EPA Review of i2LResearch USA, Inc. Protocol

Proposal for Laboratory Evaluation of Mosquito Bite Protection from Permethrin-Treated Clothing for the U.S. Army

Tim Ciarlo
Maureen Lydon
Office of Pesticide Programs

Overview

- Protocol for a laboratory study of the mosquito bite protection afforded by military uniforms containing 0.5% permethrin
- Submitted by i2LResearch USA, Inc.
- Protocol is for a special study, non-EPA guideline, that is similar to a previous study reviewed in April 2014 by the HSRB involving etofenprox-treated military clothing
- Research is proposed to satisfy EPA registration requirements for efficacy

Overview 2

- Sponsor will test the hypothesis that permethrin treatment provides bite protection when mosquitoes are exposed to treated fabric compared to an untreated control. Sponsor's target level of mean bite protection is ≥90%.

- Permethrin is recommended by the World Health Organization for use in public health vector control programs as a direct spray to infested areas or indirectly by treating fabrics, such as mosquito nets

Differences from Skin-Applied Repellent Studies Reviewed by the HSRB

- Field study versus laboratory
- Different "repellent" effect
- Different efficacy measures
  - CPT vs Mean Bite Protection
- Subjects will receive mosquito bites

Science Assessment: Proposal for Laboratory Evaluation of Mosquito Bite Protection from Permethrin-Treated Clothing for the U.S. Army

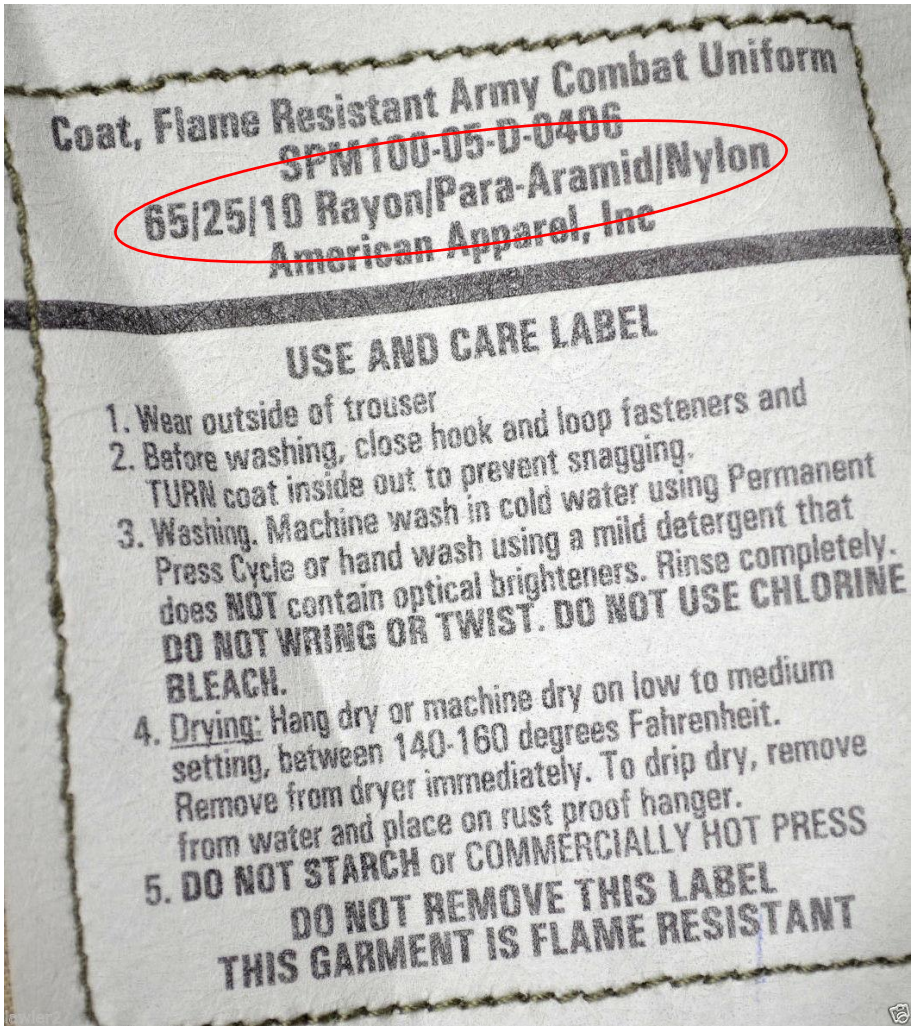
Tim Ciarlo
Registration Division
Office of Pesticide Programs

Study Objectives

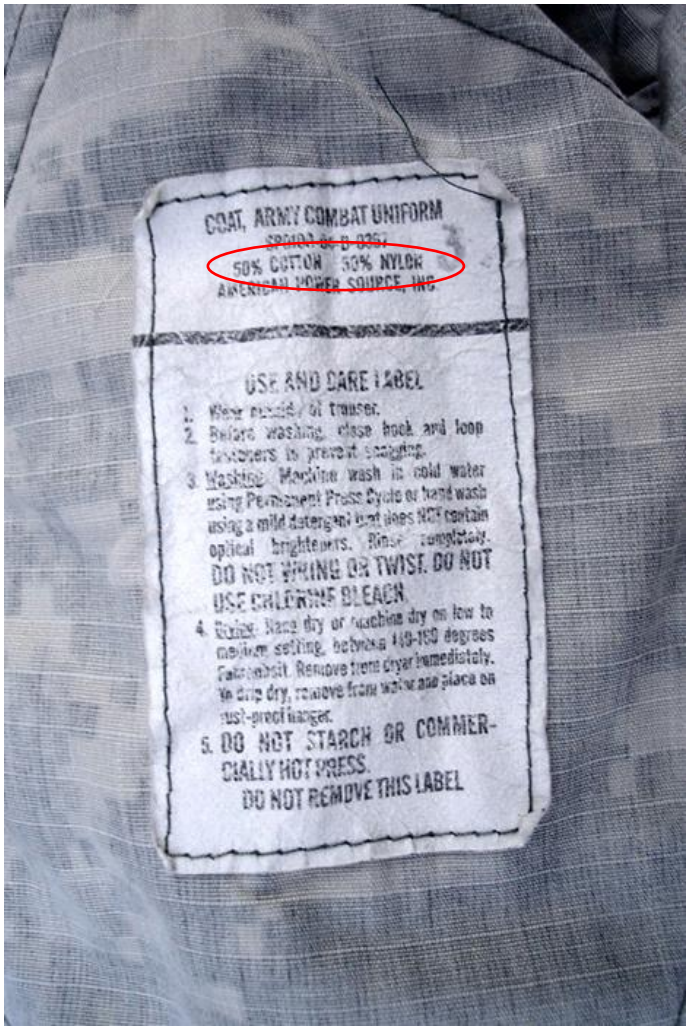
- This study is designed to determine the bite protection level of permethrin-treated U.S. Military Flame Resistant Army Combat Uniforms (FRACUs) and Army Combat Uniforms (ACUs) treated initially at an application rate of 0.5% wt/wt, and to assess the bite protection performance after 0, 20, and/or 50 washes.

Study Objectives 2

- FRACU



- ACU



Study Objectives 3

- The purpose of this research is to determine whether permethrin-treated FRACUs and/or ACUs meet the EPA specifications for minimum bite protection level.
  - ≥90% at each fabric treatment level
- The research has societal value because U.S. military personnel serving domestically and abroad are at risk of contracting insect-transmitted diseases.

Acute Toxicity of the Test Material

- Acute Dermal = LD_{50} >2,000 mg/kg body weight
- Acute oral = LD_{50} >2,280 mg/kg body weight
- Low irritant to eyes
- Minimal irritant to skin
- Not a skin sensitizer

MOE Estimate
Dermal NOAEL = 500 mg/kg/day

- Estimated maximum amount of permethrin in 6 treated sleeves = 696 mg/subject
- Assuming 15% permethrin transfer to skin, each subject can receive up to 104 mg in one day (696*0.15 = 104)
- Assuming 70 kg subject, estimated human exposure (EHE) is 104/70 = 1.49 mg/kg
- Margin of Exposure (MOE) = NOAEL/EHE = 500/1.49 = **335**
- EPA's Level of Concern of MOE = 100

Experimental Design

Testing Paradigm for Ae. aegypti

	Subject Right Arm		Subject Left Arm	
Test Set	**Treatment Condition**	**Specimen Designation**	**Treatment Condition**	**Specimen Designation**
1	FRACU Untreated Unwashed Control	Sleeve 1	ACU Untreated Unwashed Control	Sleeve 2
2	FRACU Treated Washed 50x	Sleeve 3	ACU Treated Washed 50x	Sleeve 4
3	FRACU Treated Washed 20x	Sleeve 5	ACU Treated Washed 20x	Sleeve 6
4	FRACU Treated Unwashed (0x)	Sleeve 7	ACU Treated Unwashed (0x)	Sleeve 8

Testing Paradigm for An. quadrimaculatus

	Subject Right Arm		Subject Left Arm	
Test Set	**Treatment Condition**	**Specimen Designation**	**Treatment Condition**	**Specimen Designation**
5	FRACU Untreated Unwashed Control	Sleeve 9	ACU Untreated Unwashed Control	Sleeve 10
6	FRACU Treated Washed 50x	Sleeve 11	ACU Treated Washed 50x	Sleeve 12
7	FRACU Treated Washed 20x	Sleeve 13	ACU Treated Washed 20x	Sleeve 14
8	FRACU Treated Unwashed (0x)	Sleeve 15	ACU Treated Unwashed (0x)	Sleeve 16

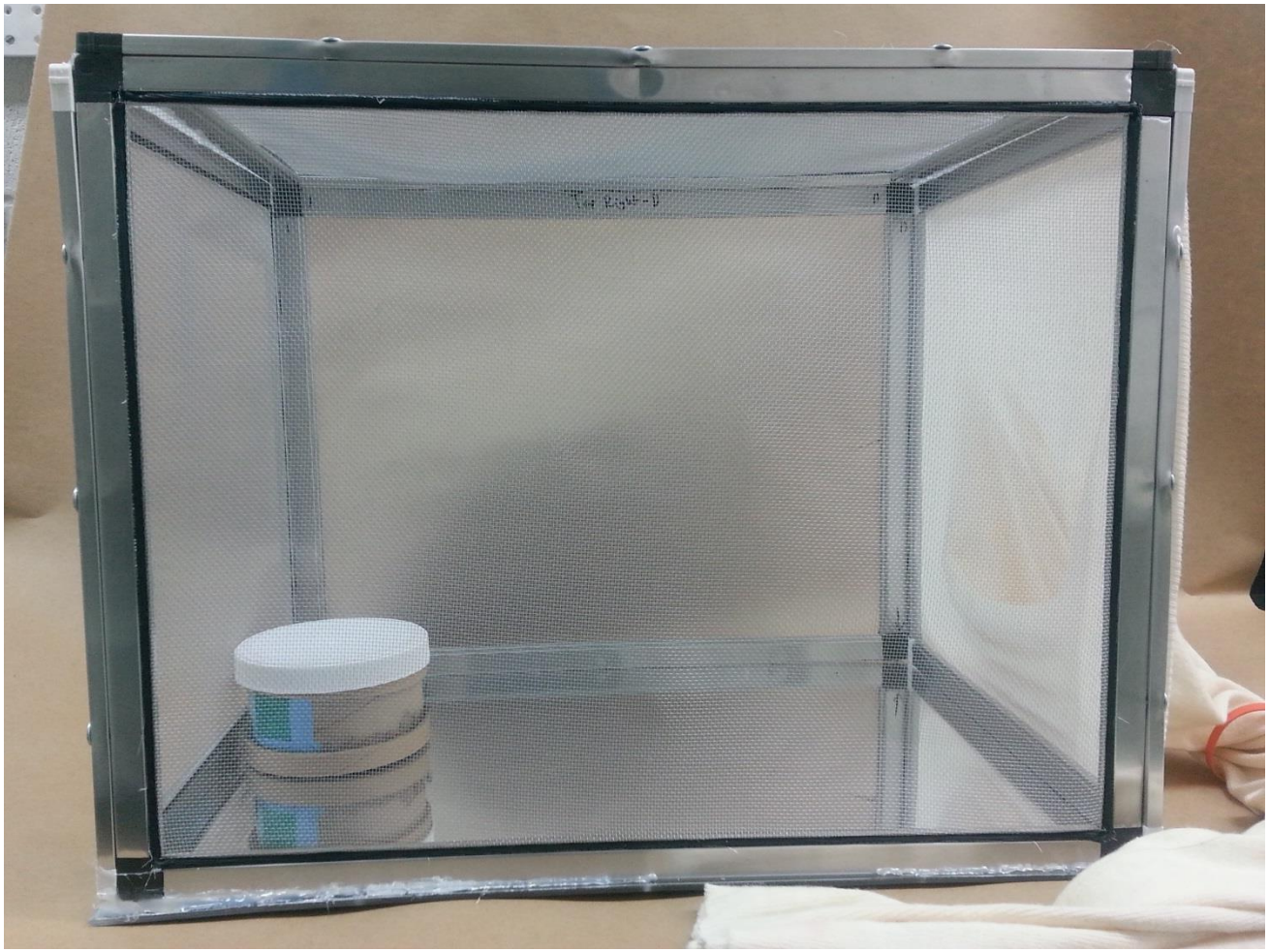
Experimental Design 2
Difference in Number of Test Subjects

Fabric and Treatment Condition	Number of Fabric Specimens	Number of Subjects	Number of Species	Total Replicates per Fabric Type
FRACU Untreated Unwashed Control	1	10	2	20
FRACU Treated Washed 50x	1	10	2	20
FRACU Treated Washed 20x	1	10	2	20
FRACU Treated Unwashed (0x)	1	10	2	20
ACU Untreated Unwashed Control	1	15	2	30
ACU Treated Washed 50x	1	15	2	30
ACU Treated Washed 20x	1	15	2	30
ACU Treated Unwashed (0x)	1	15	2	30

Experimental Design 3

- The test cages are approximately 59,000 cm^3 in volume and each will contain 175 to 225 female mosquitoes (density of ~1 mosquito/300 cm^3)
- Female mosquitoes will be preselected from stock cages. A technician will place an ungloved hand near the screened stock cage to attract mosquitoes and will then use a motorized aspirator to transfer them to test cages

Experimental Design 4
Test Cage



Endpoints and Measures

- Unit of measure for determination of efficacy is percent bite protection
- Presence of blood in the mosquito's abdomen will confirm a 'mosquito bite'
- For each test set, the treatment % bite values will be corrected to account for the bite through values in the untreated control using Abbott's Formula

Endpoints and Measures 2

- Percent bloodfed in untreated control treatment after test interval
- Percent bloodfed in permethrin treatment after the test interval
- Test interval = 15 minutes

Endpoints and Measures 3

- Percent Bite Protection =

  [1 – (treatment rate) / (control rate)] × 100%

- Treatment rate (or proportion) =
  (# bloodfed female mosquitoes after test interval) /
  (total # of female mosquitoes in test cage)
  ~~when subject used TREATED fabric~~

- Control rate (or proportion) =
  (# bloodfed female mosquitoes after test interval) /
  (total # of female mosquitoes in test cage)
  ~~when subject used UNTREATED fabric~~

Statistical Analysis Plan

- A protocol deriving a sample size for an etofenprox-treated uniform was originally presented to the HSRB in April 2014
  - April 2014 protocol presented with objective of meeting military bite protection specification of 85%/80%/70%
- EPA derived the number of subjects for this current permethrin study by simulation after modifying the (original) SAS code that was used for the earlier HSRB
  - PROC GLIMMIX (subject as random effect) rather than PROC GENMOD (subject as fixed effect)
  - Expected bite-through rate in untreated material
  - Additional sensitivity analyses (detailed in science review)

Statistical Analysis Plan

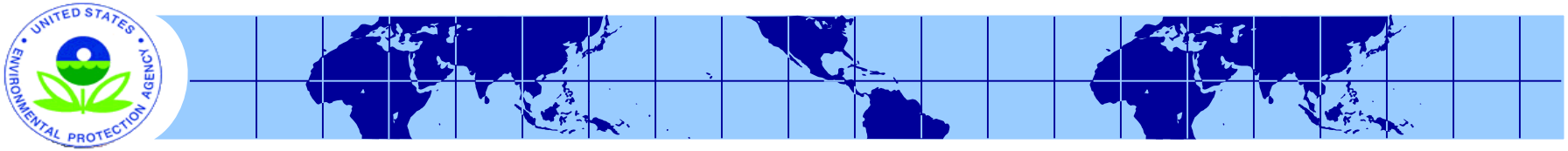
- For current protocol, two bite protection objectives considered:
  - Military percent bite protection specification of 85%/80%/70% for 0-, 20-, and 50-washes
  - EPA percent bite protection specification of 90% for 0-, 20-, and 50-washes.

Statistical Analysis Plan 3

Overall, EPA considered:

- the estimated precision of the simulated sample sizes,
- the sensitivity of the sample size estimates to the parameter inputs, and
- the “value-added” of additional subjects’ participation in reducing the half-width of the 95% confidence interval

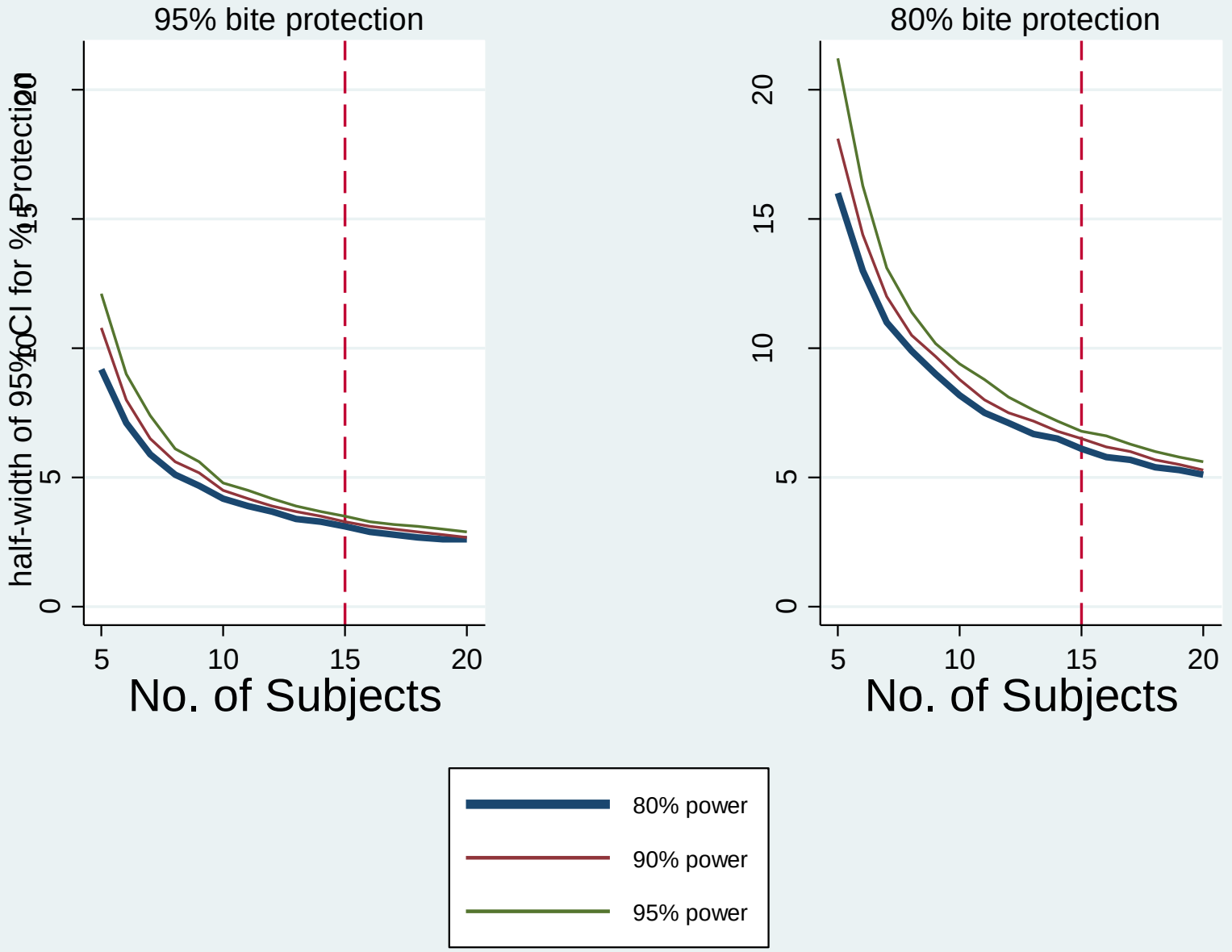
…in arriving at what it judged to be an appropriate sample size that helps to ensure the scientific integrity of the proposed research



Power Analysis to Evaluate Effect of Sample Size on Precision of Bite Protection For ACU and FRACU Uniforms, (as measured by half-width of 95% CI)

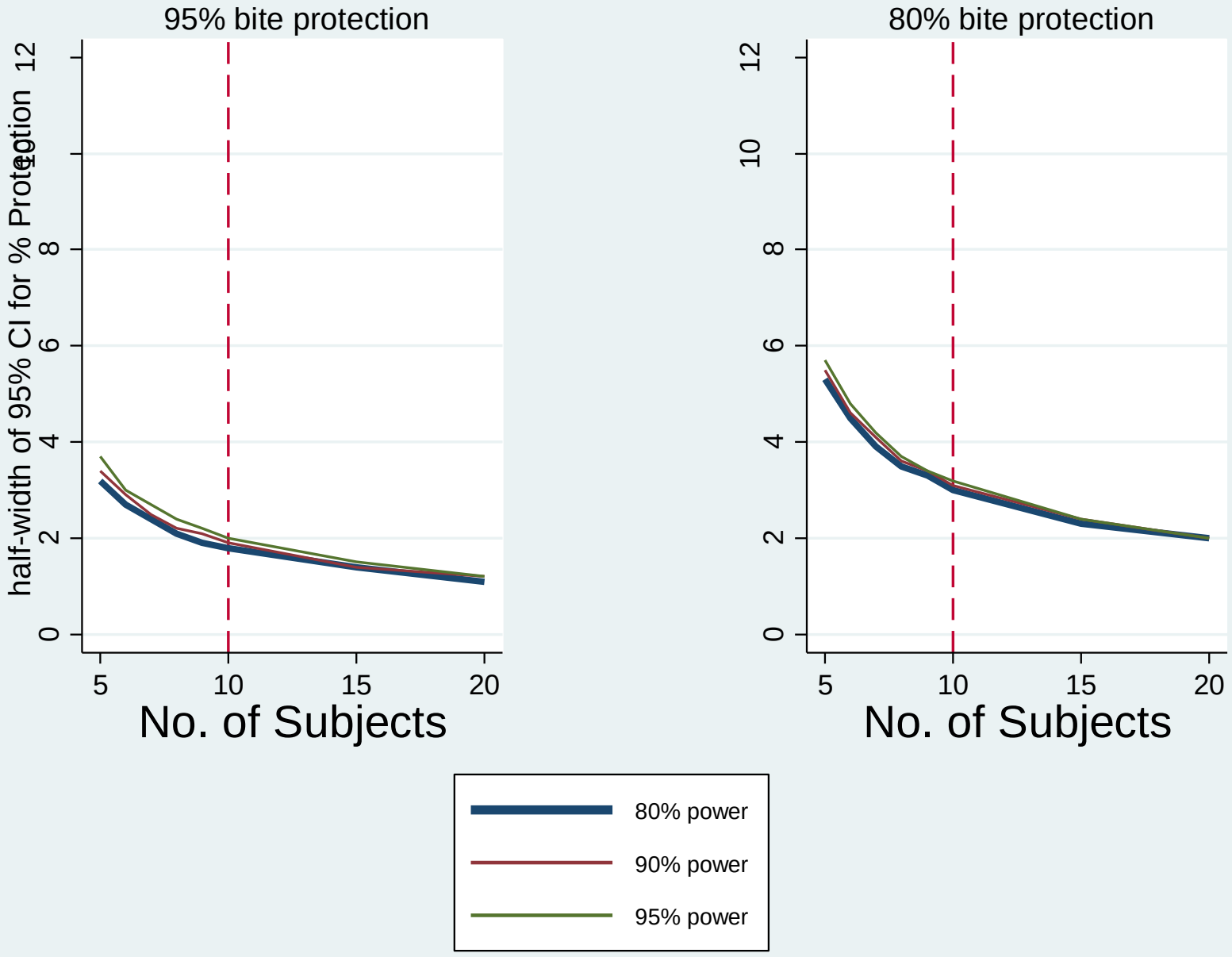
Fabric	Assumed True Bite-Through Rate in Control	Required Bite Protection Standard (% bite reduction)		Simulated True Bite Protection	No. of Subjects	Estimated Half Width of 95% C.I. Using SAS GLIMMIX Model: (subject as random effect)				EPA Conclusion
		Source	*Level*			*Mean*	*80% power*	*90% Power*	*95% power*	
ACU	10%	Mil. Spec.	85%/ 80%/ 70%	80%	15	5.6	6.1	6.5	6.8	The use of **15 test subjects** can be reasonably assured of meeting both Mil. Spec. (85%/80%/70%) and EPA Spec. (90%) bite protection standards for the ACU based on simulations at 80% to 95% power
		EPA Spec.	90%	95%	15	2.8	3.1	3.3	3.5	
FRACU	75%	Mil. Spec.	85%/ 80%/ 70%	80%	10	2.9	3.0	3.1	3.2	The use of **10 test subjects** can be reasonably assured of meeting both Mil. Spec. (85%/80%/70%) and EPA Spec. (90%) bite protection standards for the FRACU based on simulations at 80% to 95% power
		EPA Spec.	90%	95%	10	1.6	1.8	1.9	2.0	

ACU Sample Size as a Function of Percent Bite Protection



Power Analysis to Evaluate Effect of Sample Size on Precision of Bite Protection For ACU and FRACU Uniforms, (as measured by half-width of 95% CI)									
Fabric	**Assumed True Bite-Through Rate in Control**	**Required Bite Protection Standard (% bite reduction)**		**Simulated True Bite Protection**	**No. of Subjects**	**Estimated Half Width of 95% C.I. Using SAS GLIMMIX Model: (subject as random effect)**			**EPA Conclusion**
		Source	***Level***			***Mean***	***80% power***	***90% Power***	***95% power***	
ACU	10%	Mil. Spec.	85%/ 80%/ 70%	80%	15	5.6	6.1	6.5	6.8	The use of 15 test subjects can be reasonably assured of meeting both Mil. Spec. (85%/80%/70%) and EPA Spec. (90%) bite protection standards for the ACU based on simulations at 80% to 95% power
		EPA Spec.	90%	95%	15	2.8	3.1	3.3	3.5	
FRACU	75%	Mil. Spec.	85%/ 80%/ 70%	80%	10	2.9	3.0	3.1	3.2	The use of **10 test subjects** can be reasonably assured of meeting both Mil. Spec. (85%/80%/70%) and EPA Spec. (90%) bite protection standards for the FRACU based on simulations at 80% to 95% power
		EPA Spec.	90%	95%	10	1.6	1.8	1.9	2.0	

FRACU Sample Size as a Function of Percent Bite Protection



Statistical Analysis 4

- Data Analysis
  - The numbers of bloodfed and total female mosquitoes found with treated and control fabric for each subject will be analyzed as binomial distributed data in a generalized linear mixed model (GLMM) using a log link.

Measures to Ensure Reliability

- Standard Operating Procedures (SOPs) will be in place that must meet Good Laboratory Practices requirements.
- Subjects' attractiveness to mosquitoes will be determined prior to testing
  - minimum of 10% bite-through with untreated controls
- Laboratory technicians will assist subjects with placing the test sleeves on their arms and excluding all exposed skin from mosquito exposure. Laboratory technicians will assist subjects with insertion and removal of their arms in/from the cages.
- Counts of bloodfed mosquitoes and the total number of mosquitoes in the cage will be determined by a research technician.

Compliance with Scientific Standards

- The following elements are generally acceptable with refinement and clarification:
  - Experimental design
  - Statistical analysis

Highlights of EPA Science Comments

- This section highlights some, but not all, of the revisions requested by EPA before the research proceeds.
- EPA's science review documents all of the requested revisions.
- i2LResearch and LaunchBay have agreed to implement all proposed revisions to the protocol.

Highlights of EPA Science Comments 2

- Please revise "repellent treated clothing…" to "insecticide-treated clothing…" Permethrin is not a repellent. It is a toxicant.
  - *i2LResearch: Revised accordingly √*

- It should be noted that subjects that have a noticeable smell of fragrance products will not be allowed to participate since this may confound results. Also, revise "12 hours" to "24 hours."
  - *i2LResearch: Revised accordingly √*

Highlights of EPA Science Comments 3

- Since this is a lab study, all subjects who withdraw should be replaced. It is insufficient to only continue with the remaining subjects. Please revise accordingly.
  - *i2LResearch: Revised accordingly √*

- Replace "test substances" with "permethrin-treated and untreated uniform fabrics."
  - *i2LResearch: Revised accordingly √*

Highlights of EPA Science Comments 4

- Describe the fabrics in more detail, e.g., composition of fabric types and openness vs. tightness of the weave.
  - *i2LResearch: Revised accordingly √*

- In addition to the total number of mosquitoes with confirmed bites, it would be important to record the exact total number of mosquitoes in each cage (The number of mosquitoes with confirmed bites + no bites).
  - *i2LResearch: Revised accordingly √*

Highlights of EPA Science Comments 5

- The original protocol submitted by i2LResearch USA, Inc. proposed that 8 individuals serve as test subjects. However, the justification for the proposed sample size provided in the initial protocol pertains to studies where Complete Protection Time (time from application to a mosquito landing) is evaluated, which is not applicable for this study design where Mean Bite Protection will be evaluated.
  - *i2LResearch: Revised accordingly √*

Highlights of EPA Science Comments 6

- The investigators mentioned and referenced a website for Kaplan-Meier estimator. The Kaplan-Meier Estimator is not relevant to this study. This study is designed to measure bite protection, not the "time to event" measure of the Kaplan-Meier statistic.
  - *i2LResearch: Revised accordingly √*

Highlights of EPA Science Comments 7

- Raw numbers for mosquitoes with visible blood in the abdomen (obviously fed) as well as those mosquitoes which need to be crushed to see that blood feeding occurred should be provided.
  - *i2LResearch: Revised accordingly √*

Charge Question

Science

- Is the protocol "Laboratory evaluation of mosquito bite protection from permethrin-treated clothing for the U.S. Army after 0, 20 and/or 50 washings" likely to generate scientifically reliable data, useful for estimating the level of mosquito bite protection provided by the different textiles treated with permethrin?

Ethics Assessment:

i2LResearch/LaunchBay Protocol for a Laboratory Evaluation of Mosquito Bite Protection from Permethrin-treated Clothing for the United States Army

Maureen Lydon

Office of the Director
Office of Pesticide Programs

Value to Society

- Proposed study would test the mosquito bite protection of up to two fabrics, ACU and FRACU, that have been treated with permethrin via the *Invexus* process
- Study would determine whether permethrin-treated ACUs and/or FRACUs meet the target level of mean bite protection at $\geq$ 90%
- If target levels are met, the treated fabrics would provide a high level of bite protection for US Army soldiers wearing ACU and FRACU

Subject Selection

- Participants will be recruited through advertising using digital and social media.
- Recruitment firm will also use Spanish language advertisement and on-line Spanish newspaper that advertises in the recruitment area.
- Ad will contain link to study-specific website with prescreening qualification form.

Subject Selection 2

- Once completed, pre-screening qualification form will be uploaded to secure and encrypted portal, that only i2LResearch staff can access.
- The respondents who meet the eligibility criteria will be contacted initially by the recruitment firm or i2LResearch.

Subject Selection 3

- Using the telephone screening script, respondents will be asked basic eligibility questions. The purpose of the study, permethrin, test procedures and compensation will be briefly explained.
- Eligible/interested respondents will receive a follow-up call from i2LResearch to review study steps, the focus of the required training, inclusion/exclusion criteria, compensation, freedom to withdraw, and offer to provide the consent form in advance of the training to interested subjects.

Subject Selection 4

- Next step is two hour training prior to the test day.
- Part of training is provision and review of the consent form. i2LResearch will ask subjects six questions to ensure understanding of the consent form.
- Eligible interested subjects will sign consent form and receive a copy.

Inclusion/Exclusion Criteria

- 15 inclusion/exclusion criteria are complete and appropriate, except the study sponsor needs to:
  - Exclude subjects with sensitivity or allergies to insecticide-treated fabrics or insect repellents.
  - Exclude individuals with open cuts or scrapes or allergies to latex or skin care products.

Inclusion/Exclusion Criteria 2

- One of the inclusion criteria:
  - Read and speak English fluently

Rationale:

- Current product labels in English
- Language does not affect attractiveness to mosquitoes
- Research offers no benefits to subjects

Inclusion/Exclusion Criteria 3

Rationale (continued):

- Through the recruitment process, to the extent feasible, researchers will work to ensure that the ethnic groups represented in the demographics of Army soldiers -- the intended users of the treated clothing -- are reflected in the recruitment pool

Informed Consent Process

- Proposed consent process with EPA comments included is satisfactory
  - Potential subjects meet with i2LResearch for two hour training
  - Subjects given consent form, time to read it, opportunity to ask questions
  - Two hour training covers a range of topics

Informed Consent Process 2

- Training will discuss:
  - purpose of study and subject's role
  - length of test day and breaks
  - identity and function of permethrin
  - risks and steps being taken to mitigate risks
  - inclusion/exclusion criteria
  - content of the consent form and procedures
- Study staff will review and demonstrate the procedures of a 15 minute exposure interval and show subjects how fabric will be applied to their arms for the study

Informed Consent Process 3

- To confirm understanding of the consent form, subjects will be asked six questions, outlined in section 2.2.6 of the protocol
- Once the consent form incorporates EPA's comments, it will include all elements required by federal regulations

Compensation

- Each subject will be paid $30 for taking part in each training session.
- For each test day, test subjects will be paid $104.00 ($13 per hour) for any length of participation up to 8 hours.
- In the unlikely event that a test day exceeds 8 hours, subjects will be paid $19.50 (time and a half) for each additional hour, rounded up to the nearest hour.

Compensation 2

- An alternate who is not needed to replace a test subject will be able to leave and will be paid $50. The decision as to whether an alternate is needed is expected to occur within the first 2 hours of the test.
- Any subject who appears for testing, but must withdraw from the test for health-related or emergency reasons, will receive full payment as for an eight-hour day (even if they worked less than eight hours), plus any overtime worked.
- Any subject who chooses to withdraw from the study for a non-health or emergency related reason will be paid for the hours which they participated on that test day.

Highlights of EPA Comments

- This section highlights some, but not all, of the revisions requested by EPA before the research proceeds.
- EPA's ethics review documents all of the requested revisions.
- i2LResearch and LaunchBay have agreed to implement all proposed revisions to the protocol.

Highlights of EPA Comments 2

- Increase number of test subjects proposed by EPA to ensure scientific integrity of study
  - *i2LResearch: Revised accordingly √*
- Increase the number of alternate subjects to four per test day to ensure that there are subjects available to replace those who choose to withdraw
  - *i2LResearch: Revised accordingly √*

EPA Comments 3

- Revise the protocol so that if a subject withdraws after testing has begun, he or she is replaced with an alternate. Original protocol said study would proceed with remaining subjects.
  - *i2LResearch: Revised accordingly √*
- Revise the protocol so it states that an eligible and interested subject may choose to participate in up to two test days
  - *i2LResearch: Revised accordingly √*

EPA Comments 4

- To minimize discomfort, increase time between test days to 72 hours if a subject is participating in more than one test day
  - *i2LResearch: Revised accordingly √*
- Add breaks of up to 10 minutes for subjects between each exposure and a 30 minute lunch break for interested subjects that overlaps with one of the 10 minute breaks. If subject needs longer break, that's allowed
  - *i2LResearch: Revised accordingly √*

EPA Comments 5

- Ensure beverages and snacks are available to subjects
  - *i2LResearch: Revised accordingly √*
- During training session, i2LResearch should explain and demonstrate the procedures of a 15 minute exposure interval step-by-step, in addition to reviewing the other topics identified for the training session. This helps to further ensure fully informed consent.
  - *i2LResearch: Revised accordingly √*

EPA Comments 6

- Regarding questions to ensure understanding of the consent form, replace one of the questions. Ask subjects what they will wear on their arms during the exposure period.
  - *i2LResearch: Revised accordingly √*
- Add language to exclusion criteria, as previously discussed
  - *i2LResearch: Revised accordingly √*

EPA Comments 7

- Work to ensure the recruitment pool represents the demographics of the members of the Army who are the intended users of the permethrin-treated fabrics.
    - *i2LResearch: Revised accordingly √*
- Submit advertisement and pre-screening qualification form to EPA and overseeing IRB for review and approval prior to implementation.
    - *i2LResearch: Revised accordingly √*

EPA Comments 8

- As part of recruitment, post a Spanish language advertisement online, and use an online Spanish language newspaper that advertises within the recruitment area
  - *i2LResearch: Revised accordingly √*
- Work to ensure that the size of the recruitment pool is at least two times that required for the study. Update protocol to reflect this in all applicable sections.
  - *i2LResearch: Revised accordingly √*

EPA Comments 9

- Revise telephone screening script to reflect EPA's comments. Intent is to incorporate applicable changes from the protocol into the screening process as well. EPA shared suggested revisions to the screening script with the HSRB.
  - *i2LResearch: Revised accordingly √*

EPA Comments 10

- Update consent form to:
  - discuss permethrin and its uses
  - reflect updated exclusion criteria
  - update numbers of subjects & alternates
  - provide breaks between exposures
  - update topics covered during training
  - provide snacks and beverages to subjects
  - update language in form regarding no participation by pregnant/nursing women

- *i2LResearch: Revised accordingly √*

EPA Comments 11

- Regarding medical monitoring, i2LResearch should give the on-call nurse copy of final approved protocol and brief the nurse on study process and test substances. i2LResearch should contact on-call nurse at initiation of each test day to confirm that testing has begun for that day and reiterate that i2LResearch will call nurse as necessary
  - *i2LResearch: Revised accordingly √*

EPA Comments 12

- In hazards section of protocol, add psychological risks related to pregnancy testing and associated description offered by EPA for consideration
  - *i2LResearch: Revised accordingly √*
- As part of risk mitigation, screen a subset of the colony of mosquitoes to be used in order to check for pathogens
  - *i2LResearch: Revised accordingly √*

EPA Comments 13

- Revise language associated with covering medical costs so that it reads:

  If a subject is injured as a result of wearing the permethrin-treated fabric or from study procedures, study sponsor will directly pay for medical expenses necessary to treat subject's injuries that are not covered by their insurance

  - *i2LResearch: Revised accordingly √*

EPA Comments 14

- Clarify the benefits section and provide more details
  - *i2LResearch: Revised accordingly √*
- Add a reference to complying with FIFRA §6(a)(2) adverse effects reporting requirements
  - *i2LResearch: Revised accordingly √*
- Clarify the existing statement that "adverse effects will be followed until resolution is reached"
  - *i2LResearch: Revised accordingly √*

EPA Comments 15

- Revise protocol to state, if subjects request standard, over-the-counter antiseptics and hydrocortisone cream, it will be provided immediately upon completion of the test at no cost to subjects
    - *i2LResearch: Revised accordingly √*
- In application section of protocol, clarify that different sized sleeves "will be" created to fit subjects' needs, instead of "may be" created
    - *i2LResearch: Revised accordingly √*

EPA Comments 16

- i2LResearch and LaunchBay have agreed to incorporate and implement EPA's comments

Risks and Risk Minimization

Protocol provides appropriate measures to minimize five categories of risk:

- Adverse reaction to test substances
- Exposure to biting mosquitoes and mosquito-borne diseases
- Physical discomfort of multiple mosquito bites
- Unanticipated loss of confidential information
- Psychological risks related to pregnancy testing

Risks and Risk Minimization

Steps to minimize adverse reaction to test substances:

- Protocol excludes candidates who are known to be sensitive to insecticide-treated fabrics or insect repellents are excluded.
- Protocol excludes subjects with cuts, scrapes, skin diseases or skin conditions such as psoriasis, atopic dermatitis or eczema. These conditions could increase the possibility of a reaction to test material.
- Subjects will be told that if anyone experiences any skin reaction, experiences an injury, or simply feels unwell, he or she should inform i2LResearch staff right away. Such subjects will immediately be given appropriate care, may be withdrawn from testing, and may be transported to a local hospital if necessary. The closest hospital to the laboratory test site and directions will be identified prior to the test date.

Risks and Risk Minimization

Steps to minimize exposure to mosquito-borne diseases:

- To eliminate the risk of contracting any mosquito-borne diseases, the study will be conducted only with laboratory-reared mosquitoes, which are not known to harbor any pathogens.
- In order to ensure the mosquitoes used in the study are not carrying any diseases, a subset of the colony will be screened for pathogens as described in the protocol.
- In addition, the supplier will document that these laboratory-reared mosquitoes are disease free, and that they have never received a blood meal.

Risks and Risk Minimization

Steps to minimize physical discomfort of multiple mosquito bites:

- Protocol excludes candidates who are allergic, hypersensitive to or phobic of mosquito bites.
- Subjects are alerted in the consent form to the possibility of experiencing a skin reaction to mosquito bites, and are advised to inform the study director or other staff member, if they believe they are having a reaction.
- Over-the-counter topical anti-itch gel, or cream to relieve itching, will be available for use by subjects after completion of the study.
- There will be at least 72 hours between test days and subjects can only participate in up to two test days.
- A nurse familiar with the protocol will be on-call to provide advice or assistance in case medical advice is needed during the test day.

Risks and Risk Minimization

Steps to minimize the unanticipated loss of confidential information:

- All efforts will be taken to maintain the confidentiality of the pregnancy test results. The test results will not be disclosed to anyone other than the test subject, the verifying employee, and/or the Study Director.

- In addition, the subjects' identities and participation in the study will be protected as follows: each subject will be assigned a code number, and only subjects' code numbers will appear on data sheets. The subjects' names will not appear anywhere on the data sheet, or in the reports. The study records will be maintained at the testing facility in locked cabinets and electronic files kept on a password-protected computer server.

Risks and Risk Minimization

Steps to minimize psychological risks related to pregnancy testing:

- The protocol provides for discrete handling of the pregnancy testing that is required of female subjects on each test day.
- Female subjects self-administer the pregnancy test in a private bathroom.
- After completing the test, each female subject is asked if she would like to continue in the study. If her answer is no, then no further questions are asked; she will not be asked to share the result with anyone. If her answer is yes, the result of the pregnancy test will be verified by only one member of the research team who will be female.
- For females who proceed with the testing, the result of the pregnancy test is not recorded.

Benefits

- No direct benefit to subjects
- Primary direct beneficiary is study sponsor
- If the treated materials are proven effective and meet the target level of mean bite protection of ≥90%, that's a high rate of mosquito bite protection. Indirect beneficiaries would include US Army soldiers who wear permethrin-treated ACUs and FRACUs

Risk-Benefit Balance

- Risks have been effectively minimized
- Risks are reasonable in light of the expected societal benefits of the knowledge likely to be gained

Respect for Subjects

- Effective methods for protecting subjects' privacy
- Proposed level of compensation is appropriate
- Subjects free to withdraw at any time
- Study sponsor will directly pay for medical care for research-related injuries, not covered by subjects' insurance

Independent Ethics Review

- Schulman IRB reviewed and approved the protocol and informed consent materials
- Schulman IRB has AAHRPP accreditation, is registered with OHRP, and is independent of the investigators

Applicable Ethical Standards

- This is a proposal for third-party research involving intentional exposure of human subjects to a pesticide, with the intention of submitting the resulting data to EPA under the pesticide laws
- The primary ethical standards applicable to the conduct of this research are 40 CFR 26, Subparts K and L, and FIFRA 12(a)(2)(P)
- Attachment 1 to the EPA Review contains a point-by-point evaluation of how this protocol addresses the requirements of 40 CFR 26 Subparts K and L

Compliance with Ethical Standards

- Requirements of §26.1111, §26.1116, and §26.1117 have been met
- Requirements of §26.1125 have been met
- Requirements of §26.1203 have been met

Findings in EPA Ethics Review

- i2LResearch and LaunchBay agreed to address EPA's comments
- No deficiencies relative to 40 CFR 26, Subparts K and L, or to FIFRA §12(a)(2)(P)
- Protocol meets the applicable requirements of 40 CFR Part 26, Subparts K and L

Charge Questions

Science

- Is the protocol "Laboratory evaluation of mosquito bite protection from permethrin-treated clothing for the U.S. Army after 0, 20 and/or 50 washings" likely to generate scientifically reliable data, useful for estimating the level of mosquito bite protection provided by the different textiles treated with permethrin?

Ethics

- Is the research likely to meet the applicable requirements of 40 CFR Part 26, Subparts K and L?