

Quick guide for the disinfestation with Steam-X Super-Heated Steam Gun

Operating manual for the treatment of bed bugs with steam



Index

Introduction

1.1 General information	4
1.2 Getting to know the enemy: bed bugs	5
1.3 How to identify a bed bug infestation	6

Why steam?

2.1 Advantages of using steam	7
2.2 Advantages of using Steam-X Super-Heated Steam Gun	8

Preparing for steam treatment

3.1 Preparing the area to be treated	9
3.2 Preparation of the steam generator with Steam-X Super-Heated Steam Gun	10
3.3 Tips to ensure a complete and effective treatment	11

Conclusions

4.1 Post-treatment monitoring and prevention measures	12
4.2 Evaluation of treatment success	12

References 13

Attachments

5.1 Bug biology	13
5.2 Inspection form	15
5.3 Procedure when reporting	16
5.4 Steam-X Super-Heated Steam Gun certification	17-27

Introduction

1.1 General Information



Bed bugs (*Cimex lectularius*) are a significant problem for people.

These small pests, known for their ability to hide in the most unnoticeable crevices and for their resistance to insecticides, feed on blood in the same way as hematophagous insects (mosquitoes, phlebotomuses, horseflies, fleas, etc.).

Their stings are generally painless, which allows the insects to feed without being disturbed. However, they cause itching, skin irritation and, in some cases, allergic reactions. A bed bug infestation can lead to considerable psychological and physical discomfort for affected individuals.

Therefore, it is essential to have a thorough understanding of the biological characteristics and behaviour of these insects, as well as to make use of control and prevention means and strategies in order to deal effectively with infestations. Nowadays, the presence of bed bugs is no longer just a problem that afflicts hotels, Bed&Breakfasts or private homes, but also affects public transport, train stations and airports. In fact, the presence of these insects has also been found on seats in buses, trains and planes.

At European level, in countries such as France, Germany and England, national institutions declared a national emergency in 2023, while Italy has recently experienced a significant increase in the number of infestations.

This technical manual is designed to offer detailed and practical information on bed bugs with the aim to support the professionals in this field with the management and eradication of these parasites.

1.2 Getting to know the enemy: bed bugs

Health significance

Bed bugs cause erythema and allergic reactions through their bites, which, although painless, cause annoying itching that can also lead to secondary infections and cause sleep disturbances, anxiety and psychological stress. Although they do not transmit diseases, their impact on the quality of life and the costs associated with pest control are quite significant, making effective pest management necessary.

Biological characteristics of bed bugs

Bed bugs are nocturnal parasites of an oval and flattened shape, they measure about 4-5 mm as adults and are reddish-brown in colour. Feeding exclusively on human blood, their colour turns darker after sucking the host's blood.

Their life cycle includes three developmental stages: egg, nymph and adult. The nymph becomes an adult by moving from one moult to the next feeding on blood. The mature eggs hatch after laying, depending on temperature, after 10 days (20° C) or 4-5 days (35° C). The complete cycle from egg to adult under ideal conditions of 20° C takes about a month on average.

Behaviour and eating habits

Bed bugs are attracted by body heat and carbon dioxide emitted during human breathing. They feed every 5-10 days, but can survive for months without feeding. Generally, one of their meals can last between 3 and 10 minutes and, during this time, the insect attaches itself to the host's skin and feeds until it is completely satiated.

During the day, the pests hide in cracks and crevices near resting places, as well as mattresses and bed bases, as they cannot stand light. Moreover, it is not unusual to find them inside electrical sockets, in baseboards, cupboards, behind pictures or curtains.


Introduction

1.3 How to identify a bed bug infestation

Recognising a bed bug infestation requires a lot of attention and patience.

In order to carry out an accurate inspection, it is necessary to have some essential tools to identify their presence early on, including:

- Torch
- Magnifying glass
- Screwdriver
- Insect collector (like a jar with a screw-on lid which you can find in pharmacies)
- Insect brush or tweezers

 **Caution:** *Following the guest's report, it is essential to carry out the environmental inspection as soon as possible, in order to promptly take the required measures to eliminate the presence of bed bugs*

Signs that could indicate the presence of bed bugs are as follows:

1. **Bites on the skin:** if the host wakes up with bites on the skin, often arranged in lines or groups, these could be a sign of bed bugs. These bites can be itchy and tend to appear mainly on arms, legs, neck or other exposed body parts during sleep.
2. **Traces of blood or faeces:** after feeding, bed bugs can leave traces of blood on sheets or pyjamas, a sign of their passage. In this regard, it is essential to carefully observe the seams around mattresses or pillows. It is also possible to find dark-coloured excrements (0.3 mm to 1 mm in diameter) in the drawers of furniture next to beds, in wardrobes or inside suitcases.
3. **Unpleasant smell:** it is possible to perceive an 'unpleasant' smell in the bedroom or in the vicinity of the bed and/or sofa; this could be a sign of infestation. However, the smell alone does not provide any evidence that bedbugs are actually present in the room.
4. **Direct sighting of bed bugs:** it is likely to have seen with the naked eye (and even photographed) young or adult bed bugs on the mattress, bed or surrounding furniture. However, it is still necessary for pest control personnel to visit the site to verify and determine the extent of the infestation.

2.1 Advantages of using steam

- **Immediate effectiveness**

High-temperature steam quickly kills adult bed bugs as well as their eggs and nymphs. The high temperatures penetrate into cracks and folds in mattresses, recesses where bedbugs tend to hide.

- **Penetration into crevices**

Steam has the ability to penetrate cracks and hidden places where bed bugs often take refuge, reaching areas that would otherwise be difficult to treat with insecticide sprays or powders.

- **Absence of neurotoxic insecticides**

Pest elimination by steam does not require the use of insecticides or other chemicals: this is particularly advantageous for 'sensitive' environments such as bedrooms, hospitals and schools, where exposure to chemicals could be dangerous.

- **Reducing insecticide resistance (absence of resistance)**

Bed bugs can develop resistance to insecticides over time, making chemical treatments less effective. In contrast, steam benefits of the physical heat given by the high temperature, to which bedbugs cannot develop resistance.

- **Safety for human and pet health**

Steam is safe for people and pets. It does not leave toxic residues in the surrounding environment and does not represent a risk for human health.

- **Versatility and accessibility**

Steam generators are versatile tools that can be used on a wide range of surfaces and materials, including mattresses, furniture, upholstery, carpets and curtains.

- **Removal of hidden dirt**

In addition to eliminating bed bugs, steam also removes hidden dirt and allergens, improving the overall cleanliness of the environment.

- **Environmental sustainability**

The use of steam makes it possible to manage the infestation of these pests through an environmentally friendly and sustainable method, reducing or eliminating the need for harmful chemicals.

Why steam?

2.2 Advantages of using Steam-X

- Steam gun equipped with an internal 400W heating element that allows the output steam to overheat to 130°C.
- The accessory's special terminal softens the steam flow to prevent the dispersion of the bugs into the surrounding areas.
- Equipped with a steam hose and plug for connection to all our steam generators from 4 bar to 8 bar pressure.
- High-temperature steam is lethal for bedbugs and their eggs.
- Unlike chemical pesticides, steam is a completely environmentally friendly cleaning method that does not release harmful residues into the environment.
- The gun can be used on all surfaces.
- Supervision of the environment and regular use of steam helps to prevent the reappearance of new infestations.

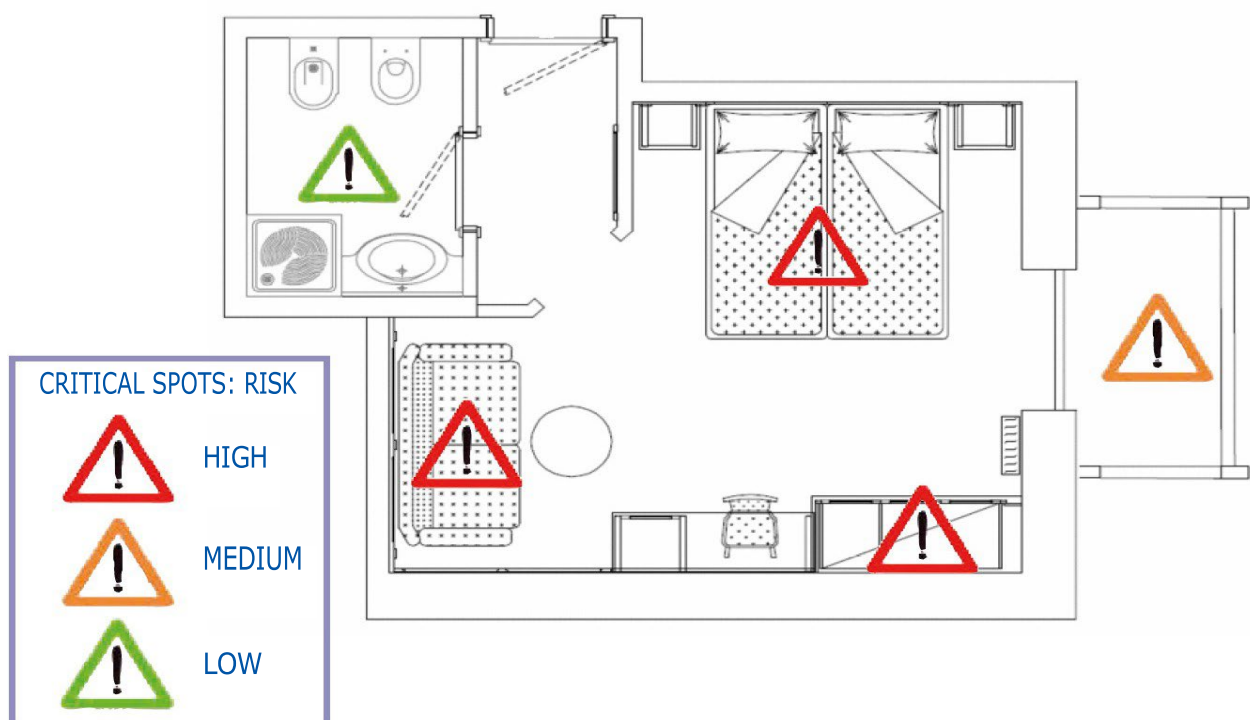


Preparing for steam treatment

3.1 Preparing the area to be treated

To properly combat bedbugs, certain steps must be followed to ensure the effectiveness of the pest control treatment. First, tell the customer not to move anything, as far as possible, until the technician arrives. Next:

- Remove bed linen (sheets, blankets, pillowcases) and proceed to machine wash at 60°C at least.
- Inspect the mattress, also carefully check the bed frame. If possible, dismantle the bed to inspect each component.
- Remove clothing and other textiles: move all clothing, pillows, curtains and other textiles in the room. Also wash these items at high temperatures and dry them properly.
- Reduce clutter: reduce clutter in the room and inside furniture and cupboards to facilitate the operator's access to all infested areas.
- Clean the environment: in addition to catching bugs and their eggs, use a vacuum cleaner with a HEPA filter to remove dust and other debris from the area.
- Dispose of waste: immediately empty the vacuum drum into a sealed plastic bag and then steam the surface to eliminate all living forms of bedbugs or other organisms.



Preparazione per il trattamento con il vapore

3.2 Preparation of the steam generator with Steam-X

1. Fill the tank or the boiler, depending on the model you purchased, with water.
2. Connect the power cord of the steam generator to the electrical outlet.
3. Connect the Steam-X accessory plug to the socket on your steam generator, ensuring that the black spring on the plug is correctly locked into the socket.



For the Steam-X version with a 1.5mt power cord:

- connect the Steam-X electrical plug to the vacuum socket placed on the back of the generator (for example Carmen, Junior Star Max, Steam Box Vac Mini)

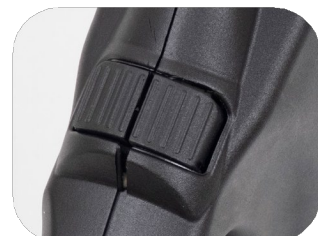
For the Steam-X version with a 5mt power cord:

- connect the Steam-X electrical plug to the wall socket.

4. Activate the general power switch on the back of the machine, press the power and the boiler button, wait approximately 5/6 minutes until the green 'steam ready' indicator on the machine lights up, at which point both the steam generator and the Steam-X gun will be ready to be usage.
5. If the accessory is correctly plugged in, the heating element inside the gun will start to heat up.

⚠ Caution: When this tool switched on for the first time, smoke may come out of the gun. This is due to moisture accumulated during storage and will cease after a few minutes. With the first usage or if not in use for a longer time, make sure to discharge the first steam flow to the ground to clean the hose of any eventual processing dross and impurities.

6. To activate the steam flow, press one of the two buttons on the accessory handle. Both buttons are set with the same steam flow rate (minimum steam flow).



⚠ Caution: Before steaming surfaces, discharge the condensation produced by the appliance into a container for approximately one minute.

7. It will now be possible to proceed with the surface treatment: work at a distance of about 20 centimetres while keeping the gun in a slow movement.



3.3 Suggestions to guarantee a complete and effective treatment

In order to guarantee a complete and effective treatment against bedbugs, it is important to follow a few key tips:

- Inspect all suspicious areas including beds, furniture, carpets, curtains, electrical outlets, baseboards or cracks in the floor or walls.
- Look for the presence of bedbugs or their passage (such as dark stains on fabrics)
- Move the dispensed steam slowly over all infected surfaces to ensure that the heat penetrates and kills the bedbugs and their eggs.
- If the bedbug infestation is particularly massive, more inspections and more steam interventions will be necessary.
- After the first treatment, it is suggested to schedule at least one follow-up visit within 7-10 days
- All signs of infestation (blood spots, faeces, adults, and nymph) must be removed in order to not confuse future inspections.
- After treatment, it is advisable to ventilate the site to ensure a complete drying of the treated spots

Conclusions

4.1 Post-treatment monitoring and prevention measures

- **Environmental cleaning:**

keep rooms and areas of possible bedbug infestation clean and tidy to reduce insect hiding places

- **Monitoring with traps:**

once the steam treatment is complete, passive monitoring devices can be placed at key locations. The number of devices will vary depending on both the size of the sites and the type of infestation found. Various models of monitoring traps specifically for bed bugs are available on the market.

- **Training:**

if you have to work in accommodation facilities, it is essential to adequately train staff in proper cleaning and tidying of the room, as well as, specifically, in bed-making. In this regard, it is important that staff are aware of where monitoring devices are located so that they can promptly report any evidence of bedbugs to the technicians in charge of carrying out disinfestations with steam.

- **Sealing:**

sealing all cracks and crevices in walls, floors and furniture to prevent bedbugs from finding new potential hiding places.

4.2 Evaluation of treatment success

Only 7-10 days after the pest control treatment the success of the intervention can be verified.

Thereafter, regular monitoring should be carried out to ascertain the effective elimination of the pests.



Caution: As bed bug eggs hatch in 7-10 days at room temperature, it is advisable to wait at least this period between the end of the treatment and the follow-up visit.

References

- *Artropodi di interesse sanitario in Italia e in Europa. Roberto Romi, Cristina Khoury, Riccardo Bianchi e Francesco Severini 2012, ii, 172 p. Rapporti ISTISAN 12/41.*
- *Cimice dei letti. manuale per la prevenzione e la gestione dell'infestazione. Gioia Capelli, Fabrizio Montarsi, Giulia Maioli. Istituto Zooprofilattico Sperimentale delle Venezie . pp. 51. 2006.*

Photografic documentation source: Istituto Zooprofilattico Sperimentale delle VenezieArti

Attachments

5.1 Bedbug biology

The bedbug (*Cimex lectularius*) is the best-known insect in the family Heteroptera Hemiptera (Heteroptera). It is a dark brick-red, ferruginous insect that can measure 6 to 8 mm in total length (females are larger than males). When it is fasting, its body is oval and strongly flattened.

When it has completed its blood meal (adult 'engorged') its shape is more elongated. Its head is short, transversely dilated, with small but very protruding compound eyes, with antennae of four articles and a rather short rostrum, which, when resting, lies almost invisible in a groove.

The thorax is rather small and to it are attached the elytra reduced to small oval scales, which leave the broad abdomen, which is almost round in shape, fully exposed. The legs are of medium length and are equipped with three-article tarsi.

In order to fulfil their life cycle they need a rather high temperature and therefore settle in rooms that are heated during winter and in the warmest places in the house.

They are, however, able to withstand low temperatures but, in this case, they suspend activity until the environment is thermally suitable. They feed during the night, usually for 3-10 minutes, and are bothered by light. They return to consume their blood meal after a week.

They can remain fasting, while maintaining their aggressive capabilities almost intact, for months on end, even for over a year. If they live in environments where the temperature fluctuates little during the course of the year, they reproduce at any time and can have three generations within twelve months.

The bugs lay, usually in the same environments where they take refuge during their resting hours, packages of pearly grey eggs. These are just over 1 mm long and half a millimeter wide, sub-elliptical, with a small operculum



Female Cimex lectularius



Male Cimex lectularius

at the anterior pole. In total, each female deposits from 200 to almost 300 eggs.

Incubation and the duration of the pre-imaginal period (juvenile stages) depend mainly on environmental conditions, the availability of food and, most prominently, on thermal conditions.

At an average temperature of around 25°C, the eggs hatch after 8 days, but it then takes 11 weeks for the newborns to reach the final stage after they have completed 5 molts.

It is also necessary for the larvae to take in a certain amount of blood for each life stage. As they do not feed, or feed poorly, the pre-adult period is very long. Even larvae can go through very long periods of fasting without suffering. If they develop under optimal conditions of environment and nutrition, it takes only 7 weeks for the bugs to move from early adulthood to their imago stage.

The cold slows down their development, but even too high temperatures are not tolerated. If, for example, 50° C is reached in their environment for a few days, all young and adult individuals die. If the bugs have plenty of food at their disposal, they can quickly multiply and get to worrying densities (over 1,000 individuals in 90 days).

Health importance (ISTISAN Reports 12/41.)

Although bedbugs are considered potential disease vectors, they are not involved in the transmission of any disease-causing pathogens in humans.

However, bedbug bites cause local itchy edema and erythema, largely due to allergic phenomena resulting from the inoculation of an anticoagulant contained in the insect's saliva.

The reaction is subjective: in some cases the local inflammation is considerable, in others practically absent. The act of scratching can cause secondary bacterial infections in the edematous area.

Heavy infestations can cause nervous system and digestive disorders in hypersensitive individuals.

It has also been reported that children living in heavily infested homes may suffer from severe forms of asthenia, which cannot always be attributed solely to malnutrition.

5.2 Inspection form

Date	Place
Operator	Room no.

	YES	NO	OBSERVATIONS
Bed sheets and pillowcases			
Head area mattress			
Foot area mattress			
General area mattress			
Bed frame			
Bed feet			
Bed slats			
Lockers			
Closet			
Baseboards			
Electric sockets			
Paintings			
Chairs			
Armchairs			
Etc.			

*other insects/ characteristic smell/ excrement stains/ etc.

Attachments

5.3 Procedure when reporting room bugs

- DATE: ____ / ____ / ____
- RESPONSIBLE STAFF FOR MONITORING ROOM N° ____
- NUMBER OF BEDROOMS INFESTED: ____
- NAME OF THE COMPLAINING CUSTOMER: ____
- ORIGIN OF THE CUSTOMER: ____
- ORIGIN OF THE PREVIOUS CUSTOMER: ____

PROCEED WITH THE FOLLOWING STEPS:

1. Close the room and do not allow anyone to enter except for the personnel involved in the inspections.
2. Proceed as indicated in the 'inspection form'
3. Collect any insect specimens in test tubes/jars to show to the pest control company.
4. Call the reference pest control company.

AFTER TREATMENT:

1. Do not enter the room until the pest control company has declared it accessible.
2. Proceed with a thorough environmental cleaning to remove any residues from the surfaces in the room.

5.4 **Steam-X** certification



ENTOSTUDIO S.r.l.
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EFFICACY EVALUATION OF "STEAM-X"

AGAINST *Cimex lectularius*

Pag. 1 di 11

VALUTAZIONE DELL'EFFICACIA DI "STEAM-X" NEI CONFRONTI DI *Cimex lectularius*

Test concluso il

11 giugno 2024

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Report code: Q016A-24-Q042A-24



INDEX

1. TEST INDEX-CARD.....	3
2. TEST OBJECTIVE.....	4
3. SUMMARY	4
4. MATERIALS AND METHODS.....	4
4.1 SPECIES OF INSECTS USED FOR THE TEST AND BREEDING CONDITIONS.....	4
4.2 DETAILS OF THE TESTED MACHINERY	4
4.3 DOSAGE APPLIED	5
4.4 TEST-ROOM SPECIFICATIONS	5
4.5 EQUIPMENT	6
4.6 DEFINITION OF TEST AND REPLICATE	6
4.7 EVALUATION PARAMETERS	6
4.8 TEST DESIGN	7
5. RESULTS.....	9
5.1 ADULTS AND NYMPHS.....	9
5.2 EGGS.....	10
6. CONCLUSIONS.....	11



1. TEST INDEX-CARD

Study conducted for:	TECNOVAP srl Via dei Sassi, 1/a – 37026 Pescantina (VR) - Italy
Study conducted by:	Entostudio S.r.l. Viale del lavoro, 66 - 35020 Ponte san Nicolò (PD) – Italy
Study conducted in:	Entostudio S.r.l. Viale del lavoro, 66 - 35020 Ponte san Nicolò (PD) – Italy
Study coordinated by:	Entostudio S.r.l. Viale del lavoro, 66 - 35020 Ponte san Nicolò (PD) – Italy
Supervisor of the study:	Dr. Andrea Drago
Responsible of the study:	Matteo Cecconello
Tested product:	STEAM-X
Species used for the test:	<i>Cimex lectularius</i>
Test start date:	May 20, 2024
Test completion date:	June 11, 2024



2. TEST OBJECTIVE

To evaluate the efficacy of the "STEAM-X" Steam System against the target species through a simulated use test.

3. SUMMARY

Target insect: *Cimex lectularius*

Formulation of the tested machinery: The machinery used for the test is a steam generator, "STEAM-X," equipped with a superheater gun. Distilled water was used for the treatment.

Method of testing: A shelter is created by placing two surfaces on top of each other to create a small gap. The shelter is then placed in a plastic arena where 5 insects (of mixed stage and sex) are placed inside the slit. After a period of acclimatization, the treatment was performed by applying steam around the shelter. The knockdown time and mortality at 24 h is then evaluated.

The same procedure was performed by placing 10 eggs in the shelter that are observed for 1 week after treatment.

4. MATERIALS AND METHODS

4.1 SPECIES OF INSECTS USED FOR THE TEST AND BREEDING CONDITIONS

The bed bugs used for the test came from the laboratory of Entostudio.

The bedbugs are kept in the laboratory at 27°C and 75% R.H.. The photoperiod lasts 12 hours. The bedbugs are confined into a falcon containing a piece of black cardboard and closed with a tulle netting. Once a week the container is placed tulle side down on a Parafilm sack filled with defibrinated rabbit blood in order to let the bed bug's feed. The females lay the eggs on the cardboard.

The colony originated in 2022 from bedbugs provided by Czech University of Life Sciences Prague (CZU).

4.2 DETAILS OF THE TESTED MACHINERY

Name: EVO

ENTOSTUDIO S.r.l. Cap. Soc.: € 40.000,00 I.V.	Viale del Lavoro, 66 - 35020 Ponte San Nicolò PD - ITALY	Tel. & Fax +39 0497402487	P.Iva IT03951900285 C.F. 03951900285	www.entostudio.com info@entostudio.com
Report code: Q016A-24-Q042A-24				



Description of machinery tested: The test was carried out with steam generator mod. Evo sanitizer (boiler power 800 W, operating pressure 4 bar), and steam gun with superheater. The compatible models for the test performed are the following: all Tecnovap steam generators with a boiler output of 800 W - 4 bar or more.



Fig. 1 Machinery used for the test

4.3 DOSAGE APPLIED

Each shelter was treated for 4 seconds, applying steam perimeterwise at a distance of 20 cm.

4.4 TEST-ROOM SPECIFICATIONS

The test was performed in a room measuring 3.15 x 2.90 x 3.03(h) m corresponding to a surface of 9.13 m² and a volume of 27.7 m³. The room is washable thanks to a resin floor and varnished walls. The room is completely white. The climatic parameters (temperature and humidity) and the illumination are automatically controlled and regulated: the illumination is provided by led lights 4000°K solar-spectrum. The test was performed at 300 Lux intensity, 25±1°C and 60±5% Relative Humidity.

The room has an adjustable air extraction system that is capable to change the air up to 2800 m³/h.



4.5 EQUIPMENT

For the test procedure, the following devices were used as specified:

- CompactSteam humidifier (CAREL S.p.A.);
- TFA, digital thermo-hygrometer to measure environmental parameters;
- Nimex Ni 2600, digital lux meter to measure light intensity;
- Daikin FDXM25F2V1B (indoor unit) and Daikin RXM25M (outdoor unit), for the air conditioning temperature;
- BITICINO H4684-AM5864, for photoperiod management;
- Arena SAMLA 78 x 56 x 18 cm (Volume 56 L)
- Stereo-microscope ZEISS Stemi 508;
- Steam generator, mod. Evo.

4.6 DEFINITION OF TEST AND REPLICATE

Test: is the complete evaluation procedure of a product. It is given by the set of replicates performed.

Replicate: corresponds to a treated arena and a control arena. The procedure begins with the introduction of the insects into the container.

4.7 EVALUATION PARAMETERS

Dead: Individuals which do not show any movement and which do not react to external stimuli (i.e. when touched with a pair of tweezers) are considered as dead.

Moribund: after the test starting the individuals that react to stimuli but are unable to move in a coordinated manner, e.g. to upright itself or walk properly, are considered as "moribund". These insects left into the arena. If moribund insects turn dead, the data is reported to the moment when they were observed as moribund.

Alive: Individuals which do not show any behavioural alterations.



4.8 TEST DESIGN

The test was performed in a plastic arena measuring 78 x 56 x 18 (h) cm, having an area of 0.43 cm² and a volume of 56 L.

Each arena corresponds to a replica.

A shelter was placed in the center of the arena (Figure 2), the shelter was made by placing two surfaces, non-porous (ceramic tile, vitrified side) and porous surfaces (wood tile), one on top of the other so as to create a gap of about 4 mm.

For each type of surface, 3 treated replicates and 3 control replicates were performed.

For each replicate, 5 insects (of mixed sex and stage) were used.

The insects are introduced 15 minutes before treatment, during this acclimatization period the light is left on so as to encourage the insects to stay inside the slit.

Before applying steam, the machinery used is prepared as per the directions below:

- Distilled water is introduced into the reservoir
- The gun with superheater is connected to the steam outlet of the machine
- The boiler is then turned on for about 5 minutes, the time to reach the required working operating pressure (165°C - 6 bar)
- Steam condensate is discharged into a container for about 1 minute in order to standardize the pressure of the machinery before application.

Once the minimum power of the machinery was set, steam was applied around the shelter for 4 seconds while maintaining a distance of 20 cm.

At the end of the application, the shelter was opened and the culling time was evaluated (Figure 3). The culled insects were then moved to a plastic container where 24-hour mortality was recorded.

The same procedure was performed, on shelters created with nonporous surfaces, using 10 eggs for each replicate. After application of the product, the eggs were moved to a plastic container and observed for one week to check for hatching or not.

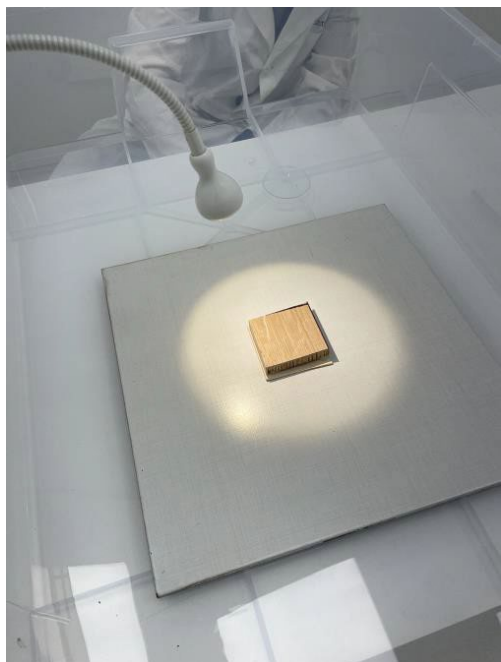


Fig 2. Shelter used for the test



Fig 3. Steam-treated bugs.



5. RESULTS

5.1 ADULTS AND NYMPHS

The following are the results obtained.

Tab. 1: Results for Wood surface

May 20, 2024	Treated (N. knocked down insects)			Mean (%) ± SEM	Control (N. knocked down insects)			Mean (%) ± SEM
Time (min)	Treat. 1	Treat. 2	Treat. 3		Cont. 1	Cont. 2	Cont. 3	
2	5	5	5	100.00±0.00	0	0	0	0.00±0.00
5	5	5	5	100.00±0.00	0	0	0	0.00±0.00
Alive 24 h	0	0	0	0.00±0.00	5	5	5	100.00±0.00
Moribund 24 h	0	0	0	0.00±0.00	0	0	0	0.00±0.00
Dead 24 h	5	5	5	100.00±0.00	0	0	0	0.00±0.00

Tab. 2: Results for ceramic tile surface

May 20, 2024	Treated (N. knocked down insects)			Mean (%) ± SEM	Control (N. knocked down insects)			Mean (%) ± SEM
Time (min)	Treat. 1	Treat. 2	Treat. 3		Cont. 1	Cont. 2	Cont. 3	
2	5	5	5	100.00±0.00	0	0	0	0.00±0.00
5	5	5	5	100.00±0.00	0	0	0	0.00±0.00
Alive 24 h	0	0	0	0.00±0.00	5	5	5	100.00±0.00
Moribund 24 h	0	0	0	0.00±0.00	0	0	0	0.00±0.00
Dead 24 h	5	5	5	100.00±0.00	0	0	0	0.00±0.00



5.2 EGGS

The following are the results obtained.

Tab. 3: Results for *Cimex lectularius*

June 04, 2024	Treated Tiles (N. of eggs hatched)			Mean (%) ± SEM	Control Tiles (N. knocked down insects)			Mean (%) ± SEM
	Treat. 1	Treat. 2	Treat. 3		Cont. 1	Cont. 2	Cont. 3	
N. of egg per replicate	10	10	10	100.00±0.00	10	10	10	100.00±0.00
Hatched 1 day	0	0	0	0.00±0.00	4	4	4	40.00±5.77
Not Hatched 1 day	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 2 days	0	0	0	0.00±0.00	7	7	8	66.67±6.67
Not Hatched 2 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 3 days	0	0	0	0.00±0.00	10	10	10	100.00±0.00
Not Hatched 3 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 4 days	0	0	0	0.00±0.00	10	10	10	100.00±0.00
Not Hatched 4 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 5 days	0	0	0	0.00±0.00	10	10	10	100.00±0.00
Not Hatched 5 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 6 days	0	0	0	0.00±0.00	10	10	10	100.00±0.00
Not Hatched 6 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00
Hatched 7 days	0	0	0	0.00±0.00	10	10	10	100.00±0.00
Not Hatched 7 days	10	10	10	100.00±0.00	0	0	0	0.00±0.00



6. CONCLUSIONS

Based on the tests conducted, the following was found:

Nymphs and adults:

In the treated replicates, for both surfaces tested, culling was 100% after 2 minutes and mortality 100% after 24 hours. No culling and mortality values were recorded in the control replicates.

Eggs:

In the treated replicates, no eggs completed hatching during the 7-day evaluation period.

In control replications (those not treated with steam), however, all eggs hatched after 4 days of observation.

Ponte San Nicolò,

Reported on **June 30, 2024**

Signed

Andrea Drago PhD