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Bait Evaluation Methods for Urban Pest Management

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<http://dx.doi.org/10.5772/53421>

1. Introduction

Baits are a preferred type of formulation used in urban pest management, especially for the control of cockroaches, ants, and increasingly termites. With precise placement in areas away from contact with human population, especially children, and a reduced rate of active ingredient (AI) application in a given structure area, baits are more economical and pose less risk for consumers and the environment than other formulations. However, baits are very difficult to evaluate for efficacy. For baits, pest acceptance and horizontal transfer of bait are essential in order to control pest populations.

Baits are composed of one or more insecticide active ingredients incorporated into an attractive food matrix, which varies according to the type of target pest, and even according to species within a certain pest type. Although commercial development of species-specific baits may represent a serious commercial problem due to limited market, this has been done in the past, for instance with imported fire ant baits in the USA. However, typically, baits are developed to target a group of similar insects, e.g., cockroaches, which may vary in their response to the bait formulation, resulting in varying degrees of control depending on the pest population composition.

In order to perform successfully, baits must attract the target insect and be ingested in sufficient amount that will cause the desirable level of control in the pest population. For non-social insects, such as cockroaches, transfer of the active ingredient among different segments of the pest population (e.g., adults and immature forms, reproductives, etc) is desirable but not necessarily an essential characteristic of the baits. However, in social insects (ants and termites) the transfer of the active ingredients between the foragers and the remaining of the population, and specially the reproductive caste, is essential in providing adequate control of the pest population within reasonable time.

2. Cockroach bait evaluations

Cockroach consumption of a bait and subsequent control can be complex. More than one of these cockroach pests may occur at a location with each having its own food requirements [1], susceptibility to insecticides [2], and aversion to certain bait formulations [3, 4]. Additionally, within each species, feeding patterns [5] and insecticide susceptibility [6] can vary among stadia.

When cockroach baits are placed close to harborage, they are usually in direct competition with other food and water resources. Therefore, baits, which are often gels with 40-60% moisture [7, 8], need to out-compete other sources of dry food as well as other moisture sources so cockroaches will consume them. Although cockroach control by baits is primarily due to bait consumption, not all insects within a population are actively seeking food, so not all individuals may consume a bait. Besides consumption, cockroaches are exposed to insecticidal active ingredients by contacting baits with antennae or palps. Contact exposure results in some toxicant transfer and mortality [9]. Also, cockroaches can be affected by contact with small amounts of translocated active ingredient (trampling), or when they consume contaminated feces (coprophagy), dead or dying cockroaches (cannibalism), or vomit (emetophagy) [10, 11, 12, 13]. These effects can result in secondary and sometimes even tertiary mortality [14].

The combination of consumption, contact, and secondary exposure results in mortality of various cockroach species and stadia (Figure 1).

Therefore, it is important to consider the consumption of insecticidal baits and the consequent ingestion of active ingredients, the mortality of different life stages within the pest cockroach population, and any possible effect of indirect to the active ingredient in the bait without actual bait consumption.

Consumption. In testing consumption in the laboratory, the use of mixed-age cockroach populations (adults of both sex and nymphs all in the same arena) is important to simulate bait consumption in natural infestations (Figure 2).

Weight-change controls, which are protected from consumption by the insects but otherwise under the same conditions as the bait being tested, must be used so adjustments can be made due to moisture change in bait and any other used in the experiments. To better understand potential differences between different products when consumed by different insect populations, it is important to estimate the consumption of bait in relation to the size of the insect consuming it. Bait consumption (B_{con}) per g of insect can be calculated as using the following equation:

$$\text{Consumption (mg)/g cockroach} = ((F_B - \{F_B * [(WC_B - WC_A) / WC_B]\} - F_A) / W_t)$$

where F_B is the weight of bait (mg) available to cockroaches at the start of the experiment, WC_B is the weight of weight-change control bait (mg) before the experiment, WC_A is the weight of weight-change control bait (mg) at the end of the experiment, F_A is the weight of

bait (mg) remaining after the experiment and consumption by the cockroaches, and W_t is the total weight of cockroaches placed in the arena.

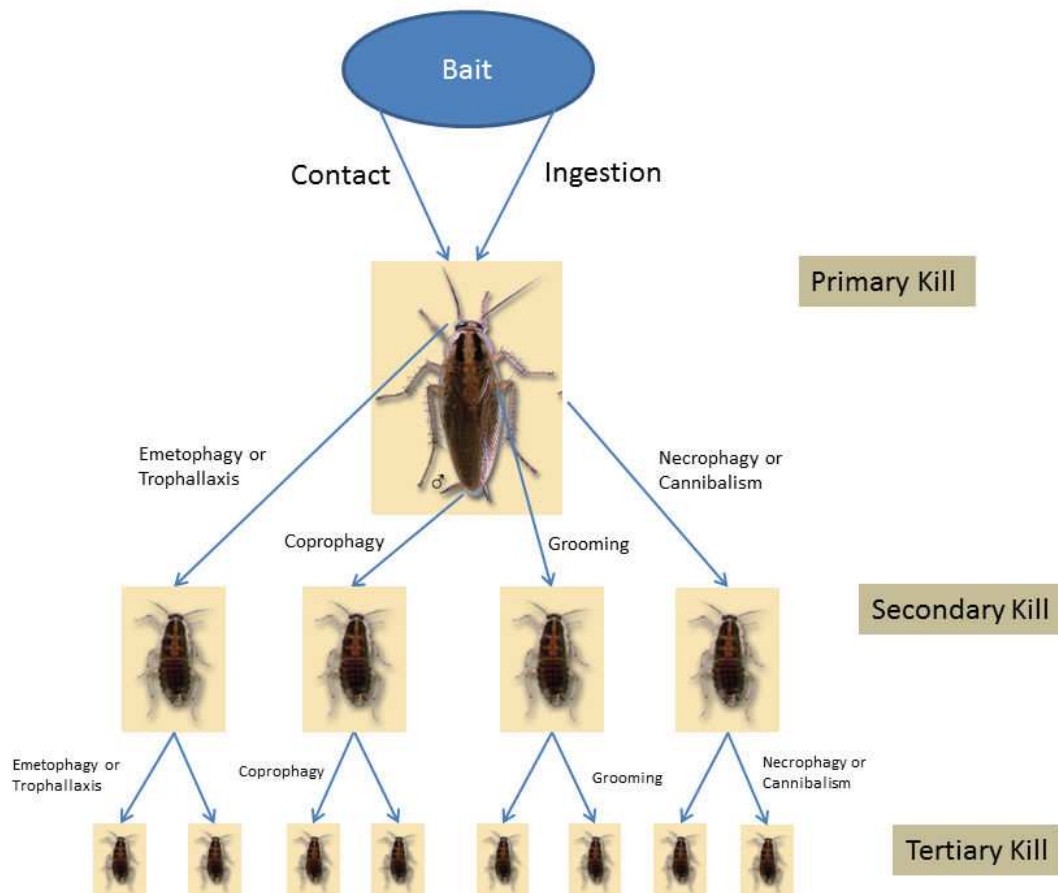


Figure 1. Cockroach baits can control populations in several ways, including primary kill, secondary kill, and tertiary kill.

Precise measurement of gel bait consumption by cockroaches is complicated by several factors:

- a. Cockroaches are likely to remove and spread bait that is never eaten, especially in small arenas with large insect populations. Although the contact with the bait without consumption can be an important element in producing the total cockroach mortality, it can cause the measurement of bait consumption to be very unreliable and variable among the different experimental replicates. The use of larger arenas with smaller cockroach populations and plenty of harborage areas away from the bait can help in minimize this effect.
- b. Rapid water loss cause gel bait especially, but also other bait forms, to change weight even in the absence of any consumptions. This factor is most severe on baits with very high water content, and formulations that do not limit water loss. Unless changes in

water content can be estimated precisely, consumption can be overestimated. Any other calculations that result from the bait consumption (active ingredient consumption, bait preference in relation to alternative food, etc) can be greatly affected by over or under-estimation of the bait. Also, an estimate of the amount of bait necessary for control of a population of cockroaches will be greatly affected by any miscalculations due to imperfect water loss estimates. Rapid water loss can also affect the palatability and nutritional content of the baits, which will greatly affect the bait's effectiveness as well as any measurements associated with its consumption.

- c. Differential consumption of food by different cockroach life stadia can vary over short periods of time. For experimentation, strict selection of insects within specific age groups can minimize any problems associated with the inclusion of insects that will not consume any bait, or any other food, for some time into the experimental period.

One solution to minimize some of the effects on measurements of consumption is to limit the measurement to a specific time period (e.g., 24 h following the initial exposure). Although water loss and trampling may be more severe during the initial hours after application, some of these problems can be resolved by using weight-loss controls. Limitations on how the cockroaches can reach the bait may be used to limit trampling on the baits but researchers must be careful in not limiting access to the bait, especially if bait stations are crowded.



Figure 2. Cockroach arena set up for bait evaluations. Harborage and water vials are on left, bait and untreated food is in center, and protected water controls are on right.

Active Ingredient Consumption. Because mortality is dependent on the actual amount of active ingredient consumed by the insects, it is important to determine that consumption (Figure 3).



Figure 3. Cockroaches leave harborage in choice tests and can choose to ingest either bait or untreated food, like laboratory chow.

Baits with lower active ingredient content may be more palatable to the insect and be consumed in relatively high amounts, while a bait with high active ingredient content may partially deter consumption by the pest population. The active ingredient (AI) consumption/g cockroach can be calculated as follows:

$$\text{AI / g cockroach } (\mu\text{g}) = B_{\text{con}} * C_{\text{ai}} * 1000$$

where B_{con} is bait consumption (mg)/g cockroach, and C_{ai} is the percentage of active ingredient in the formulated bait.

From the point of view of pest control, the delivery of the active ingredients is the most critical factor when using baits. Different active ingredient concentrations combined with varying bait palatability determine the total amount of active ingredient ingested by the pests and their consequent mortality (Figure 4).

In general, when consuming baits, individual cockroaches will consume more active ingredient than necessary to cause death. Depending on the bait product and the cockroach species, the quantity of active ingredient ingested can vary from just above what is needed to kill the insect to more than 1000 times the LD_{50} . This excess consumption of AI may be impor-

tant in avoiding or delaying development of insecticide resistance by killing virtually all cockroaches exposed to these baits [15].

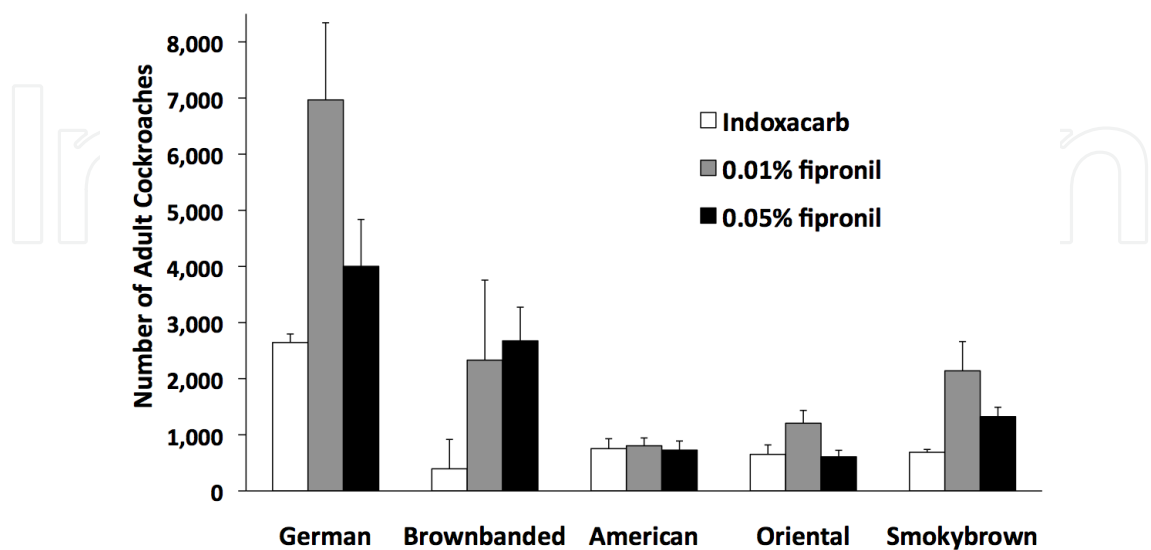


Figure 4. Number of adult cockroaches that could potentially consume and be killed by a 30-g tube of gel bait based on the consumption/g cockroach and the weights of average adult cockroaches from 5 different species (German: 56.7 ± 0.83 mg; brownbanded: 82.8 ± 0.49 mg, American: 959.4 ± 8.44 mg, smokeybrown 679.6 ± 19.02 mg, oriental 499.6 ± 9.51 mg).

Speed of kill is another aspect associated with the amount of active ingredient ingested by the cockroach. Although a quick kill can be advantageous in eliminating the pest problem within short time after the control action, other aspects, such as the possibility of transferring the active ingredient to other insects, can be maximized by limiting the amount of active ingredient that any individual cockroach will consume. Thus, understanding the different factors affecting the amount of active ingredient likely to be consumed by the average cockroach will help in the development of baits with greatest chances for success. Differences in susceptibility to insecticides [2, 6] and in feeding patterns [5] for different life stages of the pest, may also affect how fast bait materials will act. Adult males and non-gravid females are more likely to encounter and consume bait due to more consistent feeding patterns than nymphs.

Percent Bait Consumption. In the development of bait product, it is important that the bait competes well against other preferred food that the insect may find in their habitat. Thus, some measurement of bait preference over other foods is important in the development of baits. An important indication of how well a bait product will perform is the percentage of the total food consumption that is actually represented by the bait. The greater the percentage of bait in the total food consumption, the greater and faster mortality can be expected in the pest population. Percentage bait selection over an alternative preferred food can be determined by calculating the percentage of bait in the total food consumption, as follows:

$$\% \text{ Bait Selection} = [B_{\text{con}} / (B_{\text{con}} + AF_{\text{con}})] * 100$$

where, B_{con} is the bait consumption (mg) and $+ AF_{con}$ is alternative food consumption (mg).

Cockroach baits have been optimized for some of the most common pest species, and minor pest species may be much less attracted to the commercial baits. Preference for the baits may be due to a balanced mixture of moisture and nutrients specially given the high water need by most cockroach species. However, different species of cockroaches have different water needs and this will be reflected in the percent of bait consumed when alternative foods vary widely in water content in relation to the bait. With high water losses [16], high cuticular permeability [17], high metabolic rate [17], cockroaches need to balance moisture and nutritional needs [18] in order to survive and reproduce at optimal levels.

Secondary Effects. Secondary effects to the consumption of bait may be very important, especially in relation to the portion of the cockroach population that may not consume any of the bait. Gravid females and nymphs during the ecdysis may not consume the bait before dries up or is consumed by more aggressive bait consumers. The only way to affect the cockroaches that do not consume the bait directly is through secondary effects that result from consumption or other contact with bait contaminated debris, feces, and other materials.

To test these secondary effects, the arenas used in primary consumption studies should be set aside and remain unchanged except for the removal of live and dead cockroaches and any unused bait. Any remaining alternative food as well as harborages, water vials, containers, frass and any debris can be left in the arenas. These arenas can then be supplied with fresh food and water and a new population of cockroaches. Secondary effects (mortality) due to the contact with an environment contaminated by cockroaches consuming insecticidal baits can be evaluated against separate populations of cockroaches which are added to the contaminated arenas immediately after the primary consumption experiment and at different time intervals. A mixed population of cockroaches should also be used for these secondary effect experiments so that mortality in natural infestations can be simulated. Data similar to that collected in primary consumption experiments can be obtained in these experiments.

To reach the portion of the cockroach population that will not consume the bait, perhaps the best solution is the design of baits that offers greater opportunity for secondary mortality through contact with either dying insects or debris moved by insect that visit and consume the baits. The development of baits with these characteristics requires testing under conditions that maximize transfer of the material between segments of the cockroach populations.

Contact Effects With No Consumption. The effect of direct contact with bait without consumption can also provide better understanding on how different baits can affect cockroach populations. These experiments are difficult because they require the sealing of mouthparts in cockroaches so they cannot consume the bait. These experiments produce better results when the insect life stage used is sufficiently resistant to lack of water, or is placed in an ambient environment where water loss does not cause serious mortality in the test population. Adult male German cockroaches have been used in such experiment due to superior survivability without feeding.

Once their mouthparts are sealed using a droplet of melted paraffin wax or other non-lethal method, the cockroaches are placed into arenas containing pre-weighed portion of a bait and other materials used for the direct consumption experiments. Mortality and other parameters can be observed during a short period of time (2-5 days) while the insects survive despite the lack of food and water consumption. These experiments have short durations due to the need to evaluate treatment mortalities within the time period when mortality in the control insects is still within reasonable levels. Beyond 3-5 days, mortality in the control insects will increase rapidly, mostly due to the lack of water in insects with their mouthparts sealed, and any results will be heavily influenced by that the control mortality.

Contact mortality with baits, besides being difficult to document, may be of lesser importance in population control, especially because cockroaches cannot survive long without ingesting food or water. Contact mortality relies on cockroaches investigating the bait without consuming it as would occur with bait-averse populations [19, 20]. Certain active ingredients, such as fipronil, are more likely to cause higher contact mortality than others; however, differences in bait matrices may also cause varying levels of contact mortality [19], and some formulations may have the potential for killing high proportions of cockroaches by contact alone.

Although different pest cockroach species have varying food preferences and, within the same species, there is great variability in the amount of bait individual cockroaches may consume, baits remain the most efficient method for control of cockroaches. Although maximization of bait consumption must take priority in bait product development, other factors that enhance secondary mortality and contact toxicity must be considered. Evaluation of these bait products in relation to direct mortality, by bait consumption, as well indirect mortality, by secondary and even tertiary contact with the active ingredient in the bait, are also important in development and evaluation of cockroach baits.

3. Ant bait evaluations

The control of social insects using baits relies greatly on the fact that in these social colonies, mortality of the individual workers has little effect on the survival of the colony. It is only by removing the reproductives, or at least a sufficient number of workers and juveniles to directly affect the reproductive potential of the colony, that a pest ant colony can be eliminated. Some of the active ingredients used in baits formulated for ant control completely bypass any effect on the worker, and concentrate their power with specific chemicals that interfere with the reproductive potential of the queen or queens. Because the reproductive individuals in ant colonies do not normally gather food or consume material that has not been somehow prepared by other colony individuals, reaching the reproductives is the greatest obstacle for any active ingredient formulated in an ant bait.

Because ant workers do not ingest large solid particles, ant bait formulations that target most urban ant pests must contain a liquid component. An ant head dissected shows the structures that prevent solids larger than 0.5 microns from being ingested (Figure 5). Food

enters through the mouth and passes into the infrabuccal pocket. The infrabuccal pocket is a location for food particles too large to swallow. Food in the infrabuccal pocket passes through the buccal tube that is lined with setae that serve as filters. Particles too large to pass through this filtering mechanism remain in the infrabuccal pocket. These food particles can later be transferred to larvae for that ingest and digest these particles. Liquids that are ingested pass through the buccal tube into the pharynx and down the esophagus to the crop and midgut for storage and digestion. For baits to be ingested by urban pest ants they are usually liquids or granules that are soaked with liquid baits.

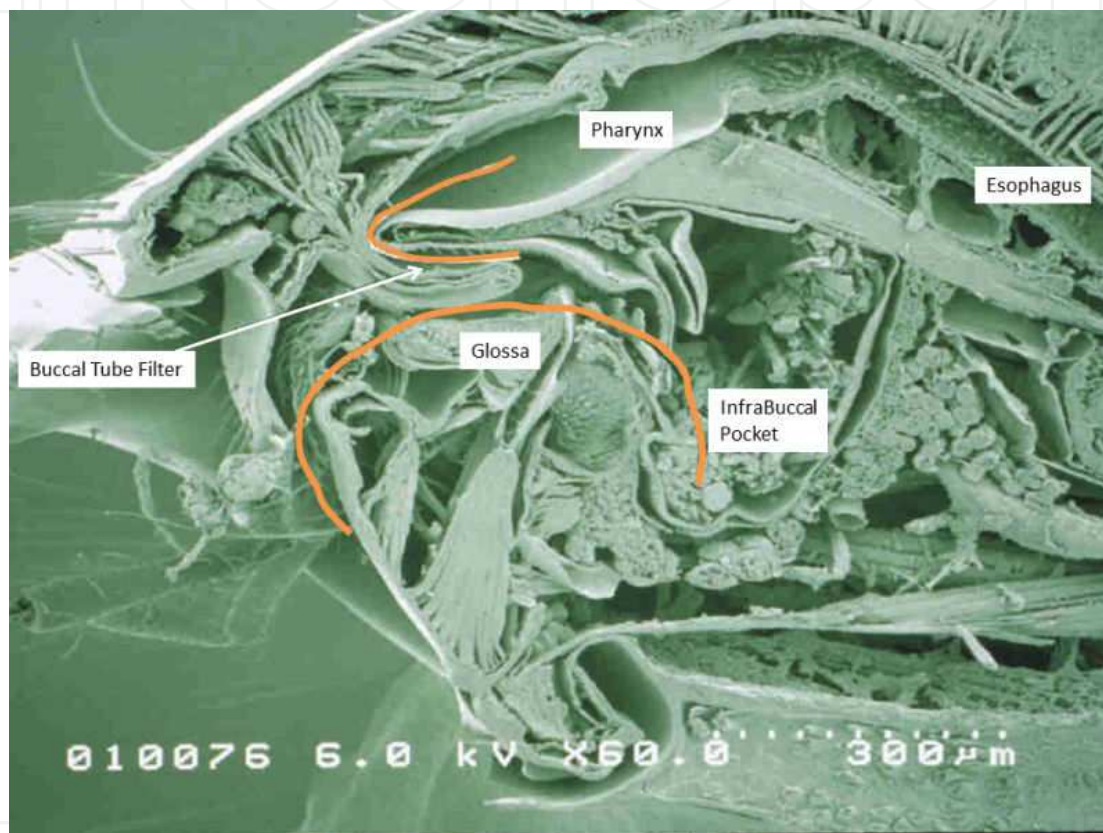


Figure 5. Cross section of an carpenter ant head showing the structures associated with ingestion of food.

Baits that target fungus-gardening ants target ants in a very different way, and are therefore develop following a completely different model. Most urban ant baits come in liquid, gel or granular formulations, but the granular formulation consists of a matrix containing a liquid that can be removed by the ants. Gel baits are only appropriate for indoor use or other special situations where protection from the climatic conditions is possible. Liquid baits normally require application into a holding device which the ants will have access to the bait. Granular baits are more practical for application outdoors, although they are also convenient for indoor applications.

Baits work by taking advantage of ant biology and behavior such as social grooming and trophallaxis. Once the bait is discovered, the foraging ants pick up the bait and transport it

back to the colony. The brood, especially late instars, may be important in the digestion solid bait particles into a liquified form that can be transferred to workers and reproductives in the colony. The amount of brood in the colonies, in the laboratory and field, could be responsible for the foraging preference. Fourth instar larvae do most of the protein digestion in the ant colony [21, 22] and their presence in a colony can change ant foraging preference to proteinaceous materials.

It is through food sharing that the toxicant in the bait can be transferred to the rest of the colony. Because the bait is picked up directly by the ant workers and is later shared within the colony, relatively low amounts of the toxicant can be used in targeting a pest ant population [23]. Ant foragers are usually the older workers that first pick up or consume the bait. They share the toxicant with other workers and queen tenders, and eventually after 3-4 days the toxicant reaches the queen, which affects reproduction in the colony. Even if the queen dies, eggs may hatch, larvae may pupate and develop into workers. Eventual control of a large colony may take 1-5 months.

The current baits out on the market for ant control include gel baits, liquid baits, and solid granular baits. A liquid or gel bait is usually one that requires a bait station and constant reapplication due to the elements and are usually used with ants that display mass recruitment to food sources.

In many cases there is little distinction between liquid and solid baits in terms of what the ants actually harvest in the field, as in the instance of popular fire ant baits. Fire ant baits consist of oil placed on a carrier. Foraging worker ants only feed from liquids so workers only remove oil off the bait granule. Thus the granule serves as a vehicle for the toxicant and attractant but it may not be carried into the nest at all. The active ingredient will enter the colony as a liquid.

Because granular baits can be broadcast over larger areas, this is the preferred formulation to reach most of the ant species. Granular baits take advantage of foraging patterns of different ant species. Granular baits consist of attractants, a carrier and active ingredients [24]. Four characteristics are important in a granular bait: 1) delayed toxicity, 2) easy transfer among individuals in the colony, 3) non-repellent active ingredient, and 4) attractive formulation for the target ant species [25, 23].

1. **Delayed toxicity:** In most ants, only a small percentage of the worker population actively forages outside of the nest. The use of active ingredient with delayed toxicity can guarantee maximum distribution of the bait within the colony before the ants start showing signs of toxicity. If there is enough delay, the active ingredient will be fed to larvae and reproductives before foraging and food sharing activities are shut down in the colony. This guarantees mortality of different castes within the nest, and the elimination of immatures.
2. **Easy transfer:** This should be applied both to the bait itself, so that it can be handled by a maximum number of individuals within the colony, but especially to the active ingredient. With fast and easy active ingredient transfer, most of the colony can receive a le-

that dose of the active ingredient before initial gatherers are affected and start showing toxicity effects.

3. **Non-repellency:** Because baits rely on the pick up and transfer by workers in the ant colony, non-repellent materials will be more easily masked within the bait formulation. Repellent active ingredients could also be used if they could be sufficiently masked by formulations components so not to prevent rejection of the bait by foragers.
4. **Attractive formulation:** The first step in the bait use process is attracting foragers so they are enticed to seek the bait and carry it back to the nest. Attractants added to the bait can overcome other deterrent characteristics of the bait or avoid any defense behavior that would normally prevent ants from returning toxic components to the nest. A great deal of work is dedicated to examining ant preferences to bait components so formulations can be picked up preferentially by the ants in an environment that will likely have many other food sources.

Several aspects of the granular bait formulation should be considered during the development of new products, including granule composition and size, attractive additives, and active ingredient.

The ideal granular bait should contain granules of similar size that can be easily applied to areas when needed. The carrier of the active ingredient is the most important part of the granular formulation because both the particle size and the materials and components used determine the spreading characteristics, the effectiveness of recruitment and removal of the bait, and the residual life of the active ingredient [26].

Size of the granular carrier may determine the size of ants that can be targeted with a particular formulation (Figure 6). In general, smaller ant species prefer smaller particle sizes to larger ones when given a choice [27]. If the particle size can be matched to a particular target pest ant species, this can increase the efficacy of the granular bait. Ants can normally carry granules with size that roughly match the size of head of foraging workers, but in some species, ant workers may collaborate in carrying larger pieces. Also, ants may subdivide larger bait particles before carrying the bait back to the nest. However, for baits developed for different ant species, this size matching may not be a preferred option.

A large granule size is more convenient because more active ingredient can be added to a larger particle size, allowing more active ingredient to be introduced into the colony with fewer particles collected by the foragers. Ant foraging normally fits what has been described as the optimal foraging theory [28, 29], which states that ants should take the biggest pieces of food particles that they can carry, in order to increase their net energy intake per unit of effort (Figure 7).

However, the difficulty in transport by the ants navigating the larger granular bait into the nest must be considered, especially for ant species that do not cooperate during foraging.

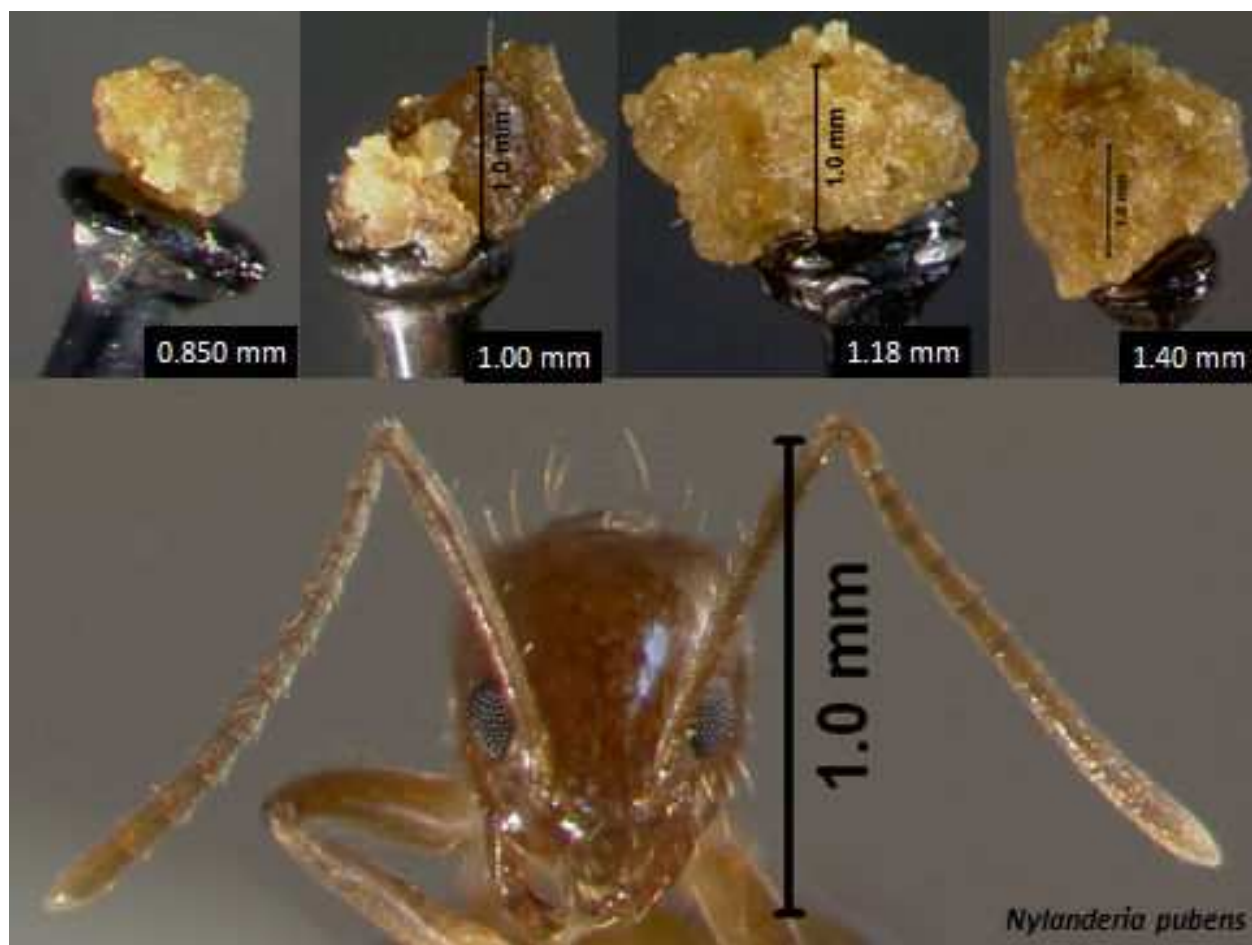


Figure 6. Workers head, *Nylanderia fulva*, in comparison with dog food granules used in size preference experiments.

Because different ants have different requirements for protein, lipids and sugar, and these requirements are likely to vary throughout the year or the life of the ant colony, the composition of the granule can be critical in bait development. Two approaches have been used in the formulation of granular baits for ants: a) use of non-nutritious granule to which food attractants are added, b) use of food particles as the granule matrix to which the active ingredient is added. With either approach, the quality of the food attractant determines which ants are attracted to the bait. Although several bait compositions use sugars as the only attractant, some products have been formulated with protein and lipid attractants, and even insect tissue.

A non-nutritious granule normally used in formulating ant baits is de-fatted corncob, a byproduct from corn processing [30, 24]. It is capable to absorbing relatively large amounts of the liquid additives such as the oil used in many fire ant baits. Fine granules from dog food or other animal diets serve as a nutritious granule for ant baits because it fulfills ant nutrient requirements, is easy to prepare in a uniform granular size and it readily absorbs additives.



Figure 7. *Nylanderia fulva* foraging in a laboratory setting on 1.00-mm (Top) and 1.40-mm (Bottom) dog food granules used in size preference experiments.

The addition of insect tissue [31], which attempts to mimic the natural diet of many ant species, adds complexity to the bait but also to the production process, with inevitable cost consequences. The use of a readily available byproduct, such as silkworm pupae can facilitate production and cut costs. Other insects that can be mass reared at low costs, such as crickets or waxmoth larvae, are also interesting alternatives. Laboratory reared crickets ground into a slurry with addition of small quantity of water have been used in our laboratory to be added to dog food granules tested as a bait to tawny crazy ant (*Nylanderia fulva*).

Traditional baits for *S. invicta*, consist of oil (attractant) on corn cob matrix (carrier) [30, 24]. On the other hand, baits that contain proteins and carbohydrates are very attractive to species such as *L. humile* and *Paratrechina* spp. that are not attracted to the lipid-based fire ant baits [24].

In order to enhance a carrier, it is important to know the ant species food preferences, based on field observations, laboratory experiments, and reports in the literature on similar ant species. Observations of feeding habits such as ants feeding on honeydew from aphids, plant nectaries and insect tissue [32], tending to aphids and mealy bugs [33] can serve as clues in the development of ant baits. Preference studies indicating the balance between components in the ant diets and other aspects of ant nutrition [21, 34, 35, 36, 37, 38, 39] also offer valuable clues that can help the development of new ant baits.

Ant Bait development. Bait development should be initiated with preference tests in the laboratory (Figure 8), but should quickly move into field tests due to the great variability food preference and gathering between laboratory and field populations of the same ant. Laboratory colonies are usually fed a constant diet that is rich in all nutrients need by the ants, while field ant populations are more likely to go through periods when their diet is relatively low in certain nutrients or components such as live insects or sugars. Differences between controlled environment in the laboratory and more variable environment in the field lead to differences in foraging behaviors preference to bait components [40, 41, 42, 43]. Differences in the presence and proportion of different developmental life stages in the colony [44] can also be important factors in determining differences between laboratory and field results.

Field tests should also be conducted at different times of the year in order to characterize the bait preference and effect given different levels of foraging on a specific formulation throughout the season. Because foragers need to move very little between the nest and the foraging arena in laboratory colonies, food choices may be different from those for field ants which usually will travel much further from the nest both in scouting for new food sources and in foraging trails. Distance between the nest and food source can affect choice and quantify of food gathered by an ant colony.

In the laboratory, arena preference tests with multiple bait choices can be used in the elimination of candidate formulations. Later tests with limited choices can be used later to further define preference for specific formulations. Careful design of the foraging arena will avoid preference biases for baits that are found more readily.

If using colony fragments in foraging experiments, careful attention to the size and composition of these experimental colonies is important to preserve the foraging behavior and other characteristics that match those of the full colonies (Figure 9). For instance, foraging of different baits can be drastically affected by the presence or absence of brood, and the brood age structure in an ant colony.

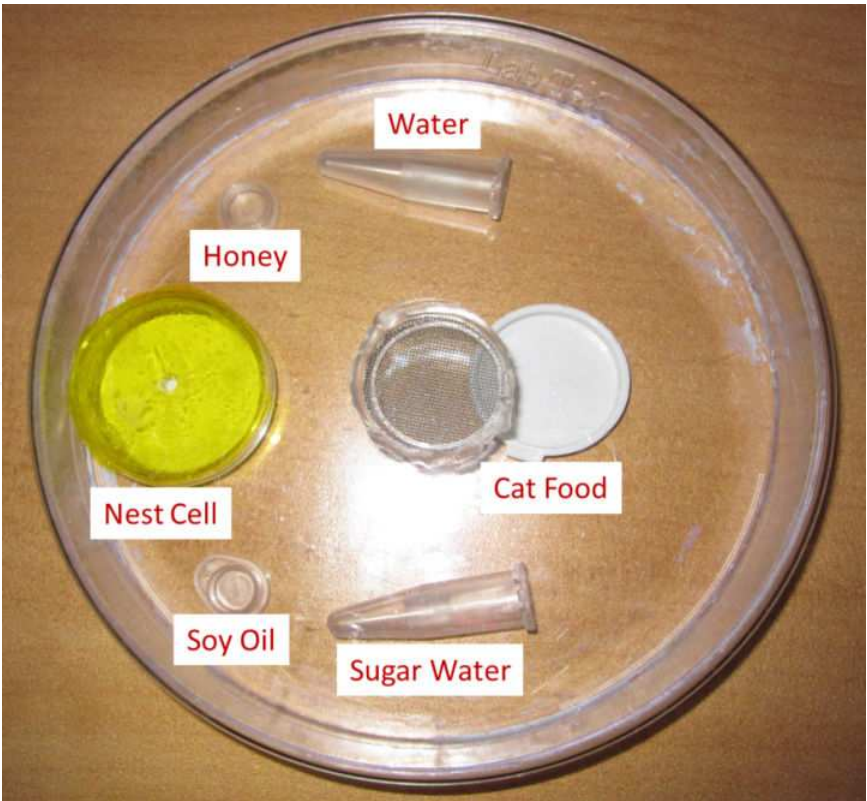


Figure 8. Testing arena used for experiments on *Nylanderia fulva* using granular bait matrix applied with active ingredient.

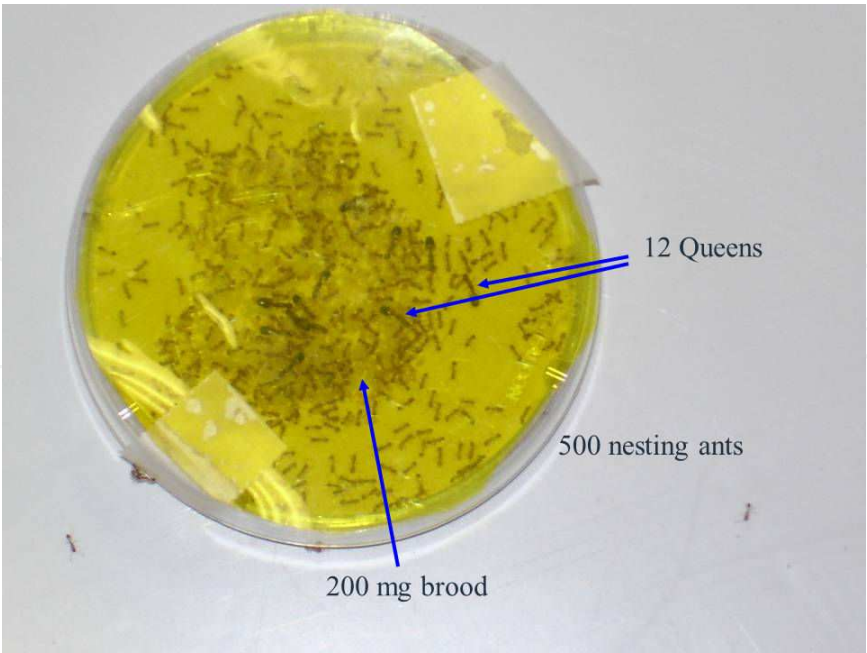


Figure 9. Colony fragments of Pharaoh ants are set up in cells containing brood, workers and queens.

Eventually, bait development must include field experiments to determine the fate and efficacy of the material when applied in situations for which the bait is designed. Careful observations on ant behaviors associated with finding, gathering, and moving the bait material to the ant colony are useful in understanding potential shortcomings of the developed products. Use of fields in different locations that represent the variety of situations where the ant baits can be used is essential in defining clearly the effects of different parameters and factors on the performance on the ant bait.

The data collected from the different experiments will vary, and should be adjusted for each ant species and situation. However, at minimum, the data should allow estimation of the quantity of material necessary to eliminate pest ant colonies, and the total mortality after different periods of time. Of particular importance, is the effect that bait may have on reproductive individuals. In polygyne colonies, it is especially important that the bait achieves maximum distribution within the colony, so that reproductive individuals throughout the colony can be effectively controlled.

4. Termite bait evaluations

Methods of subterranean termite exclusion and prevention of structural infestations have broadened from soil termiticides and barrier treatments to include monitoring and baiting systems. Baiting systems have increased in registration and use since the introduction of the first bait 18 years ago (tradename Recruit, Dow AgroSciences LLC, Indianapolis). The specificity and mode of action of these active ingredients requires much less product to be applied to the environment. Hexaflumeron was the first active ingredient (AI) registered in the United States to be used in a termite bait formulation and there are currently several other AI in use, all of which fall into two classes: insect growth regulators (IGR) and energy production inhibitors. Both classes are considered to be slow-acting and rely on foraging termites to transfer small amounts of consumed bait material throughout the colony through contact, trophallaxis, grooming, fecal consumption and cannibalism. Baiting systems using IGRs are intended to be used as stand alone treatments. Bait formulations with AI affecting energy production are used in conjunction with soil treatments. There are baits designed to be used in-ground to prevent structural infestation and others for use above-ground in areas with known termite activity. A successful baiting system should be proven to affect termite populations (Figure 10).

Active ingredient evaluation. A non-repellent, lethal and slow-acting active ingredient is required for a termite bait to be effective. When evaluating a potential bait toxicant one must first determine the toxicity of it towards the termite species it will be used against. For example, at what concentration will it kill 90% of exposed termites (LC_{90})? How long does it take for that 90% to die (LCt_{90})? Termites are highly social and do not fare well when kept in small numbers so great numbers of exposed termites should be placed together

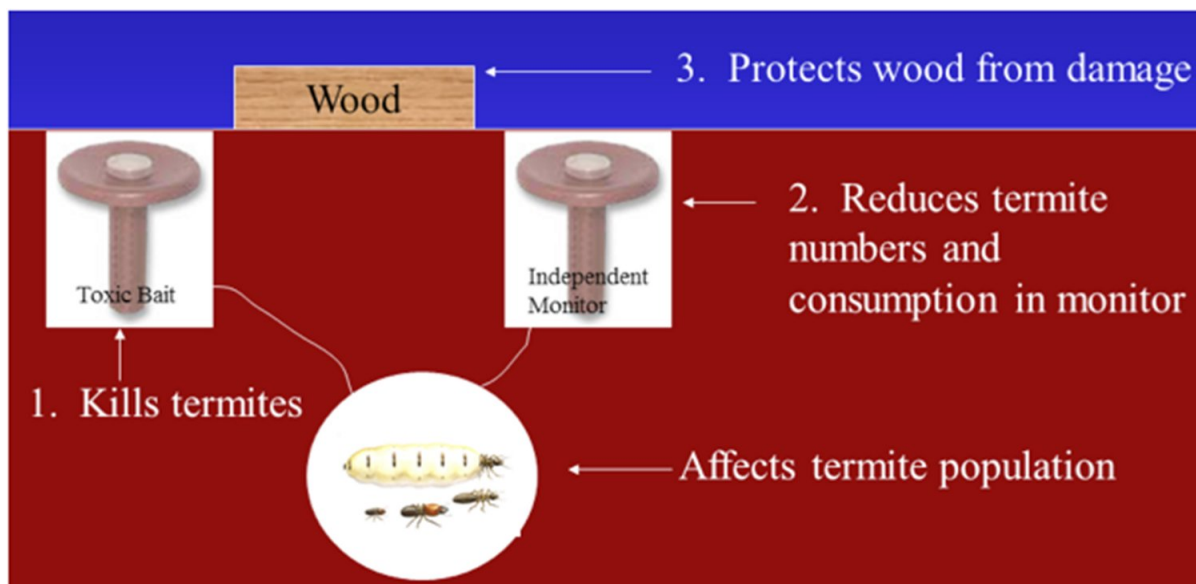


Figure 10. Termite bait efficacy data can document termite populations are affected in 3 ways.

Once the ideal bait toxicant concentration has been determined it must be tested to determine whether it will be readily consumed or show a feeding deterrent effect when adhered to wood or other cellulose containing matrix.

Bait formulation evaluation. The formulation of a palatable bait matrix is essential to the success of a baiting program. In the natural environment of subterranean termites there are a lot of potential food sources providing competition for baits. If there is no preference shown towards a bait it will not be very successful when installed below ground. Above ground baits must be more palatable because the termites present already have a source of wood that they are consuming. Impregnating wood or another cellulose containing material with active ingredient is the most common formulation method for commercially available baits.

Choice tests can be used to determine termite feeding preferences and any foraging biases. In a laboratory setting one can prepare a trial arena filled with moistened soil (10% wt:wt). Each dish will contain one piece of untreated wood and one piece of bait matrix, both of which have the same moisture content, dimensions and orientation within the arena. One thousand subterranean termites are introduced in a Petri dish with a small opening covered by a piece of filter paper at the end. Once the filter paper has been consumed the termites will be free to tunnel and forage throughout the arena. Locations of food choices should be randomized to eliminate any directional biases. Repeating this experiment multiple times with multiple colonies will show which food is preferred by termites. Which source did the termites consume the most of by weight? Was the first food source contacted the only one consumed or was there cross over between both food sources? Statistical analysis will indicate if termites prefer the bait matrix to untreated wood. This simple assay can be repeated and altered to include multiple wood and bait choices, different soil types and moisture levels.

Baiting system evaluation. Using a baiting program to prevent structural infestation may take considerably longer to be effective than soil treatments and thus the methods used to evaluate them are different. In order to gain registration, evaluations of termite baits must ultimately fulfill requirements set forth by state government guidelines (*Florida: 5E-2.0311 Performance Standards and Acceptable Test Conditions for Preventive Termite Treatments for New Construction*). Stand-alone baiting systems must be tested and meet specific requirements in field plot and building tests.

Independent Monitors	Building Monitoring	Reinfestation of Buildings
• >90% reduction in termite activity	• Cessation of live termite activity	• Visual inspection showing no reinfestation within 2 years
• >90% of test buildings protected	• >90% of test buildings protected	• Research and visual inspection showing no reinfestation within 1 year
• Protection within 12 months of initiation of feeding on bait active ingredient	• Protection within 12 months of initiation of feeding on the formulated bait	

Table 1. Performance standards for stand-alone termite baits in structures with existing infestations.

Evaluation of below-ground baits: field plot tests. Once it has been determined that feeding on bait has started, infested field plot tests require a reduction in each termite population by at least 50% or a reduction of wood consumption at independent monitors by a minimum of 50% in at least 75% of baited population colonies within one year.

The minimum required thresholds must be maintained for at least 6 months. In order to meet this requirement one can place monitoring stations, which have the same shape, appearance and moisture levels of a baiting station but contain untreated wood instead of bait, throughout a field plot or around a structure known to contain termites. Monitoring stations containing untreated wood are installed in the ground in augered holes at consistent intervals (every 10-20 feet).

Monthly inspections of monitoring stations will continue until live termites are found. Monitoring stations without termites are not switched to bait stations until live termites are found. When live termites are found monitoring stations are deemed ‘active’, wood will be replaced by a bait tube containing active ingredient and termites contained in the wood will be placed in the bait station. Plastic bucket traps (with uniform holes allowing for termite entry) containing wooden blocks may be placed around these stations and checked monthly. The purpose of these bucket traps is to be able to count and assess the nature of termite activity and chart differences over time. Commercially available bait stations require different monitoring intervals but for evaluation it is recommended to be conducted monthly to better determine when control has been achieved.

During monthly inspections, the number of termites present and the amount of bait consumed will be recorded. Bait matrix consumption is typically a visual estimate of the percent consumed as bait weights can be misleading. If baits are completely consumed, compromised or damaged they will be replaced with new bait. Once termite presence and bait consumption ceases monitoring resumes and monthly inspections will continue for at least 6 months. If monitoring stations are found to be active a new bait tube will be installed. The amount of active ingredient consumed can be measured at the end of the study by drying the bait and comparing initial and post-treatment weights using the percentage of active ingredient by weight in the matrix formulation.

The question of whether a colony has been eliminated or merely suppressed can be difficult to answer and may require months to years of monitoring before and after a baiting system is put in place. A suppressed colony will exhibit a period of inactivity in which no termites will be found in monitoring stations yet eventually recover and continue foraging [45]. To better determine the level of control achieved it is recommended to use cuticular dyes and genetic markers as detailed below.

Detecting presence of multiple colonies and foraging areas. Once a monitoring station is attacked by termites they will be collected and keyed to species level. If one is interested in determining the number of colonies present and their respective foraging areas in a field site there are two options: mark-release-recapture or cuticular dyes. Mark-release-recapture involves collecting live termites at monitoring stations, bringing them into the laboratory and feeding them filter paper impregnated with fat-soluble cuticular dyes such as Nile Blue A, Sudan Red 7B, or Neutral Red (Fisher, Pittsburgh, PA) [46]. Once termites are dyed they are placed back into the stations from which they are collected. Mark-release-recapture will also help in estimating population sizes.

A less obtrusive method involves placing a cellulose matrix impregnated with cuticular dyes, Nile Blue A or Neutral Red, in bait tubes which will allow for long-term tracking of termites from station to station. This eliminates the need for termites to be handled. Once the termites are dyed and back in test site the procedure is the same. During monthly monitoring of stations the locations and numbers of dyed termites can be recorded and a map of foraging activities produced [47]. Both methods can be enhanced through the use of genetic markers to help differentiate between colonies [48, 49, 50].

Evaluation of above-ground baits: building tests. Evaluation of above-ground baits can only be conducted in buildings with active subterranean termite infestations. Mud tubes are broken and both monitoring and bait stations are installed in line with the disturbed tube. Weekly monitoring is recommended because bait toxicants are introduced to the colony very quickly. Bait consumption will be visually estimated as a percentage and should be replaced if too much has been consumed or the bait has been compromised. If baits are too dry water may be added but too much water will become a deterrent to consumption. Termites will be counted in monitoring stations but not removed. Once feeding on bait and monitoring stations has ceased, baits will be replaced with monitoring stations.

Above-ground baiting programs must show $\geq 90\%$ reduction in termite activity in $\geq 90\%$ of test buildings within one year from the initiation of bait consumption. A successful above-ground baiting program must show that there has been no re-infestation within one year after activity has ceased. This must be verified by combining a visual inspection with termite detecting tools including infrared devices, moisture meters, radar, chemical detection, bath trap inspection ports, canine detection or fiber optics. The alternative is to wait until two years after the last evidence of termite presence and conduct a visual inspection of the site.

5. Summary

Baits have many advantages for use in urban environments. The advantages extend from use in IPM programs, to non-impact of humans who are living and living or working amid infestations, and to advantages associated with controlling the pests (Table 2).

IPM	Human	Pest Control
<ul style="list-style-type: none">• Preserve beneficial organisms involved in biological control	<ul style="list-style-type: none">• No odor	<ul style="list-style-type: none">• Slow acting
<ul style="list-style-type: none">• Reduced risk	<ul style="list-style-type: none">• Lower exposure to pesticides	<ul style="list-style-type: none">• Non-repellent
<ul style="list-style-type: none">• Can be used in sensitive areas	<ul style="list-style-type: none">• No mixing needed	<ul style="list-style-type: none">• Attractives and phagostimulants to enhance consumption
<ul style="list-style-type: none">• Long lasting	<ul style="list-style-type: none">• Less preparation prior to pesticide application	<ul style="list-style-type: none">• Secondary mortality by transfer
<ul style="list-style-type: none">• Application as point sources		<ul style="list-style-type: none">• Long lasting
<ul style="list-style-type: none">• Less active ingredient		<ul style="list-style-type: none">• Transfer of AI within pest population
<ul style="list-style-type: none">• Narrow spectrum of insects controlled		<ul style="list-style-type: none">• Translocation• Overcome insecticide resistance

Table 2. Advantages of baits in pest management in urban environments in relations to IPM principles, humans and pest control.

Insecticides used in urban environments are almost always in proximity to people, pets, and food. As a result, the safety of products used and efficacy of the formulation in urban pest control are of extreme importance. People can be affected by the use of a wrong formulation, or buildings can be destroyed when ineffective products are used. As a result of screening active ingredients and formulation, a variety of insecticides have been developed for urban pest management (Table 3). Most of the active ingredients are listed by the USEPA as reduced risk products. As reduced risk, there is an expedited registration process for baits containing these actives.

Type/Active Ingredient	Mode of Action	Pest Groups
• Oxadiazine Indoxacarb	• Sodium channel blockage	• Cockroaches, ants
• Neonicotinoid Imidacloprid Dinotefuran	• Acetylcholine receptor stimulation	• Cockroaches, ants, flies
• Spinosins Spinosad	• Acetylcholine receptor stimulation	• Flies
• Phenylpyrazoles Fipronil	• GABA receptor blockage	• Cockroaches, ants
• Avermectins Abamectin Emamectin	• Glutamate receptor stimulation	• Cockroaches, ants
• Chitin synthesis inhibitors Hexaflumuron Noviflumuron Diflubenzuron	• Block chitin formation	• Termites
• Amidinohydrazone Hydramethylnon	• Inhibit energy production	• Cockroaches, ants
• Pyrroles Chlorfenapyr	• Inhibit energy production	• Termites
• Borates Boric acid Sodium borate Disodium octaborate tetrahydrate	• Non-specific metabolic disruption	• Cockroaches, ants, flies

Table 3. Active ingredients, modes of action, and pests controlled with baits used for pest management in urban environments.

Baits have become one of the most popular formulations used by pest management professionals for use against cockroaches, ants, and termites. One of the advantages of bait formulations is that they are usually ready to use, in low concentrations, and can be placed only where and when needed. Hazards of using baits is minimized by using child-resistant bait stations or careful placement directly into harborages. The use of baits requires more time than spraying and costs may be higher because of the use of food-grade ingredients in the formulation.

Overall, baits are a very effective and successful insecticide formulation for urban pest control. As a result, the industry has been expanding testing and screening programs for label expansions so insects other than ants, cockroaches, and termites can be controlled.

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