach and alcohol, and all work surfaces will be covered with fresh, plastic backed laboratory bench paper. All pipettes are calibrated, dedicated to this procedure, and only used with sterile, filter pipette tips. All Hamilton syringes were purchased new, checked for accuracy, and dedicated to this project.

Material storage & preparation: Prior to use, empty Solaray® capsules and pharmaceutical grade lactose monohydrate NF (Spectrum Chemical Mfg. #61-1730890) are stored in an airtight bag in a gasket sealed plastic box, in a cabinet in a lab that does not utilize scintillation-detectable radioactivity or human tissues. This box contains all microdosing supplies in an area separated from traditional laboratory supplies.

The [^{14}C]-BaP is shipped as a concentrated solution (0.10 mCi/mL; specific activity = 27 nCi ^{14}C/nmol BaP = 0.103 nCi/ng) in toluene. This concentrated solution is stored in sealed containers as received from the supplier at -80°C in a secure carcinogen laboratory in LPSC 383. In order to ensure radiochemical purity (greater than or equal to 98% for the purified concentrated stock) and to prepare a diluted sample suitable for human consumption, the compound is collected as a single peak from an HPLC run. The identity of the compound is confirmed by co-elution with a non-radioactive standard.

A working stock solution will be prepared by diluting the 0.10 mCi/mL [^{14}C]-BaP concentrated solution. The stock solution will contain 50 ng [^{14}C]-BaP /25 µL = 2 ng/µL (5.4 nCi ^{14}C/25 µL = 0.2 nCi/µL). For example, to make 4 mL of stock solution, 8 µL of the 0.10 mCi/mL solution will be aliquoted into a clean vessel. Toluene is carefully evaporated off, and the [^{14}C]-BaP aliquot resolubilized into 4 mL 95% food grade ethanol. The radioactivity will be checked with a liquid scintillation counter and adjusted to the target concentration if needed.

The stock solution will be aliquoted into clean 2 mL amber glass vials and stored under argon at -80°C. Each stock solution is checked for ≥ 95% [^{14}C]-BaP radioisotope purity on a quarterly schedule throughout the study. Stock solutions will be checked for both purity and concentration on UPLC using UV detection for BaP and radioisotope detection for [^{14}C].

Capsule preparation, verification and storage: Siddens and Uesugi have been trained to prepare the capsules. As an added safety measure, a second trained study team member will also be present during capsule preparation for quality control to verify capsules are produced per specified SOPs.

Each capsule will contain only the [^{14}C]-BaP and lactose as an excipient. Empty vegetarian capsules, size 0,

composed of vegetable cellulose, will be opened and held in a microtube rack. The empty capsules will be filled with lactose. The 25 µL aliquot of either stock solution will be applied to the lactose with a calibrated pipet fitted with a sterile filter tip. The ethanol is allowed to evaporate, and then the capsules are sealed with their caps.

Five (5) capsules will be prepared in each batch: Up to two (2) subjects may participate in a study cycle per week and three (3) capsules will be used for quality control verification.

> Capsules for quality control: Immediately after preparation, 3 of the 5 capsules will each be dissolved in 5 ml ultrapure water and 15 ml scintillation cocktail and counted on a liquid scintillation counter. Results will be emailed directly to the director of OSU Radiation Safety (RSO). Once the RSO has determined that the three evaluated capsules are within 10% variance and 10% error, the results are emailed to Siddens or Uesugi. Siddens will then contact Williams and Uesugi that the cycle(s) may proceed. If the capsules are not given RSO approval, a new set of capsules will be prepared and the subject(s) will be rescheduled if needed.
>
> Capsules for subjects*:* Two capsules will be stored in sealed vials in the dark in a zipper-lock plastic bag with silica desiccant at -20°C until subject consumption. The bag will be labeled appropriately (see sample label). Any capsules not used within 1 week will be discarded.

Dispensing: Capsules will be transported to the Clinical Research Center (407 LPSC) in sealed vials within the labeled zipper-lock plastic bag. The bag will be stored within a plastic box for transportation to the clinic.

One (1) capsule containing 50 ng [^{14}C]-BaP will be administered to the subject with 100 mL of water by the nurse coordinator at the start of the study cycle.

Inventory control: Information including number of capsules, date of preparation, and operator are recorded immediately after capsules are prepared. Date of consumption and disposal of test capsules are recorded on day 1 of the cycle. If test capsules do not pass approval by the OSU radiation safety officer, the information is still recorded and a new set of capsules will be prepared.

Disposal: Capsules that are not needed, expired or rejected for human use will be disposed of in the radiochemical dry waste at Oregon State University.

	Please see IRB Protocol 8233 for lactose, capsule, and [^{14}C]-BaP quality assurance documents.			
18	Does the use of the test article involve a route of administration or dosage level, use in a subject population, or other factor that significantly increases the risks (or decreases the acceptability of the risks) associated with its use? If "yes", explain:	☐	☒	
	As noted above, the use of [^{14}C]-BaP is not intended to be therapeutic. The great majority of exposure (>95%) of the larger molecular weight PAHs, such as BaP, is through the diet in a variety of foods including breads and cereals, grains, vegetables and smoke-cured or barbequed meats[2]. Dietary intakes of total PAHs in the U.S./North America have been estimated at 160-3,000 ng/day with BaP alone at 270-700 ng/day. In Asia, dietary levels of total PAHs are estimated at 55,000 ng/day[2]. A 2005 report from the FAO/WHO Joint Expert Committee on Food Additives and Contaminants listed a mean BaP daily dietary intake of 270 ng (70 Kg individual) with 700 ng as a high-level intake[1]. The European Union maximum limit for BaP in smoked meats is 5,000 ng/Kg fresh weight. The dose of 50 ng would be the equivalent of approximately 10 g of smoked meat at the EU allowable limit. Compiling all of the animal data, a **Virtually Safe (Lifetime) Dose (VSD) of 42-350 ng/person/day** has been established as the best estimate for a lifetime exposure to BaP producing no more than 1 cancer per million people[2-4]. Therefore, based on the chemical mass involved in microdosing (significantly lower than background exposure and levels determined to be VSD), this study poses *de minimus* risk to subjects.			
	Potential attachments triggered by this section: • Investigator brochure • Approved labeling • Package insert • Documentation of quality or purity or Certificate of Analysis • "Safe to Proceed" from the FDA • Correspondence from the FDA • Letter from the FDA or industry sponsor setting forth the IND number			
SECTION 25 - Dietary Supplements		Yes	No	N/A
	A **dietary supplement** is a product taken by mouth that is intended to supplement the diet and that contains one or more "dietary ingredients". The "dietary ingredients" in these products may include: • Vitamins • Minerals • Herbs or other botanicals • Amino acids			

	• Other substances found in human diet, such as enzymes In some cases, dietary supplements or substances generally recognized as safe (GRAS) are considered to be drugs when they are used to diagnose, cure, mitigate, treat, or prevent disease. Under FDA regulations, research that involves use of a drug other than the use of a marketed drug in the course of medical practice must have an Investigational New Drug (IND) Application, unless the study meets one of the exemptions from the IND requirement.			
	Guidance and decision trees on Drugs, Biologics, and Dietary Supplements			
1	Are one or more dietary supplements being studied as part of this project?	☐	☒ *Skip to section 26*	
2	Name of dietary supplement (include generic and trade name, if applicable):			
	3,3'-diindolylmethane (DIM) - This protocol involves the use of a dietary supplement (BioResponse DIM® 150), but the supplement is not being studied.			
3	Approval Status (e.g., FDA Approved, FDA Approved/Unapproved Use, not FDA approved):			
	FDA Approved			
	If a dietary supplement is being used in the study as a drug, the study is regulated by the FDA. Please see the Guidance on Drugs, Biologics, and Dietary Supplements for additional information.			
4	Has an IND application been submitted to the FDA?	☐	☒	
5	If no, provide justification:			
	We are studying the metabolism of [^{14}C]-BaP, not BioResponse DIM® 150.			
6	Chemical formula:			
	$C_{17}H_{14}N_2$			
7	Dosage strength(s):			
	300 mg per day			
8	Rationale for choosing the drug or substance dose:			

	BioResponse DIM® 150 (purchased locally) will be used as the standardized source of 3,3'-diindolylmethane (DIM). We have chosen the BioResponse supplement, rather than purchasing and encapsulating DIM from a chemical supply company, as the oral bioavailability is significantly greater[26] and it more accurately reflects human consumption.			
9	Method/route of administration:			
	Subjects will be instructed to take 300 mg (2 x 150 mg capsules) orally with dinner for 7 days prior to the study cycle.			
10	Mechanism of action:			
	In animal models, DIM is capable of inducing or inhibiting phase 1 (e.g., CYP1A1; CYP1B1) and phase 2 (e.g., GSTM1 or UGT1A10) enzymes known to metabolize BaP for elimination. In human clinical trials (for prevention of breast cancer) it has been shown to increase the ratio of (2-hydroxy E2/16α-hydroxy E2). The 2-hydroxy metabolite is sometimes referred to as the "good estrogen" and 16α-hydroxy as the "bad estrogen"[10]. CYP1B1 is capable of metabolizing estrogen to 4-hydroxy-estrogen which is a "catechol" capable of producing oxidative stress at high levels. DIM actually does not impact CYP1B1 markedly and most reports from animal studies indicate inhibition (which would be protective).			
11	Known drug interactions:			
	It is possible that DIM could impact the metabolism of other substrates for these enzymes. However, CYP1A1 and CYP1B1 (required for formation of carcinogenic metabolites) rarely are involved in metabolism of drugs. This fact and our exclusion criteria with respect to use of therapeutic drugs makes drug interaction unlikely. There are no known drug interactions in humans taking DIM reported in the literature. In addition, the impact of DIM would be transient as it has a half-life in humans of a few hours. Any induction or inhibition of enzymes would only last 1-3 days as these enzymes turnover fairly rapidly. It is possible that there could be a transient change in estrogen metabolite levels in women taking DIM.			
12	Manufacturer/Sponsor:			
	BioResponse, LLC			
13	Manufacturer/Sponsor Location:			
	Boulder, Colorado			
14	Name of supplier:			

	BioResponse, LLC, purchased locally.			
15	Summarize preclinical and early human studies (for studies with an IND):			☐
	n/a			
16	Will an FDA-approved drug be administered for an indication, dose, route of administration, or subject population that is different from what has been approved for the marketed product? If "yes", explain and provide justification and safety information:	☐	☒	
17	Provide the plan for the storage, dispensing, handling, inventory control, and disposal of the dietary supplement:			
18	Does the use of the test article involve a route of administration or dosage level, use in a subject population, or other factor that significantly increases the risks (or decreases the acceptability of the risks) associated with its use? If "yes", explain:	☐	☒	
	Potential attachments triggered by this section: • Investigator brochure • Approved labeling • Package insert • Certificate of Analysis or other documentation of quality/purity • "Safe to Proceed" from the FDA • Correspondence from the FDA • Letter from the FDA or industry sponsor setting forth the IND number			
SECTION 26 - Medical Devices		Yes	No	N/A
	A **medical device** is an instrument, apparatus, implement, machine, contrivance, implant, in vitro reagent, or other similar or related article, including a component part, or accessory which is: • recognized in the official National Formulary, or the United States Pharmacopoeia, or any supplement to them, • intended for use in the diagnosis of disease or other conditions, or in the cure, mitigation, treatment, or prevention of disease, in man or other animals, or			

	• intended to affect the structure or any function of the body of man or other animals, and which does not achieve any of its primary intended purposes through chemical action within or on the body of man or other animals and which is not dependent upon being metabolized for the achievement of any of its primary intended purposes.			
	Guidance and decision trees on medical devices			
1	Are one or more medical devices being studied as part of this project?	☐	☒ *Skip to section 27*	
2	Indication if data related to the safety or efficacy of the device will be collected:			
				
3	Rationale for choosing the device to be used:			
				
4	Device description:			
				
	The device description can include: • pictures of the device (where applicable); • physical, chemical and/or biological processes/principles used by the device to generate device output, if applicable; • physical and biological characteristics of the device output, if applicable; • explanation of the user interface and/or how the device interacts with other devices or with the user (medical professional and/or patient); • explanation of the materials used in the device; • a brief explanation of how the device is manufactured (where necessary); • discussion of the mechanism of action and how the device and/or, if applicable, device output is used; • for an IVD, detailed technical description of your device including instruments, reagents, components, software, principles of operation, and accessories (if there are changes to a previously cleared or approved device, then you should describe these changes); • discussion of the scientific basis for development of the device or an explanation of expected clinical utility;			

	In addition to pictures and a written description, other information about the clinical use of the device, such as a surgical technique guide or video of how the device is used in the clinical setting, may be helpful.			
5	Proposed Intended Use/Indications for Use:			
				
	Use description can include: • identification of the disease or condition the device is indicated to prevent, mitigate, screen, monitor, treat, or diagnose; • identification of the target population; • part of the body or type of tissue to which applied or with which the device is interacting; • frequency of use; • physiological use; • Statement of whether investigators intend to commercialize or market the device; statement of whether the device is intended for prescription and/or over-the-counter use; For an IVD device, this information should include a detailed draft of the intended use of the device including the intended use population, the analyte/condition to detect, and the assay methodology.			
6	Is there an intent to commercialize or patent the device?	☐	☐	
	If there is commercialization intent or if investigators hold IP rights or interests in this device, it is considered a financial interest that should be disclosed in the conflict of interest section of the protocol. Additional disclosures may be required in the consent form.			
7	Will the device(s) be stored in a locked environment under secure control with limited access and in an area of the PI's control?	☐	☐	
8	Will proper instructions on the use of the device will be provided to the subjects?	☐	☐	
9	Will a log be kept regarding the receipt, use, and/or dispensing of the device and the disposition of remaining devices at the conclusion of the investigation?	☐	☐	
10	Will the device be labeled in accordance with the FDA's requirements?	☐	☐	
	Label that will be placed on the device; including an indication of single-use or directions for sterilization between uses.			

SECTION 27 - Food or Beverages		Yes	No	N/A
	If food or food extract will be used as a drug, complete the drug section above instead.			
	**21CFR172**: Food Additives Permitted For Direct Addition To Food For Human Consumption (FDA approved) **21CFR182**: Substances Generally Recognized As Safe (FDA affirmed) **21CFR184**: Direct Food Substances Affirmed As Generally Recognized As Safe (FDA affirmed) **21CFR186**: Indirect Food Substances Affirmed As Generally Recognized As Safe (FDA affirmed) **FEMA GRAS LIST**: Flavor & Extract Manufacturers Association, Flavor Ingredient Library (FDA consulted and had no questions) **EAFUS**: Everything Added to Food in the United States. Inclusion on this list does not indicate approval or affirmation – it simply provides the references and regulations pertaining to a food ingredient			
SECTION 27.1 - Food as Courtesy or Compensation		Yes	No	N/A
1	Does the study involve providing participants with commercially purchased food intended as a courtesy or compensation?	☒	☐ *Skip to section 27.2*	
2	If the study involves providing participants with commercially purchased food intended as a courtesy or compensation, will allergen information (clear labeling or menu with ingredients listed) be provided to the participants? If no, explain why:	☒	☐	
				
SECTION 27.2 - Food as Intervention		Yes	No	N/A
1	Does the study involve participants ingesting, tasting, or smelling a food or beverage, or a component thereof for the purpose of research?	☒	☐ *Skip to section 28*	
2	Name of food, beverage, or component thereof:			
	Brussels sprouts			
3	Supplier (e.g., manufacturer name, grocery store, restaurant):			
	Local grocery store			
4	Is this study a taste and food quality evaluation or consumer acceptance study?	☐	☒	

5	Will participants consume wholesome foods without additives?	☒	☐	
	21CFR172: Food Additives Permitted For Direct Addition To Food For Human Consumption (FDA approved)			
6	Will the food or beverage consumed contain only ingredients that are at or below the level and for a use found to be safe by the Food and Drug Administration?	☒	☐	☐
7	Will the food or beverage consumed contain only agricultural chemicals or environmental contaminants at or below the level found to be safe (or approved) by the Environmental Protection Agency or the Food Safety and Inspection Service of the U.S. Department of Agriculture?	☒	☐	☐
8	Has there been a determination by the FDA that the product is Generally Recognized as Safe (GRAS)?	☐	☒	
9	If yes, provide GRAS number:			☒
10	Has the determination of GRAS been self-affirmed as a result of published safety data?	☐	☐	☒
11	Is the food, beverage, or component food-grade and intended for use in humans? If yes, provide documentation. If no, explain and indicate how safety for use in humans is known and attach documentation that demonstrates substantial equivalence to ingredients found in the food supply:	☒	☐	
	Brussels sprouts are a commonly consumed whole food.			
	Certificate of Analysis from the manufacturer; package insert			
	All GRAS substances are food-grade. The same substance at another grade is not GRAS.			
12	Risks to subjects are minimized by ensuring that the proposed products are food-grade. If product is not food-grade, compare the analysis to the same product in the USP Food Chemicals Codex and provide an assessment of whether the proposed product is substantially equivalent:			☒
13	Provide the plan for the storage and handling of the food:			
	The Brussels sprouts used in this study will be commercially frozen and purchased at a local grocery store. We will re-package them into individual daily serving portions and weigh portions in the OSU Moore Family Center kitchen in Milam Hall. Subjects will receive 7 days' worth of frozen Brussels sprouts and instructed to thaw, gently warm, season as desired and consume each evening with dinner for 7 days prior to the study cycle. Study team members handling and preparing the Brussels sprouts will have a current Oregon			

	Food Handler's License and will follow the kitchen preparation and sterilization procedures as outlined in the Oregon Health Authority, Division 150, and Food Sanitation Rule.			
	GMP certificate or food-grade certificate that indicates that the product can be lawfully used in the US food supply Documentation that the product is an approved food additive (21 CFR 172) Safety data if GRAS status is self-affirmed			
SECTION 28 - Radiation		Yes	No	N/A
	The Radiation Safety Officer will review this submission.			
1	Does the study involve exposing participants to radiation?	☒	☐ *Skip to section 29*	
2	Will any participant be asked to undergo an x-ray procedure (including radiographic and DEXA) while in this study?	☐	☒	
2a	All x-ray procedures are for research use only, and will not be used for medical screening or diagnosis.	☐	☐	
2b	If diagnostic x-ray scans are performed for medical screening, and all procedures are routine, standard, clinical procedures.	☐	☐	
2c	All participants receiving a diagnostic radiation procedure would have the same procedure for clinical reasons even if they were not in this study.	☐	☐	
3	Will radioactive materials (including nuclear medicine, metabolic, nutrition, toxicity, or drug studies) be administered to any participants as part of this study?	☒	☐	
3a	All procedures involving administration of radioactive materials are routine standard, clinical procedures prescribed by a physician.	☐	☒	
3b	All participants receiving a radioactive material would have the same procedure for clinical reasons even if they were not in this study.	☐	☒	
4	Summarize the "authorized users" training and experience in the use of (ionizing) radiation-emitting devices and/or radioactive materials with human participants:			
	PI/Program Director Williams oversees all radioisotope activities in his laboratory and has made use of radioisotopes his entire career. His experience includes use of many carcinogens, drugs and phytochemicals labeled with a variety of different isotopes covering a range of doses in the laboratory and in animals. This is the fifth human subject study he has conducted as PI using ultra low dose ^{14}C-labeled carcinogens administered to human subjects to study carcinogen metabolism at everyday levels of exposure.			

	Lisbeth Siddens has worked at OSU for over 30 years as a laboratory technician and has worked on a variety of procedures utilizing isotope-labeled chemicals. Recent work has focused primarily on ^{3}H- and ^{14}C-labeled carcinogens. Siddens has previously done radiation compliance for other OSU faculty and is currently doing so for the Williams laboratory. She is also approved by chemical safety for handling of extreme carcinogens so her experience is ideally suited for this study. Siddens cross-trained with McQuistan and Madeen, former study team members involved in carcinogen and isotope handling, record training, stock preparation and capsule preparation for this study. Sandra Uesugi, RN has previous laboratory experience handling ^{14}C-labeled radioisotopes. Uesugi is approved to assist Siddens with capsule preparation for study subjects providing quality control and to ensure that a second trained individual is always present to assist and confirm and verify capsules are produced per specified SOPs. Uesugi has also been trained by Siddens to prepare BaP capsules in the event that Siddens is not available to prepare capsules for this study.			
5	Will ionizing or radiation-emitting devices/procedures or drugs be used or evaluated as part of this research protocol?	☐	☒	
6	For each **x-ray machine and associated procedure**, indicate the total number of exposures per single study and per complete research protocol. A single study is comprised of all procedures performed on a participant during a single visit/session.			☒
6a	Device or procedure name:			
6b	Total exposures per participant, per study visit:			
6c	Total exposures per participant, per protocol:			

7	For each **radioactive material**, indicate the activity (mCi) per single dosage, and the total number of dosages per single study and per complete research protocol. A single study is comprised of all dosages received by the participant during a single visit/session.			☐
7a	Radioactive material name:			
	[7-^{14}C]-benzo[a]pyrene (BaP), specific activity 27 µCi/µmol			
7b	mCi dosage:			
	0.0000054			
7c	Total dose per participant, per study visit:			
	1			
7d	Total exposure per participant, per protocol:			
	3 (or 2- if subject has completed Protocol 8233 or Protocol 8554 within the past 12 months, they will complete only the Brussels sprouts and [^{14}C]-BaP cycle and DIM and [^{14}C]-BaP cycle, not [^{14}C]-BaP -only cycle)			
8	**Radioactive drug(s)** prepared on-site will be prepared, assayed, tested, and labeled in accordance with the FDA's requirements?	☒	☐	
9	**Radioactive drug(s)** prepared on-site will be prepared, assayed, tested, and labeled in accordance with an Radioactive Drug Research Committee's (RDRC) requirements?	☐	☒	
SECTION 29 - Biological Samples		Yes	No	N/A
1	Does the study involve the collection or receipt of biological samples?	☒	☐ *Skip to section 30*	
1a	If yes, indicate what biological samples will be received or collected:			
	Blood and urine			
2	Will samples be obtained prospectively from living individuals?	☒	☐	
3	Where possible, risks should be minimized by using procedures already being performed on the subjects for diagnostic or treatment purposes. Check all that apply:	☒	☐	

3a	Samples are being collected *separately* from any clinically indicated procedure or another approved study	☒	☐	
3b	Study involves taking *additional* samples during clinically indicated procedure or another approved study	☐	☒	
3c	Study involves using samples *leftover* from a clinically indicated procedure or another approved study	☐	☒	
4	If blood samples will be collected, provide the maximum volume to be collected in an 8-week period and the frequency of collection:			☐
	The maximum amount of blood collected in an 8 week period is 310 mL. The [^{14}C]-BaP only cycle involves 120 mL of blood collection. The amount collect for the Brussels sprouts and DIM cycles are each 155 mL. The total amount of blood collected from each subject is 430 mL (1 cycle at 120 and 2 at 155 mL) or 310 mL if they are only doing Brussels sprouts and DIM cycles (2 x 155 mL/cycle).			
	Guidance on Minimal risk blood draw			
5	Will any clinical lab testing be conducted? If yes, describe the purpose of the tests and whether results will be disclosed to participants and/or their treating physician:	☒	☐	
	We will conduct a urine pregnancy test on female subjects at the screening visit and on day 1 of each cycle to ensure no potential for fetal exposure to [^{14}C]-BaP. A positive test result will exclude subjects from further study participation. Results of these tests will neither be disclosed to subjects nor their physician.			
6	Will the human biological material be tested/collected in a CLIA-certified lab?	☐	☒	☐
	If yes, attach the CLIA certificate			
	Results from lab tests, including urine pregnancy tests, cannot be disclosed to participants unless the lab conducting the test is CLIA-certified. For more information, please see the OSU Guidance for CLIA Certification: http://research.oregonstate.edu/irb/clia-certification-guidance			
		Yes	No	N/A
7	Will the study involve genetic testing?	☒	☐	
7a	Will test results be disclosed to the participant or their physician?	☐	☒	☐
7b	Will disease risk be quantified, including the limits on certainty of the testing?	☐	☒	☐
7c	Will a change in a family relationship be disclosed, such as mistaken paternity?	☐	☒	☐

7d	Does the participant or family member have the option not to know the results? If yes, how will this decision be recorded:	☒	☐	☐
	Subjects will not be contacted in the future as there will be no relevant genetic clinical data to share. The subject can indicate on the consent form whether or not s/he agrees to have a small number of genes (maximum 7) known to be involved in metabolism and excretion of BaP analyzed. The results will not be shared as there is no established disease risk.			
7e	Could other clinically relevant information be uncovered by the study? If yes, explain how (or if) disclosure of this information will occur:	☐	☒	☐
				
7f	Do any practical limitations exist on the participant's right to withdraw from the research, withdraw data, and/or withdraw DNA? If yes, explain:	☐	☒	☐
				
7g	Is the participant permitted to participate in the study if they decline to participate in the genetic testing?	☒	☐	☐
8	Will samples be stored for future studies?	☒	☐ *Skip to 8e*	
8a	Indicate how long samples will be retained, how they will be stored, and what they will be used for:			
	Samples and data will be identified only by an anonymous code. After analysis for this study, the coded samples will be stored in LPSC 389 at OSU indefinitely unless not allowed by subjects. Data will be stored securely on password protected computers or in locked file cabinets in LPI. If subjects decline permission to store samples beyond the scope of this study, their samples will be properly discarded after analysis. Samples and data will only be used in future PAH-related studies.			
8b	The information in the consent form should convey the disease, condition, or specific field of study for future projects. Explain whether and how participant permission will be sought for future studies of existing samples:			
	As stated in the consent form, we will not contact subjects for permission to use their samples and data in future projects. The consent form includes a section for subjects to give permission to use their samples and data in future projects without being contacted for consent. They may withdraw permission at any point during the study.			
8c	Indicate whether participants will be contacted by researchers in the future for the purpose of updating information:			

	No			
8d	Indicate whether and how participants can opt out of any sharing or future use of their sample:			
	The consent form includes a section for subjects to give permission to use their samples and data in future projects without being contacted for consent. They may change their answer at any point during the study. Since the AMS analysis at LLNL is the primary method of analysis for this study, subjects will not be offered the option to opt out of sharing their sample with LLNL.			
	The Biological Safety Officer will review this portion of the submission unless the materials are exempt from biosafety review			
8e	Provide location(s) of material handling, manipulation, and storage at OSU (i.e., building and room):			
	Samples will be collected in LPSC 407. Processing and storage will take place in LPSC 389.			
8f	Provide the names of all personnel who will be working with biological materials:			
	Lisbeth Siddens and Sandra Uesugi			
	All personnel who will be working with biological materials must have current Blood-borne Pathogens Training and any required vaccinations. Information about current status accessed from the EH&S website			
SECTION 30 - Privacy and Confidentiality		Yes	No	N/A
	Many of the terms used in this section are defined in the glossary under the heading "Privacy, Confidentiality, and Identifiers".			
1	*Privacy*, in the context of a research protocol, means respecting an individual's right to be free from unauthorized or unreasonable intrusion, including control over the extent, timing, and circumstances of obtaining personal information from or about them. Explain how privacy will be respected when identifying and recruiting potential participants:			☐
	With the exception of contacting individuals on the LIFE registry maintained by the Center for Healthy Aging Research (CHAR) at OSU, only subjects requesting information about the study will be contacted, and no subjects will be prospectively contacted by researchers without contact first being initiated by the individual. We will protect email correspondence by deleting after the subject has completed the study, is removed, withdraws from the study, or has been found to not qualify. Telephone and in-person screenings will take place in a private location with time between subject visits to maintain privacy.			

	We will immediately destroy notes taken during telephone screening conversations with excluded subjects. We will securely retain records from subjects who are excluded from the study after being enrolled.			
2	Will direct and/or indirect identifiers be requested or recorded?	☒	☐	
3	If no, will all data be collected anonymously or provided to researchers without identifiers?	☐ *Skip to section 31*	☐	☒
4	List the direct identifiers (e.g., names, social security numbers, addresses, telephone numbers, student ID, medical record number, mTurk ID, photographs, video recording):			☐
	Name, address, email address, telephone number, date of birth			
5	Indicate whether identifiers or codes will be retained that could link the identity of the participant to the sample:			☐
	Samples will be labeled only with codes. Only study team members will have access to study samples. If a student employee is hired to assist with processing of de-identified study samples, s/he will always be supervised by Siddens, Uesugi or Williams.			
6	List the indirect identifiers (e.g., combination of demographic and other variables such as gender, race, ethnicity, age, zip code, company affiliation, class standing, department, audio recording):			☐
	Age, gender, race, ethnicity – this data will be collected on forms containing only subject codes, not direct identifiers			
7	Describe the steps that will be taken to minimize the chances of a breach of confidentiality during and after data collection (e.g., coding system, pseudonyms, etc.)			☐
	Only the nurse coordinator will have access to the document linking identifiers to subject code numbers. Personal information collected during telephone screening, the consent documents, and compensation acknowledgement form will be the only documents containing direct identifiers. We will store this information separately from coded data. These documents will be stored in a locked file cabinet in a secure office. Direct identifiers will be stored in hard-copy only.			
	Details of data security will be collected in later section.			
8	Will a copy of the consent form, test results, or other research study information be placed in the participants' record (e.g., medical, personnel, or education record)?	☐	☒	
SECTION 31 - Record Retention		Yes	No	N/A
1	Will the Principal Investigator store research records in a secure and audit accessible manner for a	☒	☐	

	minimum of three years post-study termination?			
2	If not, provide justification for early destruction:			☒
				
3	Will the student researcher <u>also</u> store research records after the study has closed?	☐	☐	☒
3a	Will the records stored by the student contain individually identifiable information?	☐	☐	☒
4	If the study is FDA-regulated, confirm the PI will <u>also</u> comply with the following relevant records retention requirements:			☐
4a	In accordance with **21 CFR 312 (drugs)**, an investigator or sponsor shall retain the records and reports for 2 years after a marketing application is approved for the drug; or, if an application is not approved for the drug, until 2 years after shipment and delivery of the drug for investigational use is discontinued and FDA has been so notified.	☒	☐	☐
4b	In accordance with **21 CFR 812 (devices)**, an investigator or sponsor shall maintain the records required by this subpart during the investigation and for a period of 2 years after the latter of the following two dates: The date on which the investigation is terminated or completed, or the date that the records are no longer required for purposes of supporting a premarket approval application or a notice of completion of a product development protocol.	☐	☐	☒
5	Will a link between study code numbers and direct identifiers be retained after data collection is complete?	☐	☒	☐
6	If yes, explain why this is necessary and state how long the link will be retained:			☐
				
7	If audio and/or video recording, indicate whether these files will be destroyed after transcripts and/or coding is verified. If A/V files will be retained, provide justification for retention:			☒
				
8	Will data be stored for future studies?	☒	☐ *Skip to next section*	
8a	Indicate how long data will be retained, how it will be stored, and what it will be used for:			
	See section 29			
8b	The information in the consent form should convey the area of study for future projects. Explain whether and how participant permission will be sought for future studies of existing data:			

				
8c	Indicate whether participants will be contacted by researchers in the future for the purpose of updating information:			
				
8d	Indicate whether and how participants can opt out of any sharing or future use of their data:			
				
SECTION 32 – Sharing Data and Biological Samples		Yes	No	N/A
1	Will data and/or samples be shared with individuals or entities external to OSU (e.g., made public, shared with sponsor, sent to collaborators, given to people at the site of research, etc.)?	☒	☐ *Skip to section 33*	
1a	Why and with whom?			
	A portion of the de-identified coded study samples will be forwarded to LLNL for analysis with no personal identifying information contained within the shipment, so there is no risk of loss of confidentiality with the mailing of samples nor the analysis of samples at LLNL. Data returned to OSU will be identified only be de-identified codes, so there is no risk of loss of confidentiality.			
1b	Will shared data and/or samples be individually identifiable?	☐	☒	
1c	Will data security plan at the external site match or exceed the OSU data security plan?	☒	☐	☐
1d	If the data security plan at the external site will not match or exceed the OSU data security plan, explain:			
				
1e	Describe how security will be maintained in transit:			☐
	A portion of the de-identified coded study samples will be forwarded to LLNL for analysis with no personal identifying information contained within the shipment, so there is no risk of loss of confidentiality with the mailing of samples nor the analysis of samples at LLNL. Data returned to OSU will be identified only be de-identified codes, so there is no risk of loss of confidentiality.			
2	If the study is federally funded, provide the plan to comply with federal data sharing requirements:			☐
	Oregon State University adheres to the NIH Grants Policy Statement on Availability of Research Results: Publications, Intellectual Property Rights, and Sharing Research Resources as described in https://grants.nih.gov/grants/policy/nihgps_2013/nihgps_ch8.htm#_Toc271264947. Specifically, material transfers would be made with no more restrictive terms than in the Simple Letter Agreement or the UBMTA and without reach through requirements. Should any intellectual property arise which requires a			

	patent, we would ensure that the technology remains widely available to the research community in accordance with the NIH Principles and Guidelines document. We do not anticipate generating and unique genetic animal or biological reagents (e.g., plasmids). All publications and presentations of results will be made available to interested researchers. The centralized data management organization facilitating the publication of data to the NIEHS Chemical Effects in Biological Systems Knowledge Base for public availability of Program data. The bioinformatics software updates will be released in the Bioinformatics Resource Manager (BRM) software as version 2.2. BRM is a client-server application, and account registration is required to store each user's personal data securely on remote servers. We also want to ensure our AMS data with human carcinogen micro-dosing, is available to EPA as quickly as possible. We intent to provide EPA ORD, NCEA, IRIS with password protected access to de-identified clinical data at the earliest possible time.			
3	Will the intent to share data be disclosed to research participants as part of the consent process? If not, provide justification:	☒	☐	
SECTION 33 – Publication		Yes	No	N/A
1	Could participants be identifiable in publication or presentation (e.g., results will be reported using direct quotes, group or tribe name, company name and position title)	☐	☒	
2	Will manuscripts, presentation materials, theses, or dissertations be stored in Scholars Archive?	☒	☐	
3	Will individually identifiable data or specimens be stored in an archive or repository? If yes, describe:	☐	☒	

SECTION 34 - Data Security

Use this matrix to determine the data security level and related requirements for this study.		Breach of confidentiality poses what level of risk?		
		No Risk	Minimal Risk	Greater than Minimal Risk
Are data and/or subjects:	De-Identified or anonymous?	Level 1	Level 1	Level 2
	Identifiable or coded?	Level 1	Level 2	Level 3

	Complete only the subsection for the appropriate level of data security below.			
SECTION 34.1 - Level 1		Yes	No	N/A
1	Will the following security **requirements** be met: • Information will be shared and stored in a manner that provides access only to authorized individuals. • If information is stored on a computer, the system will have fully patched operating systems and applications, and current antivirus software with current virus definitions. Information may be stored in cloud-based servers.	☐	☐	
2	If not, provide justification:			☐
3	Will the following security **recommendation** be met: • A plan for routine back-ups of all data will be in place	☐	☐	
4	Outline any additional safeguards that will be taken:			☐
SECTION 34.2 - Level 2		Yes	No	N/A
1	Will the following security **requirements** be met: • Information will be shared and stored in a manner that provides access only to authorized individuals. • Data will not be disclosed to additional parties without prior IRB approval specifically authorizing the	☒	☐	

	disclosure. • If information is stored on a computer, the system will have fully patched operating systems and applications, and current antivirus software with current virus definitions. • Information may be stored on cloud servers licensed by OSU. A plan for routine back-ups will be in place. • **If subjects will be asked to use a third-party website or application, or if a cloud-based server will be used, the researchers will review the data security plan with the Information Security Office before initiating the study. Exceptions include servers and software licensed by OSU.**			
2	If not, provide justification:			☐
				
3	Will the following security **recommendation** be met: • Computers will have firewalls in place.	☒	☐	
4	Outline any additional safeguards that will be taken:			☐
				
SECTION 34.3 - Level 3		Yes	No	N/A
1	Will the following security **requirements** be met: • Information will be shared and stored in a manner that provides access only to authorized individuals. • Data will not be disclosed to additional parties without prior IRB approval specifically authorizing the disclosure. • If information is stored on a computer, the system will have fully patched operating systems and applications, and current antivirus software with current virus definitions. • When feasible, information will be stored in a local system of record (e.g., local server, approved cloud). • All mobile computer systems or portable storage media will be encrypted with at least the 256-bit encryption common in operating systems and encoding devices sold in the United States. • If the data are coded, and there is a linked list of codes and identifiers, this list will be stored separately from all coded data. • Identifiable information will not be stored on student researchers' computers after the study has ended, unless justified elsewhere in this section and approved by the IRB. • Computers must have host-based firewalls enabled in addition to being behind a networked firewall context. • A plan for routine back-ups of all data must be in place, with the appropriate security mechanisms for	☐	☐	

	that data, including encryption and physical security addressed. • **The researchers will review the data security plan with the Information Security Office before initiating the study.**			
2	If not, provide justification:			
				
3	Outline any additional safeguards that will be taken:			☐
				
	Researchers using cloud-based servers, or third party websites or applications, that are not licensed by OSU, and those collecting level 3 data can minimize delays by consulting with the Information Security Office prior to submitting the application materials to the HRPP for review.			
SECTION 35 - Mandatory Reporting		Yes	No	N/A
	Under Oregon state law, all OSU employees are mandatory reporters.			
	Reporting requirements related to child abuse or neglect.			
	Reporting requirements related to sexual assault or misconduct			
	What is the impact of mandatory reporting legislation on IRB-approved research?			
	What do I do if a research participant tells me about an experience with sexual harassment or sexual violence?			
1	Study includes collection of information regarding **child abuse or neglect** OR it is reasonable to expect that child abuse or neglect could be observed or revealed to the researchers	☐	☒	
2	Study includes collection of information regarding **sexual harassment or sexual violence** OR it is reasonable to expect that such information could be revealed to the researchers	☐	☒	
3	Study includes collection of information regarding **harm to self or others** OR it is reasonable to expect that such information could be observed or revealed to the researchers	☐	☒	
4	Researchers plan to report information regarding harm to self or others and this is disclosed to potential participants as part of the consent process	☐	☐	☒
5	If yes to any of the above items in this section, describe the relevant training that study team members have, or will receive, to minimize risks to participants and comply with the reporting requirements**:**			☒
				

SECTION 36 - Certificate of Confidentiality		Yes	No	N/A
	Certificate of Confidentiality			
1	A Certificate of Confidentiality has been automatically deemed issued because this study is NIH-funded and includes individually identifiable data	☒	☐	☐
2	A Certificate of Confidentiality from the NIH has been obtained or will be sought for this study because it includes the collection of individually identifiable, "sensitive" data	☐	☐	☒
SECTION 37 - Risks		Yes	No	N/A
	Minimal risk means that the probability **and** magnitude of harm or discomfort anticipated in the research are not greater in and of themselves than those ordinarily encountered in daily life or during the performance of routine physical or psychological examinations or tests.			
	It is not sufficient to describe the risks as "minimal" without identifying what those risks are. Include risks of potential harm that are physical, mental/emotional, legal, financial, insurance, employment, or social and reputational risks.			
	In all cases where participants are known to the investigators, there is the chance of a breach of confidentiality. However, if there is no potential for harm associated with such a breach, it need not be listed in this section. The ways in which the potential for a breach of confidentiality will be minimized should be articulated in the anonymity and confidentiality section.			
	Investigators should consider risks to entire groups under study (e.g., tribes, ethnic or racial groups, economic classes).			
1	Does the study involve greater than minimal risk to adult participants?	☒	☐	
2	If children will be enrolled, which of the following four federal categories applies (check "yes" only once for 2a – 2d):			☒
2a	Research does not involve greater than minimal risk to children.	☐	☐	☒
2b	Research involves more than minimal risk to children but the study holds prospect of direct benefit to the participants.	☐	☐	☒
2c	Research involves more than minimal risk to children and there is no prospect of direct benefit to participants, but the study likely to yield generalizable knowledge about the participants' disorder or condition.	☐	☐	☒
2d	Research is not otherwise approvable under the federal regulations but the study presents an opportunity to understand, prevent, or alleviate a serious problem affecting the health or welfare of the participants.	☐	☐	☒
3	Describe all reasonably foreseeable risks to study participants:			☐

	**BaP:** The International Agency for Research on Cancer (IARC) has determined that BaP is a Class 1 known human carcinogen. **Radiation:** The total radiation dose of 16.4 nCi for the 3 dose cycles of [^{14}C]-benzo[a]pyrene (10.8 nCi if only doing 2 dose cycles) is equivalent to about 60 minutes of natural background radiation. The risk from the ultra-low doses in this study is negligible. **Blood Sampling:** Risks of blood sampling include pain, bruising, and in rare instances, infection. Some individuals can become lightheaded or nauseated. **Blood Loss:** The total amount of blood collected from each subject is 420 mL (120 mL for the [14C]-BaP only cycle and 155 each for the Brussels sprouts and DIM cycles) or 310 mL if only doing the Brussels sprouts and DIM cycles. The maximum amount of blood collected in an 8 week period is 310 mL. **Food allergens:** Foods and beverages provided during this study may contain allergens. **Foodborne illness:** As with any perishable foods, the Brussels sprouts may spoil if improperly stored or may become contaminated. **Confidentiality/anonymity:** Loss of confidentiality or anonymity is a potential risk.			
4	Describe all steps taken to minimize risks:			
	**BaP:** Humans are exposed to PAHs, including BaP, from a number of sources, the highest being occupational and smoking, so we are excluding these populations. In the general population, the greatest BaP exposure is through diet (9.5-43.5 ng/day inhalation; 1 ng/day water; 160-1,600 ng/day diet). The dietary restrictions during this study are intended to offset the total 100 ng BaP consumed in 2 study cycles. This is equivalent to less than or equal to 4 ng/day over the 25 day (minimum) study period. We estimate that following our dietary restrictions will reduce subject daily dietary exposure to BaP by 35-54 ng/day. Therefore, subjects are not exposed to an increased risk of cancer. **Radiation:** The risk from the ultra-low doses in this study is negligible. **Blood Sampling:** A qualified research nurse or certified phlebotomist will perform blood draws and carry out all necessary precautions to reduce the risk of injury to subjects. Subjects will be encouraged to hydrate with 1-2 glasses of water before blood draws to minimize risk of syncope or lightheadedness. **Blood Loss:** While the risk of negative impact due to blood loss is negligible, subjects will be asked to refrain from blood donation one month before the study until one month after completion of the final			

	cycle. **Food Allergens:** Ingredient lists and packaged foods with clearly labeled allergen information will be provided to subjects upon request. Subjects will be asked to notify the study nurse of any food allergies before ordering breakfast. **Foodborne illness:** Subjects will be instructed to store Brussels sprouts frozen and only thaw immediately before consumption. Study team members handling the Brussels sprouts will have Oregon Food Handler's Licenses, and the study Brussels sprouts will be repackaged in a facility compliant with Oregon Health Authority, Division 150, Food Sanitation Rule. **Confidentiality/anonymity:** Protection efforts are described in the *Privacy and confidentiality* section			
	Consider whether it is appropriate to provide participants with contact information for one or more resources during the recruitment or consent process (e.g., CAPS if OSU students, EAP if OSU employees, suicide prevention hotlines, address of local shelters, etc.).			
5	If the study is greater than minimal risk, attach a Data and Safety Monitoring Plan or provide the following information:			☐ *Skip to section 38*
5a	Definition of an adverse event for this study:			
	DSMP is attached			
5b	Definition of a serious adverse event for this study:			
5c	Provide a plan for managing adverse events:			
5d	Summarize any reporting requirements and timelines:			
5e	Indicate if there are any individual or overall study stopping rules:			
5f	Provide the plan for reporting adverse events or unanticipated problems (e.g., breach of confidentiality, incarcerated participant, or an unresolved complaint):			
5g	If the potential harm related to physical, psychological, or financial risks is greater than minimal, describe			

	the plan for paying for related costs to participants:			
				
	NIH guidance on data and safety monitoring			
SECTION 38 - Benefits		Yes	No	N/A
1	Describe benefits to the individual participants, to society, and to science:			
	This study is not designed to be of benefit to individual subjects. Potential benefits to society include essential information to aid regulatory agencies with respect to how environmental PAHs, such as BaP, at environmentally relevant levels of exposure are taken up by the G.I. (96% of carcinogenic PAH exposure in humans is dietary), metabolized and excreted from the body. That information can be used in modeling risk assessment rather than high-dose animal studies. This will provide a mechanism for improvement of public health.			
SECTION 39 - Training and Oversight		Yes	No	N/A
	CITI training is required and should not be included in this section. This section pertains to all other training required to conduct the study, such as training study team members to collect data or samples.			
1	Is the PI the only member of the study team?	☐ *Skip to section 40*	☒	
2	Describe the plan for confirming or providing training related specifically to the study activities and for supervising all study team members:			
	Williams is responsible for the conduct and oversight of the study, for ensuring that privacy/confidentiality of subjects is maintained and that all individuals working on the study are properly trained to perform their role on the study.			
3	Describe the plan for training related specifically to obtaining informed consent and maintaining confidentiality:			
	Nurse coordinator Uesugi will obtain informed consent from study subjects. She has completed a clinical research coordinator (CRC) training course and will be responsible for maintaining confidentiality.			
4	Explain how oversight of study team members will be handled during PI absences (sabbaticals, non-contract months, etc.):			
	Extended absence of the PI is not anticipated because the PI will not use sabbatical leave during the study, is appointed at 1.0 FTE (9-month) and a tenured faculty member. A certified phlebotomist may occasionally be employed to collect blood samples as a back-up to Uesugi.			

SECTION 40 - Principal Investigator's Assurance Statements		Yes	No	N/A
	I understand Oregon State University's policies concerning research involving human participants and I attest:			
	• that the information contained in this document is accurate and complete	☒		
	• that research activities will not begin until an approval or acknowledgement has been issued	☒		
	• to the scientific merit and importance of this study	☒		
	• to the competency and availability of the study team member(s) to conduct the project	☒		
	• that facilities, equipment, and personnel are adequate to conduct the research	☒		
	Furthermore, I agree to:			
	• comply with all HRPP and IRB policies, decisions, conditions, and requirements	☒		
	• accept responsibility for every aspect of the conduct of this study	☒		
	• adhere to all aspects of the protocol, once approved	☒		
	• obtain approval prior to amending or altering the study, when required by institutional policy	☒		
	• report in accord with institutional policy, any adverse event(s) and/or unanticipated problem(s)	☒		
	• inform the HRPP if PI or another member of the study team leaves OSU or otherwise changes institutional affiliations	☒		
	• notify the HRPP office immediately of the development of any potential conflict of interest not already disclosed and, when applicable, report to an external IRB	☒		
	• accept and fulfill all expectations and responsibilities required by the FDA if the study is regulated by 21 CFR 312 (drugs) or 812 (devices)	☒		☐
	I understand that if OSU has deferred oversight to an external IRB, I am responsible for ensuring that the content of the OSU file matches the external IRB's file within 30 days of any approvals, changes, or other actions	☐		☒

SECTION 41 - Attachments
Check all that apply:
☐ Assent document(s) or guide(s)
☒ Certificate of Analysis or other documentation of quality/purity
☐ Certificate of Confidentiality
☐ CLIA certificate
☒ Consent document(s) or guide(s)
☐ Copy of curriculum if the study intervention is a workshop or a class
☐ Data and Safety Monitoring Board Charter or Report

APPLICATION & PROTOCOL

☒ Data and Safety Monitoring Plan
☐ Department of Corrections application
☐ Documentation that the product is an approved food additive (21 CFR 172)
☐ External IRB document(s)
☐ External study document(s)
☐ FDA Correspondence
☐ FDA “Safe to Proceed” letter
☐ GMP certificate or food-grade certificate that indicates that the product can be lawfully used in the US food supply
☐ Grant or contract if not already in Cayuse
☐ IND or IDE Application for Investigational New Drug or Device
☐ Investigator brochure
☒ Label for investigational drug or device
☐ Letter from the FDA or industry sponsor setting forth the IND number
☐ Letter(s) of support or permission
☐ Package insert
☒ Recruitment materials
☒ References or citations
☐ Safety data if GRAS status is self-affirmed
☒ Screening or eligibility checklist(s) or document(s)
☐ Surveys, questionnaires, interview or focus group guide(s)
☐ Translated document(s)
☐ Tribal Council Resolution or other appropriate supporting documentation from the appropriate Tribal authority
Additional attachments:
☒ Food diary
☒Demographics form
☒Cruciferous vegetable list
☒Health assessment form
☒Cruciferous vegetable food frequency questionnaire (FFQ)
☒BioResponse DIM 150 label
☒Brussels sprouts preparation instructions
☐
☐

APPLICATION & PROTOCOL

☐

PI should email completed application and all relevant attachments to IRB@oregonstate.edu

Statistical Analysis Plan

Pharmcokinetic parameters were evaluated for linearity using a best fit modeling approach. First, pharmacokinetic parameters were evaluated for significant change as a functin of dose using a standard linear regression model including a fit y-intercept. Slopes were compared using a *t*-test and an alpha value of 0.05. If the parameter changed as a function of dose, we further evaluated the parameter with a linear regression model, with *k* as the slope through the origin and a Michaelis-Menten model, which assumes saturation at V_{max} and an affinity constant K, to individual parameters (p) as a function of extermal [^{14}C]-BaP dose. The Baysesian information criterion (BIC) was used to judge the best-fit model and provide evidence of linear or saturable pharmacokinetics.

Oregon State University

RESEARCH CONSENT FORM

Study Title: Ultralow Dose PAH Binary Mixture Study
Principal Investigator: David E. Williams, PhD
Study team: Douglas Aukerman, MD, Sandra Uesugi, RN, Lisbeth Siddens, Jamie Pennington
Sponsor: National Institutes of Health, National Institute of Environmental Health Sciences
Version: August 31, 2018

SUMMARY

We are inviting you to take part in a research study. You do not have to be in the study if you do not want to. You can also decide to be in the study now and change your mind later.

This study is about a common pollutant called benzo[*a*]pyrene or BaP. The purpose of this research study is to better understand how our bodies absorb and eliminate BaP.

If you take part in this study, we will ask you to complete **two 48-hour study cycles**. You will be asked to provide blood and urine samples. During one cycle, you will receive a dose of **BaP alone**, and in the second cycle, you will receive a dose of **BaP with phenanthrene**, another common pollutant related to BaP.

The most serious risks related to being in this study are ultralow dose exposure to BaP, a known carcinogen, and potential bruising or discomfort from the blood draws.

We plan to make the results of this study public, but we will not include your name.

We would like you to ask us questions if there is anything about the study that you do not understand. You can contact the Nurse Coordinator Sandra Uesugi, RN at 541-737-3594 or sandra.uesugi@oregonstate.edu or the Principal Investigator Dr. David Williams at 541-737-3277 or david.williams@oregonstate.edu.

You can also contact the Human Research Protection Program with any concerns that you have about your rights or welfare as a study participant. This office can be reached at (541) 737-8008 or by email at IRB@oregonstate.edu .

There are more details about the study in the following pages.

Oregon State University

STUDY DETAILS

1. WHY AM I BEING INVITED TO TAKE PART IN THIS STUDY?

You are being invited to take part in this study because you are a healthy non-smoking adult aged 21-65. If you are female, you must be post-menopausal or surgically sterile.

2. WHAT WILL HAPPEN IF I TAKE PART IN THIS RESEARCH STUDY?

This research study involves a screening visit and two study cycles. We will ask you to take a 50 ng dose of BaP during each cycle with and without 1250 ng phenanthrene, another PAH commonly found in smoked foods. All the visits will take place in the Clinical Research Center (CRC) - Room 407 in the Linus Pauling Science Center at OSU.

Screening visit (60 minutes)

We will review the study activities, schedule and diet restrictions, and answer any of your questions before obtaining your written consent. We will collect demographic information, health history, height, weight, blood pressure and heart rate. If you are female, you will be asked to provide a urine sample for a pregnancy test. The study physician will perform a physical exam and review your health history. Fasting is not required for this visit.

If you were eligible for our previous *Benzo[a]pyrene Ultralow Dose-Response Study* (IRB Study # 8233), you may not need another physical exam if you have not had any significant changes to your health or medication. All other screening activities will be conducted.

Two study cycles (48 hours, 4 visits per cycle)*:

For each study cycle, we will ask you to participate in these study activities:

Food Diaries: We will ask you to record all food and beverages that you consume during the 3-days before each study cycle and during the 48-hour study cycle.

Diet restrictions: We will ask you to follow these restrictions for 2 weeks before and through the end of each cycle (16 days):

1. Avoid eating any smoked meats and cheeses
2. Avoid eating any charcoal grilled meats (gas-grilled meat is ok)
3. Avoid eating cruciferous vegetables and condiments (see list)
4. Avoid taking any supplements that contain indole-3-carbinol (I3C) or 3,3'-diindolylmethane (DIM)

Overnight Fast: We will ask you to not eat or drink anything besides water for at least 8 hours before the first morning of each study cycle. We will provide breakfast on the first morning of each cycle, 2 hours after you swallow the BaP capsule. You will be able to order your own breakfast food and beverage from a menu, and ingredient information can be provided if you have any food allergies or dietary restrictions.

Oregon State University

Study visits:

Visit 1 - (0-4-hour time points, 4.5 hours): On the first morning of each cycle, we will ask you to provide a urine sample and to empty your bladder. If you are female, we will also do a urine pregnancy test at the start of each study cycle. The study nurse will measure your weight and then place an IV catheter in a vein in your inner elbow.

We will draw a baseline blood sample and then provide a BaP or BaP + phenanthrene capsule for you to swallow with water (time 0 hours). The study nurse will draw a blood sample at 0.25 0.5, 1.0, 1.5, 2, 3, 4 hours from the IV catheter. You will have the choice to keep the IV catheter in place until the 8 hour blood draw or have it removed after 4 hours. You will need to remain near the CRC if you choose to keep the IV catheter until the 8-hour time point.

Visit 2 – (8 hour time point, 15 minutes): If you chose to have the IV catheter removed after 4 hours, you can leave the building. We will ask you to return to the CRC at the 8-hour time point for a straight needle stick blood draw.

Visit 3 – (24-hour time point, 15 minutes): We will ask you to return to the CRC at the 24-hour time point for a straight needle stick blood draw.

Visit 4 – (48-hour time point, 15 minutes): We will ask you to return to the CRC at the 48-hour time point for a straight needle stick blood draw.

We will collect a total of 120 mL (8 Tbsp.) of blood in each cycle.

Urine collection: We will ask you to collect all of your urine for 48 hours in containers that we provide. We will also provide a discrete soft-sided cooler bag for transportation. You can store any collected urine samples at room temperature in the bag until you return for your next visit. We will ask you to return any filled containers at the next visit until the end of the cycle. The longest you will need to store any samples is 24 hours between Visits 3 and 4.

Washout period: We will wait at least 3 weeks between study cycles to allow your body to completely eliminate each BaP dose.

***Participants of Ultralow Dose-Response Study (IRB protocol 8233):** If you completed the 50 ng BaP dose cycle within the past 12 months, you will only participate in the BaP plus phenanthrene dose cycle in this study to minimize your risks.

Storage and future use of data and samples: We may indefinitely store a portion of your blood and urine for possible future studies. The samples will be coded with no identifying personal information. Because it is not possible for us to know what studies may be a part of our future work, we ask that you give us permission now to use your samples and data without being contacted about each future study. Future use of your samples will be limited to studies about health effects of pollution. We will not pay you for the use of your sample or data. If you agree now to future use of your samples but decide in the future that you would like to have them removed from research tests, please contact Dr. David E. Williams, Oregon State University, 473 Linus Pauling Science Center, Corvallis, OR 97331, 541-737-3277.

We will be destroying all identifying information when data collection is complete. Once the identifying information is destroyed, we will not be able to remove your information from the larger dataset.

_____You may store my information and/or samples for use in future studies.
Initials

_____You may **not** store my information and/or samples for use in future studies.
Initials

During this study some of your blood will be used to study 1-7 specific genes. A gene is the code (DNA) present in each cell in your body and controls the behavior of that cell. We are interested in studying the genes that control the way BaP is handled in the body. This will help us understand if these genes increase or decrease the uptake, metabolism or excretion of BaP.

_____You may use my samples for gene analysis.
Initials
_____You may **not** use my samples for gene analysis.
Initials

Future contact: We may contact you in the future for another similar study. You can ask us to stop contacting you at any time.

Study Results: We will share any published results of the study with you if you request.

3. WHAT ARE THE RISKS AND POSSIBLE DISCOMFORTS OF THIS STUDY?

BaP: The International Agency for Research on Cancer (IARC) has determined that BaP is a Class 1 known human carcinogen. The amount of BaP you will take in this study is extremely small. It is less than you may eat already every day in your diet, especially in cooked meat. For example, one grilled hamburger could contain as much or more PAHs than the amount you will take in this study. The diet restrictions are designed to help reduce additional exposure to BaP and other PAHs during the study cycles, and your cancer risk is not increased by participating in this study. The US EPA and IARC have evaluated phenanthrene and determined it not to be a carcinogen.

Radiation: In order to track the BaP in your blood and urine samples, it contains a carbon-14 label. Carbon-14 emits very low levels of radiation. The total amount of radiation that you will receive in the 2 cycles is equivalent to 40 minutes of natural background radiation. At this level, the risks associated with radiation exposure are negligible and no higher than your everyday exposure.

Blood Sampling: The risks of having blood drawn from your arm include some pain when the needle goes in and a small risk of bruising, inflammation or infection at that site. Please alert the study nurse if you notice any symptoms during or after each study cycle.

Some people get lightheaded, nauseous, or faint. You are less likely to have these problems if you drink 1-2 glasses of water in the evening and morning before your study visits.

Oregon State University

The American Red Cross recommends that you do not donate more than 1 pint (32 tablespoons) of blood within a 2-month period. We request that you do not donate blood for at least one month after completing the final study cycle.

Food allergens: We will provide breakfast on the first day of each cycle and will provide clearly labeled packaged foods or ingredient lists if requested. Please notify the study nurse if you have any food allergies or diet restrictions.

Confidentiality and Privacy: There is a risk that we could accidentally disclose information that identifies you.

4. WHAT ARE THE BENEFITS OF THIS STUDY?

This study is not designed to benefit you directly. This study may help scientists and environmental regulatory agencies better understand the health effects of PAHs. Your participation will contribute to our scientific body of knowledge for risk assessment of an important group of environmental contaminants.

5. WHAT OTHER OPTIONS DO I HAVE IF I DO NOT TAKE PART IN THIS STUDY?

Participation in this study is voluntary. If you qualify for the study, you may choose to not participate. There are no alternative study activities if you choose to not participate.

6. WHAT SHOULD I DO IF I WANT TO STOP BEING IN THE STUDY?

If you decide to participate, you are free to withdraw at any time without penalty. If you choose to withdraw from this project before it ends, the researchers may keep samples and information collected about you, and this information may be included in study reports.

7. WHO WILL SEE THE INFORMATION I GIVE?

The information you provide during this research study will be kept confidential to the extent permitted by law. Research records will be stored securely. Regulatory agencies, the Food and Drug Administration, the National Institute of Environmental Health Sciences and Oregon State University employees may access or inspect records pertaining to this research as part of routine oversight or university business. Some of these records could contain information that personally identifies you.

Some of your coded blood samples will be sent to outside laboratories for analysis. Outside laboratories will only have samples identified by code and will not have access to the key connecting your name to the code.

If we contacted you through the Center for Healthy Aging Research (CHAR) LIFE Registry, we will be providing CHAR with any updates to your contact information. We will also tell them whether or not you chose to participate in this research study.

A description of this clinical trial will be available on http://www.ClinicalTrials.gov, as required by U.S. Law. This website will not include information that can identify you. At most, the website will include a summary of the results. You can search this website at any time.

Most people outside of the study team will not see research information that includes your name. This includes people who try to get your information using a court order. We could give out this information if you gave us permission.

8. WHAT HAPPENS IF I AM INJURED?

Oregon State University has no program to pay for research-related injuries. If you think that you have been injured as a result of being in this study, please contact the study team immediately.

9. WILL I BE PAID FOR BEING IN THIS STUDY?

You receive $125 for each study cycle. The total amount you will receive for completing 2 cycles is $250 or $125 if you only complete 1 cycle. If you withdraw early from the study, your payment will be prorated to the proportion of blood samples provided. For example, if you complete 9 out of 11 blood samples in a cycle you will receive $102.27 ($11.36/sample).

10. WILL IT COST ME ANYTHING TO BE IN THIS STUDY?

You are responsible for transportation to the OSU campus. We will provide convenient free parking during your study visits.

11. WHAT WILL HAPPEN IF THE RESEARCHERS THINK THAT I SHOULD NO LONGER BE IN THE STUDY?

We may take you off the study early if you do not follow study instructions, if the investigator stops the study, or if you develop serious side effects.

---

12. WHAT DOES MY SIGNATURE ON THIS CONSENT FORM MEAN?

Your signature indicates that you acknowledge that this study has been explained to you, that your questions have been answered, and that you agree to take part in this study. You will receive a copy of this form.

Participant Name:_____

Participant Signature: _____

Date Signed:_____

Name of Person Obtaining Consent:_____

Signature of Person Obtaining Consent:_____

Date Signed:_____