Technical specification Bio-Rodenticide

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1. Introduction

Field of invention

The invention is in the field of rodenticides based on botanicals. It prescribes a composition of botanicals processed in either powder or fluid form, combined with a bait of linseed. This product is embedded in a wider approach of ecological rodent control, focused on timely and collaborative effort.

Problem

Due to increased resistance of rodents to anticoagulant rodenticides, we need a game changer. With existing rodenticides in the markets there are several problems: 1) growing resistance of rodents, partly because of misuse, 2) poor planning of application (too late, not frequent, reactive), 3) sparse in coverage with negative side effects and 4) poor safety precautions. This gives rodent populations the chance to recover quickly, after which another investment has to be made to control rodents.

Despite the growing resistance to existing rodenticides and infestation of rodents increasing time and again, there is no botanical product for rodent control in the market as of yet.

Objective

The aim of this invention therefore is to offer a rodenticide product based on botanicals integrated within ecological rodent management. This can sustainably control rodent populations because of: effectiveness, larger coverage, timely and collaborative effort, eco-friendliness, longevity, safe to apply and no resistance build up (volatility).

This technical specification will detail on the bio-rodenticide itself, on ecological rodent management a manual is developed.

The bio-rodenticide is based on:

- ✓ Farmer's knowledge on local botanicals unfavourable to rats
- \checkmark Working mechanisms of the poisonous mechanism in the botanicals
- ✓ Biological volatility
- ✓ Sustainable to use, without negative side-effect to human and environmental health.

Requirements bio-rodenticide

- Effective in killing rats
- No negative side effects to other animals, humans and environment
- User-friendly
- Long term storage and working capacity (up to 12 months)

2. Background of the invention

A number of aspects have contributed to the situation where rodent infestation is ever increasing, existing chemical rodenticides are no longer effective and there is a lack of alternatives in the field of rodenticides.

More specifically:

1. There is increased rodent infestation in regions in Ethiopia, specifically Amhara, due to a soil and conservation management practices and change in agricultural practices. Providing rats

with places for shelter and more feed for longer time during the year. More land under cultivation also means less predators for rats. All in all, leading to explosive rat population.

- a. In Tigray, Ethiopia, farmer's damage varies from 9-44% on standing crops due to rodent damage
- b. New research indicates numbers of 20-50% of damage in Amhara, Ethiopia, to standing & stored crops due to rodent damage.
- 2. Existing chemical rodenticides, mostly based on anticoagulants, are no longer effective (misuse, immunization) or are no longer preferred due to negative side effects.
- 3. Hardly any innovations are taking place concerning botanical based rodenticides.
- 4. Farmers are innovating with local botanicals; this serves as the basis for the development of the bio-rodenticide.
- 5. This knowledge has been taken up by the developers of the bio-rodenticide in a process of research and development with extensive experiments both in the field as well as in the lab. In these experiments the botanicals in powder and fresh form were applied to the rodents baited with linseed, in a multitude of settings.
- 6. Throughout the experiments the effectiveness of the selected botanicals became evident. Showing mortality rats of over 80% in mixed form.
- 7. Based on this a selection on the most effective botanical treatments has been made for commercialisation.
- 8. This technical specification will describe a wider range of bio-rodenticides with botanicals and will single out one specific treatment mix to exemplify the exact procedures.
- 9. At the same time a wide range of these botanicals have the potential to be effective biorodenticides and are therefore considered.

3. Toxicity mechanisms and physiological effects on rats

This chapter will firstly give an overview of the toxicity of the botanical components. Further it describes the physiological effects to rats. From this a summary is provided linking the different components to the effects they have on rats.

3.1 Toxicity mechanisms

Each botanical has different toxicity mechanisms and levels. (this information is known, though not published here for non-disclosure reasons).

3.2 Physiological effects on rats

Find in Table 2 an overview of the physiological effects on rats after application of the botanicals.

	Overall indicators per organ for a control rat and treated rat			
Organ/ unit	Control	Treated	Meaning	
Liver	Bright red colour	Black colour	It is a first indication of poisonous content that entered the body. The liver is the organ which handles poison, so it means the liver had to process poison which explains the change in colour.	
Heart	Bright red colour	Black colour, sign of	Where the veins and the arteries join, blood is clotting. This is coagulation of blood. It means that blood clots come into the heart and block it.	

Table 2 overall indicators per organ for a control rat and treated rat

Stomac h and	Has food, big in size	blood clotting Empty or little	It is an indication of an abnormal situation in the blood, for this is normally not what you see on a freshly killed animal. It is almost impossible for blood to clot just after killing, so it is likely to be an effect of the poison. There are three possible reasons for the stomach and the intestines to be empty or small in size.
intest- ines	DIR III 2126	food, small in size.	 1 Suppression of food intake or appetite. 2 No access to food 3 The animal decided not to eat and die. *But it had the choice to eat pellets also, so this is very strange. Explanation could be that the rats have taken some of the treatment and that has resulted in a loss of appetite. So even with the choice, they have chosen to starve themselves. During the acclimatisation period they were eating the pellets and gaining weight. So if the stomach is empty and the amount of food remaining
			in the intestines is very small, it means that the animal only has few hours of being alive left. Because all the food in the intestines has run out and the animal will starve to death. Food in the intestines can only keep an animal for few hours, so it will starve.
Weight	Expect to gain weight or stay the same.	Expect to lose weight	Is a confirmation of reduced food intake and confirms the effect on stomach and intestines. It shows an overall impact on health, as normally rats should gain weight in these conditions.

3.3 Working mechanism

The compounds are mixtures of those that have the capacity to irritate skin and intestine (vesicant and purgative effect), hazardous effect when ingested directly, inhalation can cause irritation of the respiratory track, and powerful vesicating, toxic resin and carcinogenic.

Hence, few hours or days before death, the rodents treated with this bio-rodenticide mix show erected hear, shaking, reduced movement and reduced bait intake. When they die, they stuck their snout into the side or corner of cage, and appear abdomen flat out on the floor with four limbs spread. Post-mortem observations of individuals dead with a treatment mix revealed liver and heart with complete or partially darken colour (compared to the normal reddish colour of the control individuals) and often dark blood clotted in the heart. Also, the guts of the animals are either empty or with smaller quantity of food content compared to that of the control individuals which always appear filled with food. It is likely that all these physiological effects are the results of a combined effect of the active compounds of the two botanical.

(More information is known, though not published here for non-disclosure reasons.)

4. Selected treatment A

- 4.1 Selection reasons
 - Palatability by rats

- High effectiveness (mortality rate over 80% with 75% concentration)
- Low lethal dose, so lower concentration to be applied which favours rat to eat more.
- Low threshold-intake, meaning lowest amount of treatment needed to have a lethal effect.
- Effect is evidenced on vital organs (heart, liver, stomach and intestines)
- The effect is gradual in timing, not occurring on the first day, making it suitable for rodent control for the animals cannot make the association.
- Raw materials available, or with option to cultivate. Production is simple, low-cost and local.
- See Table 3 for overview of the selection criteria and assessment per treatment mix.

Table 3 Prototype criteria and selection

	Treatment A	Treatment B	Treatment C	Treatment D
Palatability	High palatability	low palatability	High palatability	Medium palatability
Effectiveness	High, mortality rate of 90%	Highest, mortality rate of 100%	Medium, mortality rate of 50%	High, mortality rate of 80%
LD50 (%)	52.16	62.25	94.27	120.22
Working mechanism	Overall poisonous impact on heart/liver, and suppressed appetite.	Overall poisonous impact on heart/liver, and suppressed appetite.	Overall poisonous impact on heart/liver, and suppressed appetite.	One component cannot be digested by rats, it solidifies in the guts and thereby disturbs the rats.
Availability of raw material	widely available with option to cultivate	Both can be produced as crop	widely available with option to cultivate	widely available with option to cultivate
Collection	Collection and harvesting	collection and harvesting	Collection and harvesting	Time-consuming
Processing	Good – materials are well suited for both mortar and pestle grinding as well as machine grinding.	Some parts are hard to crack – but possible with steel mortar and pestle, or specific grinder.	Easily chopped with blender	no processing required
Storage	when mixed as powder and linseed, can be kept for longer time	when mixed as powder and linseed, can be kept for longer time	Mould appears quickly on fresh produce	quickly becomes solid, making it difficult to properly mix

4.2 Production process

(This information is known, though not published here for non-disclosure reasons.)

4.3 Environmental safety

The effect of Treatment A on chickens is not lethal. The concentration of the treatment was set at 200%, doubling the highest possible concentration applied in the field.

Assessment on non-target effect of botanicals

In multiple day feeding tests, non-target effect of the botanicals on chickens was evaluated at 90 percent concentration above the higher concentration given to the rodents. Each day chickens were given 13.5 gram (90% concentration) of the botanicals along with pellets for consecutive four days. And the botanicals found to have no harm on the chickens (Table 4). All the chickens survived and no symptoms of sickness on the chicken were observed even after 48 h of post treatment period. The

chickens were further dissected to evaluate for possible physiological effect, however, all the organs checked were healthy compared to control.

Bio-rodenticide mixtures	Concentration (%)	Total consumed botanicals	Number of animals	Mortality	
		after 96h (g)	exposed	End of treatment after 96 h	Post treatment after 48 h
Treatment A	90	21.34	4	0	0
Treatment B	90	32.05	4	0	0
Treatment C	90	12.38	4	0	0
Treatment D	90	37.69	4	0	0

Table 4 total consumed botanicals at 90% concentration by chickens

All the botanicals found not to deter the chickens from feeding botanicals. Though the food consumed decreased after 24 hour, significant amount of botanicals were consumed (Figure 1). More than two fold of Treatment A that is required for killing 50% of the rodents have been eaten by the chickens during the experiment.

(More information is known, though not published here for non-disclosure reasons.)

Figure 2 revealed that the rats exposed to the botanicals decreased their weight during the experiment while the control continues to increase their body mass weight. However, it is evident that the rats recover after the experiment stops and gained weight during post treatment observation.

(More information is known, though not published here for non-disclosure reasons.)

5. The final product

The final product consists of:

- ✓ Bio-rodenticide packaged in resealable jar of a content of 500 gram.
- ✓ (More information is known, though not published here for non-disclosure reasons.)
- ✓ Each treatment is mixed with linseed in the right concentration: botanicals accounting for 52% concentration, and linseed for 48%.
- ✓ It includes a user manual in the local language. Explaining the farmers how to apply it in their fields.
- ✓ Local experts will be trained on best application of the bio-rodenticide and EBRM methods combined. Farmers will be able to request the help of these local experts.