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TECHNICAL REPORT

Sulfluramid is used in Brazil as active ingredient in the manufacturing of ant baits, for the control of leaf-cutting ants from the genus *Atta* (saúvas) and *Acromyrmex* (quenquéns), which are the insects that cause more injuries to national agriculture.

The leaf-cutting ants of the genus *Atta* and *Acromyrmex* are among the most important pests of the Brazilian agriculture, because their voracious attacks occur throughout the year and are spread to the entire country. The damages are immense, bringing losses to large and small crops, fruit and vegetable cultures, pastures and silvicultural areas. (MARICONI, 1970)

They are also considered the "number 1" pests among the reforesting companies.

In cultivated areas, young plants suffer the most with the attack of leaf-cutting ants, that is, soon after they are germinated or sprout.

In perennial cultures, these ants should be permanently controlled.

It is supposed that, even before Brazil was discovered, the leaf-cutting ants already represented a severe problem to the indigenous agriculture (the natives did not know how to fight them), and they were considered the determinant cause for the nomadism of indigenous in the lower areas of South America. (MARICONI,1970)

In Brasil, the losses caused by leaf-cutting ants have been mentioned since the century XVI.

Since then, several attempts were made for estimating the damages caused by this pest to various agroecosystems.

Just to give an idea of the leaf-cutting ants, it is enough to say that an adult colony can contain approximately 10 million ants, capable of cutting one ton of green leaves per year.

Sulfluramid was introduced in Brazil in 1993, after verification of its efficiency with many leaf-cutting ant species, replacing the active principle dechlorane (mirex). (FORTI et al. 2007; NAGAMOTO, 2007)

Sulfluramid, as used in the form of toxic baits, has full efficiency in the control of leaf-cutting ants. Since 1958, over 7,500 chemical compounds have been studied in many countries for ant control. Less than 1% of those 7,500 studied compounds have proven to be promising in such control. (FORTI et al. 1998)

For an insecticide to be used in the manufacture of ant baits, it shall feature a few unique characteristics that are essential for control efficiency (NAGAMOTO et al, 2004; FORTI et al, 2007)

- Acting by ingestion;
- Being odorless and non-repellent;
- Presenting a delayed toxic action;
- Being lethal at low concentrations; and
- Paralyzing cutting activities (losses caused by ants), right from the first days after its application.

Sulfluramid is currently the only registered active ingredient for the control of leaf-cutting ants, in the form of granulated toxic baits that incorporate all such characteristics. (FORTI et al., 2007)

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Baits based on Sulfluramid act by ingestion, rather than by contact. Therefore, worker foraging unable to perceive the active ingredient will distribute baits across the whole colony, reaching even the deepest chambers of the nest.

When carrying baits into their colony, ants licking the pellets and ingest small amounts of particles along with the insecticide, thus being intoxicated by ingestion.

The pellets disintegrate after being hydrated, thus enabling workers to incorporate their small pieces into the fungus garden. (MOREIRA et al, 2004b; NAGAMOTO et al 2007).

It has been evidenced that 50 – 70% of the colony workers will be contaminated by the insecticide within 24 hours. Such contamination will occur by direct ingestion of the active bait ingredient or by trophallaxis. This is a common activity in all colonies, whereby ants will exchange numberless substances between each other, such as enzymatic secretions, food, glandular substances, etc. To that end, an ant is capable of storing the ingested insecticide in its crop and later regurgitates it for other workers, thus producing a chain reaction (figures 1 and 2). (FORTI & BOARETTO, 1997)

Generalist workers and gardeners are those to be contaminated more quickly as a result of the activities they perform in fungus cultivation. Therefore, the insecticide must have a delayed action, i.e., it should not kill the ant too quickly and act at low concentrations, which is the way Sulfluramid operates.

The others insecticides, will immediately kill the workers thereby interrupting the trophallaxis chain (figure 2).

The mortality of ant workers causes an overall disorganization in the fungus garden, allowing for the growth of contaminating fungi that will render them improper for feeding ants and their queen. Without this fungus, all surviving ants and their queen will die by inanition, causing the whole colony to die in the next few days. (FORTI & BOARETTO, 1997)

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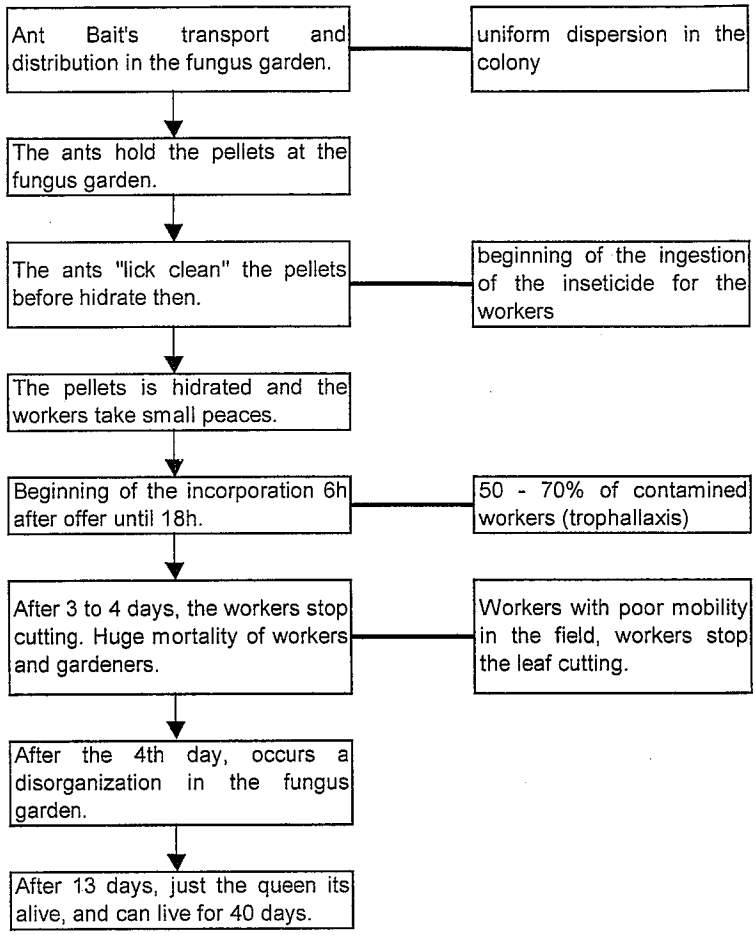

Figure 1.: Processing of the toxic bait and mode of action of sulfluramid in an *Atta sexdens rubropilosa* colony (adapted from: Forti, L.C.; Boaretto, M.A.C., Formigas Cortadeiras – Biologia, ecologia, danos e controle.1997, 61 p.).

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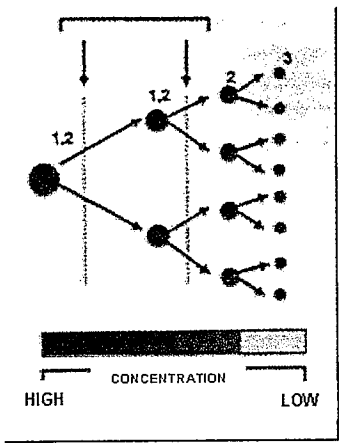

Figure 2.: Dispersion of an insecticide for trophallaxis and grooming, inside a leaf-cutting ant colony. (adapted from: Forti, L.C.; Boaretto, M.A.C., Formigas Cortadeiras – Biologia, ecologia, danos e controle.1997, 61 p.).

Several mechanical, cultural, biological and chemical methods have been studied as early as the 50s for controlling leaf-cutting ants. With the development of synthetic insecticides, chemical methods have been effectively used to control *Atta* and *Acromyrmex* ants. (FORTI & BOARETTO, 1997 ; BOARETTO & FORTI, 1997)

- MECHANICAL CONTROL

The mechanical control of leaf-cutting ants consists in excavating their nests for queen removal. Such a technique is no longer recommended for leaf-cutter colonies that are more than 4 months old, as this is when the queen will be lodged at depths exceeding 1.5m, thus rendering the technique unviable due to the great effort required.
In practice, mechanical control will be unviable in areas used for commercial plantations, in reforestation projects and grazing systems.

- CULTURAL CONTROL

Conventional soil preparation by plowing and harrowing could mean the mortality of newly formed *Atta* nests. However, with the practice of minimum cultivation adopted in several cultivars and reforestation projects, such a control has been virtually abandoned. For adult *Atta* nests, the end result could even be harmful, as soil mechanization could partially upset the anthill causing it to become temporarily inactive and giving the false impression of having been controlled.

The control of leaf-cutting ants through resistant plants, in rotation crop programs, or the handling of varietal composition of the cultivated area, could represent a beneficial strategy to the farmer for not implying additional costs, and to the environment for avoiding the risk of soil contamination by insecticides. However, no conclusive evidence has been found so far that could authorize recommendations in this sense. (BOARETTO & FORTI, 1997 ; MOREIRA et al 2004)

Several studies have been made to detect resistant materials in *Eucalyptus*, but as yet, results have been varied. For grazing systems, there is some evidence that some *Brachiaria* species are more resistant to *Acromyrmex landolti* and *Atta capiguara*, namely *B. decumbens, B. brizantha* and *B. humedicola*. The resistance mechanism seems to be associated with the inhibiting effect on the symbiont *A. landolti* fungus, as determined by *Brachiaria* species.

Nevertheless, there is evidence that the fungus cultivated by ants could metabolize toxic substances originating from such plants, rendering it more difficult to select resistant or low preference species.

Several researchers have attempted to identify vegetable species that would be toxic to leaf-cutting ants. Sesame (*Sesamum indicum*) has been investigated for use as a trap culture. However, planting this species between rows of *Eucalyptus* trees has produced no positive results. (BOARETTO & FORTI, 1997)

- BIOLOGICAL CONTROL

Natural biological control, through predators, parasitoids and pathogenic microorganisms (fungi, bacteria and viruses), is of importance in regulating leaf-cutting ant populations.

During swarms, predation by birds is very intense.

Spiders, acarids, several species of predating ants and beetles should also be mentioned. Among beetles, *Canthon virens* and *Canthon dives* are specific predators of leaf-cutting ant queens. The *C. virens* species attacks queens soon after swarm, prior to starting the penetration canal. However, we are dealing with an insect of complex biology, with a small number of progenies per female, such factors representing hindrances to mass creation aimed at field liberation.

The natural occurrence of entomopathogenic fungi has been noted in *Atta* and *Acromyrmex* ants. In Brazil, most studies have been conducted on *Beauveria bassiana* and *Metarhizium anisopliae* fungi. However, notwithstanding the high mortality results obtained in laboratory experiments, the efficiency results in the field are not conclusive. In addition to pathogens not being specific to the studied targets (as they attack other arthropods), leaf-cutting ants, along with their symbiont fungus, show defense strategies against parasites (including nematodes) and pathogens, of a morphologic, mechanical or biochemical nature. Quite often, anti-fungic glandular secretions are released by ants impairing the action of a pathogen. Furthermore, the behavioral characteristics of body cleaning, nest hygiene, changes in activity, pathogen recognition, etc., have resulted in a less successful biological control of leaf-cutting ants. (BOARETTO & FORTI, 1997 ; MOREIRA et al, 2004b)

- CHEMICAL CONTROL

Chemical control is the only method with available technology of practical use in controlling leaf-cutting ants, as a result of the aforementioned aspects of all other methods.(MOREIRA et al, 2004b)
In particular, chemical control strategies differ in their type of formulation and application directions.

- POWDER INSECTICIDES

- Dry powder products are insecticides formulated in solid vehicles (e.g.: talcum powder), to be applied with the help of manual equipment called dusters, provided with a conical container for product loading.
  The death of ants occurs by being in direct contact with the product, which must be applied into the nest entrance hole to reach the inside of the anthill.Currently used products are chlorpyrifos, fenthion, endosulfan and deltamethrin.
  The use of powder formicides faces strong limitations, such as the impossibility of product penetration into nest chambers of adult *Atta*, given its structural complexity. As yet not much known, the internal leaf-cutting ant colony structures could reach astonishing dimensions in terms of their number of fungus chambers, number and size of their channels (tunnels) and nest depth.
  Recent excavations in a colony of *Atta laevigata* led researchers into finding approximately 8,000 fungus chambers to a depth of up to 8 meters below ground level, with tunnels forming a real underground network, 3 or 4 of them converging to the colony, though rather wide, and reaching a width of 40cm by and a height of 2.5cm. (MOREIRA et al 2004a ; NAGAMOTO et al 2007)
  Therefore, it is virtually impossible for any formicides in its powder formulation to reach all fungus chambers in adult *Atta* colonies, thus being efficient to control small anthills only. (MOREIRA et al, 2004b)
  Moreover, it is important considering that a humid soil will render the application unviable (as the product will adhere to the soil) and usually leading to a phase separation (the active ingredient separating from talcum powder), resulting in irregular product distribution and, consequently, in a poor control efficiency. Another limitation is the need for having any loose earth removed 24-48 hours prior to insecticide application, resulting in a time-consuming and costly technique.
  The risks are high to both the environment and the operator.(BOARETTO & FORTI, 1997)

- LIQUID FORMICIDES
  In the past, very popular and much used for leaf-cutting ant control. However, in view of the low efficiency of already tested products requiring direct contact with ants, added to the costly work of nest boring and product losses due to soil absorption, they have been quickly replaced by toxic baits or thermal-fogging liquids. (BOARETTO & FORTI, 1997)

- THERMAL - FOGGING

Implies the generation of toxic "fumes" (droplets around 50 microns in size), using an insecticide in a mineral oil or diesel oil vehicle under heat, directly applied into the entrance holes of loose earth mounds by special equipment called thermal foggers. Two types of thermal foggers are available in the marketplace, one of them requiring no engine, therefore lighter, and the other driven by a two-stroke engine.

This method of application involves operational and economic disadvantages and brings risks to the operator health and to the environment. (CHERRETT, 1986 ; MOREIRA et al, 2004b)

Equipment operation is quite expensive as it requires constant maintenance, this being one of the main drawbacks against their viability.

It requires locating the nest entrance holes for product application, closing all other nest holes to prevent gases from escaping, which in practice is quite often impossible.

The product is heated to high temperatures to produce highly toxic gases, exposing the operator to high toxicity risks.The products used contain high concentrations of active ingredients.

The most used active ingredients are: chlorpyrifos, bifenthrin, deltamethrin and permethrin.

- TOXIC BAITS

Granulated baits represent the most widely used method for leaf-cutting ant control, consisting of a mixture an attractive (usually orange pulp and vegetable oil) and an active ingredient (insecticide), presented in the form of pellets.(CHERRET, 1986; FORTI et al 1988)

Highly efficient, this method features significant advantages over all other methods. This is a low-cost method, delivering high efficiency with reduced health hazards to humans and environment, being specific to the plague target. Its formulation is developed with extremely low concentrations of active ingredients, and its localized application do without application equipment. (CHERRETT, 1986 ; CAMERON, 1990 ; MOREIRA et al, 2004b)

Baits are directly distributed from their packaging, with no manual contact, close to active nest entrance holes or anthill trails and carried into the colony by the ants themselves.

Since its application is quite simple, presenting excellent control efficiency, the insecticide used in bait manufacturing will be of adequate quality.

An adequate insecticide used to formulate ant baits shall be lethal at low concentrations, act by ingestion present a delayed toxic action. Additionally, it shall be odorless, non-repellent, so as to be dispersed by trophallaxis to most workers in the colony. (FORTI et al , 1988)

Quick-action products will kill many ants in the first hours following their application, hindering or impairing insecticide distribution throughout the colony. They cause a certain disorder in the anthill and even temporarily stop some of its activities, such as leaf cutting. However, after some time, the anthill will be reorganized and will reappear in greater activity, causing even higher losses. (FORTI et al, 1988 ; FORTI et al , 2007)

Currently, the actives ingredients used in ant baits are: sulfluramid, fipronil and chlorpyrifos.

Sulfluramid is, among the actives ingredients, the only one with all features necessary for the good operation as an ant bait, which places it as the single efficient option to control leaf-cutting ants. (CAMERON, 1990 ; FORTI et al 2007 ; NAGAMOTO et al, 2007)

Comparative works demonstrate the low efficiency of toxic baits with the actives ingredients chlorpyrifos and fipronil (papers: "Eficiência das iscas Mirex-S Max, Blitz e Pikapau para o controle da Saúva *Atta capiguara* Gonçalves (hymenoptera: formicidae), em condições de campo" p.277-278 and "Eficiência das iscas Blitz, Mirex-S Max, Pikapau-s e Rainha no controle de *Atta bisphaerica* Forel (hymenoptera: formicidae)" p.295-297. Naturalia, V.24, 1999). (FORTI et al, 2003)

Additionally, the actives ingredients fipronil and chlorpyrifos present a much higher toxicity to mammals, water organisms, fish and bees than sulfluramid . (TOMLIN, C.D.S., 2000)

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