COLLECTION AND REARING METHODS FOR FIELD STRAINS OF *Cimex lectularius*

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Abstract:
Research shows that field strains of *Cimex lectularius* (Bed Bugs) are building resistance to pesticides, and as the world-wide complaints of bed bugs reach epidemic levels, it becomes clear that expedient studies addressing resistance must be made . In order to address this most pressing issue, field strains must be collected from active sites and then sustained on blood-meals in the lab. Sadly, due to the stigma associated with bed bugs, field collections are often hard to come by, hampered by nervous property owners who think their bed bug problem will magically go away. This poster covers collection measures once a field site is established, and also offers suggestions for rearing once samples are procured.

Introduction:
Bed bugs (Fig. 1) are small insects that feed upon human blood, a feeding that typically occurs while the host is sleeping, leaving painful bites and instilling panic into its victims, causing a fear of sleep. Bed bugs lay 4-7 eggs (Fig. 3) per day when sexually mature and when hatched, nymphs emerge (Fig 2). It is not uncommon for sufferers to saturate sleeping linens with roach poison, bleach, or kerosene , futilely attempting to thwart this enigmatic and persistent nocturnal foe. While collecting field samples, you must bear in mind that bed bugs are largely dispersed by hitch-hiking upon humans. You do not want to become the principal vector transporting these blood-suckers around town and ultimately into your own bed. Once you have collected field strains and brought them back to the lab, you will inevitably discover a few challenges standing in the way of feedings, with the two methods available being artificial and natural rearing methods. Artificial rearing methods are cost, time, and labor intensive. You will need an initial investment to purchase an incubator(Fig. 6), water bath (Fig.4), jacketed vials (Fig. 5),and weekly inputs of rabbit, chicken, or human blood. Natural methods involve a willing volunteer who offers his own body (blood-meals) for the purpose of scientific inquiry.

Collection Methods:
Familiarize yourself with bed bugs. Many web-sites are devoted to bed bug biology, and I suggest you read the sites containing comments from bed bug victims as well. This will give you valuable insight into the bed bug crisis. I also recommend you read the book by Pinto and Associates: Bed Bug Handbook as well as the SF Department of Public Health's Article 11, Sec. 581 which is the Director's Rules and Regulations on How to Control Bed Bug Infestation.

You will need a few basic tools:
Shoulder bag with strap to hold all your equipment
Small, bright flashlight
Latex gloves
Magnification lens and broad-nosed forceps
Screw-top vials and labeling tape for specimen collection
Lace-free and mid-calf high rubber rain boots to tuck your pants into (or an old pair of shoes)
Snug-fitting clothes that can easily be removed post-inspection
2 extra strength garbage bags for post-inspection shoes and clothes as well as containment housing for infested objects
Small pry bar to, gently and without damage, pry loose objects from walls and baseboards for inspection
A playing card
5 small plastic zip-lock bags for smaller objects and specimen containment
An extra set of clothes and shoes to change into following the inspection.
A clip-board with graph paper and a few colored markers to take notes of, and sketch, the afflicted rooms.
A small, powerful, multipurpose, and bag-less rechargeable vacuum cleaner with various nozzle attachments helps to suck up bed bugs and their eggs if you are bringing samples back to the lab, But, be warned, you must also bag and seal the vacuum when the inspection is complete!

Prior to entering the infested unit: Record the date, property address, and unit numbers you will be inspecting in a notebook. If known, also document the treatment history for the building / affected units. Remember, you are responsible for conducting inspections, documenting the affected areas, collecting samples, and practicing personal safety—**do not set yourself or your stuff down upon any objects in the room**. Casual contact with articles within the room is unavoidable, especially during comprehensive inspections. At the end of the inspection, you will remove your shoes and shoulder bag, place them in one of your garbage bags, and tie a firm knot at the opening.

Post-Inspection:
Everything you took into the site should be placed in one of your garbage bags *before you leave the premises*. The extra set of shoes and clothes should be, ideally, changed into at the sites bathroom; however, this is not always possible. At the very least, I recommend you change into your extra set of shoes while at your car, placing the contaminated shoes into your garbage bag and then secure the bag with a hearty knot. As soon as you get home, wash your shoes and clothes in a washing machine. All equipment used for inspections is now to be used only for bed bug services. It helps to have a large, clear plastic bin at the lab, so that you can keep everything in it, retrieving easily the specimen vials and notes you collected

Figure 1

Figure 2

Figure 3

Figure 4

Figure 5

Figure 6

Figure 7

Artificial Rearing:
Bed bugs optimally breed at 27 degrees Celsius and due to traumatic insemination (spearing the females body cavity with male genitalia) , it is recommended that you separate the males from the females at a 1:1 ratio to prevent high rates of female mortality. Fresh blood needs to be suspended in a jacketed vial that is surrounded by water heated to 37 degrees Celsius. Papers suggest that you use Nescofilm wrapped around the jacketed vial as a membrane to hold back blood and induce biting responses; however, this proved unsuccessful in my attempts, so I experimented with turkey and chicken skin, proving likewise to be ineffectual.

Natural Rearing:
After many long and trying failures at artificial rearing, I decided to self-feed my colony, starting with just a few at first, building my immunity, and ending with a typical weeks feeding to be around 1200 and growing. It is not suggested that this method be emulated as severe reactions can be experienced which may be life threatening. However, the end result is a completely cost effective and efficient method to sustain my colony, which is the goal that I set before myself at the beginning of my project. Figure 7 displays the rearing jars and lid that are used for feedings. This colony is growing very rapidly and due to its current success, there are plenty of specimens for future experiments.

Conclusion:
There was not much data to support me while developing collection procedures ,and after many frustrating attempts to artificially rear my colony, I was compelled to use non-traditional methods to keep my work alive. I even designed and built the jacketed vial in figure 5 from a bic pen, urine analysis container, film container, and a glue gun to cut the cost of an expensive custom glass vial. And, after all is said and done, staring adversity in the face and achieving relative success was by far the greatest reward during my research. Future work can now have a chance to be fruitful due to the efforts put forth.

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