Perspective

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Insecticide resistance, control failure likelihood and the First Law of Geography

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Abstract

Insecticide resistance is a broadly recognized ecological backlash resulting from insecticide use and is widely reported among arthropod pest species with well-recognized underlying mechanisms and consequences. Nonetheless, insecticide resistance is the subject of evolving conceptual views that introduces a different concept useful if recognized in its own right – the risk or likelihood of control failure. Here we suggest an experimental approach to assess the likelihood of control failure of an insecticide allowing for consistent decision-making regarding management of insecticide resistance. We also challenge the current emphasis on limited spatial sampling of arthropod populations for resistance diagnosis in favor of comprehensive spatial sampling. This necessarily requires larger population sampling – aiming to use spatial analysis in area-wide surveys – to recognize focal points of insecticide resistance and/or control failure that will better direct management efforts. The continuous geographical scale of such surveys will depend on the arthropod pest species, the pattern of insecticide use and many other potential factors. Regardless, distance dependence among sampling sites should still hold, following the maxim that the closer two things are, the more they resemble each other, which is the basis of Tobler's First Law of Geography.

Keywords: insecticide resistance survey; insecticide resistance diagnosis; area-wide survey; resistance level and intensity; insecticide resistance management

1 A FABLE: GOD, BEETLES AND INSECTICIDES

God, recognized by some as being inordinately fond of beetles,[1] would, according to others, have said on the 8th day, 'I changed My mind about insects', and invited a chemical company representative for a chat.[2] Although Haldane's irony matched against Brunner's sarcastic witticism makes for an interesting fable of the genesis of insecticide, the adaptability of insect populations allows them to withstand human interventions aimed at their control, an instance of what is sometimes referred to as ecological backlash.[3,4] A pivotal example is the phenomenon of insecticide resistance, which is a key response to arguably the most influential arthropod management tool in use since the 1940s – pesticides!

2 INSECTICIDE RESISTANCE

2.1 Evolving an influential scientific concept

Although the first reported case of arthropod resistance to pesticide dates from over 100 years ago in the 1910 s[5] – before the dawn of organo-synthetic pesticides and the worldwide use of chemical pest control in the 1940s – the earliest formal definition of insecticide resistance comes from the World Health Organization (WHO) in the late 1950s. A WHO panel of experts defined insecticide resistance as 'the development of an ability of a strain of insects to tolerate doses of toxicants which would prove lethal to the majority of individuals in a normal population of the same species'.[6]

This early conceptual definition of insecticide resistance has proven useful and recognizes insecticide resistance as a population-based and relative phenomenon evidenced by variation in responses to insecticides among arthropod populations of the same species. However, this definition does not clearly recognize the genetic basis of the phenomenon as a response to insecticide selection and, while not doing so, does not distinguish nor recognize the role that epigenetic and phenotypic adaptations can play in the expression of insecticide resistance. Regardless, the WHO definition of insecticide resistance is certainly valuable when focusing on the ecological and physiological phenomenon without considering its practical field implications.

A clear recognition of the genetic basis of insecticide resistance in its definition was soon incorporated by Crow[7] and further refined by Sawicki[8] as 'a genetic change in response to selection by toxicants that may impair control in the field'. Besides clearly recognizing the genetic basis of resistance and selection for resistance, this later definition also associates insecticide resistance with the potential, not certainty, of insecticide control failure, which is an important pest management and economic (practical) consequence of insecticide resistance. Indeed, this definition recognizes insecticide resistance as a microevolutionary phenomenon and an ecological backlash resulting from insecticide use and overuse.

Current methods for recognizing insecticide resistance are largely founded on these concepts: (i) using dose/concentration–response curves (and LD_{50}/LC_{50} estimates) to compare same-species populations and determine the magnitude (i.e. level

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or ratio) of insecticide resistance; and/or (ii) using diagnostic doses/concentrations to determine the frequency of resistant individuals in a given population; or (iii) using some variant of these aiming at recording low-frequency resistance (e.g. F_1 and F_2 screening) or incidence of the prevailing insecticide resistance mechanisms in individual insects constituting discreet populations.[9−12] Resistance to insecticidal toxins such as *Bacillus thuringiensis* (*Bt*) toxins can be considered in parallel to insecticide resistance, and the same conceptual framework applies.[13]

Concern regarding insecticide resistance potentially impairing field insecticide efficacy, or control failure, was also stated in Sawicki's concept.[8] However, insecticide resistance and control failure were not treated as synonymous, but as one potential cause (the former) and a potential consequence (the latter), while several alternative factors may also lead to control failure (e.g. unsuitable insecticide application, weather conditions, etc.), in addition to insecticide resistance. This distinction invites further investigation attempting to address both notions, either by directly correlating laboratory and field results or by mimicking field applications under more standardized conditions.[9] Therefore recognition of control failure due to insecticide resistance or the assessment of control failure likelihood remains a challenge.

2.2 An industry-based conceptual framework

The relevance of the phenomenon of insecticide resistance spans academic and industry research, stretching to pesticide users and agrochemical company marketing. In fact, the agrochemical industry has been playing a rather proactive role in insecticide resistance research and outreach in an effort to monitor, understand and manage the phenomenon. The establishment of the Insecticide Resistance Action Committee (IRAC) is a reflection of this attitude integrating industry efforts on the subject of insecticide resistance.[14] Despite the convergent interest among the different groups concerned with insecticide resistance, each has their own particular focus in their priorities and views. Thus an alternative and also influential definition of insecticide resistance is championed by IRAC: 'A heritable change in the sensitivity of a pest population that is reflected in the repeated failure of a product to achieve the expected level of control when used according to the label recommendation for that pest species'.[15]

The IRAC concept of insecticide resistance is convergent with Sawicki's regarding the genetic and microevolutionary basis of the phenomenon. However, IRAC reinforces even further the notion and connection between insecticide resistance and control failure. The distinctions among these concepts of insecticide resistance are addressed elsewhere,[16] but both introduce the notion of insecticide control failure, which is an important practical consequence of insecticide resistance and possible to estimate experimentally. First though, a definition of control failure likelihood (or risk) by an insecticide is necessary, and it can be understood as *the frequency of resistant individuals from which the resistance to an insecticide becomes an economic problem, i.e. when its efficacy is significantly compromised*.

The control failure of an insecticide due to insecticide resistance is based on the significant reduction of efficacy of a (commercial) product (i.e. an insecticide formulation) used at its recommended dose/concentration but not reaching an expected control level. Herein lies a difficulty, since the monitoring of control failure likelihood due to insecticide resistance is inferred, but not directly performed, and may be important for recognizing if insecticide resistance is indeed translated into a field problem and how serious the problem is, if it does exist. Growers and technicians rely on practical field observations to (empirically) assess control failure, but an experimental approach is necessary to assess the risk of likelihood of control failure, recognize its dimension as a problem, and map its occurrence.

Sawick's and particularly the IRAC conceptual criterion for insecticide resistance do have appeal and value because, among other things, they clearly recognize the practical problem of control failure, an ultimate and rather undesirable consequence of insecticide resistance. Therefore it is an important notion and its monitoring is welcome. The above reported effort allows the estimation of a discriminatory dose/concentration for the subsequent monitoring of insecticide resistance that considers the natural existing variability of response among distinct field populations. Nonetheless, such care does not directly assess the risk of control failure (or control failure likelihood) due to insecticide resistance, which is a consequence of the frequency of resistant genotypes, the intensity (i.e. magnitude, strength, level or ratio) and mechanism(s) of resistance associated with these genotypes, but considering the label rate and a minimum threshold of field efficacy.[9,17]

That said, it should also be recognized that the industry also explores strategies of regional market deployment and education, besides the baseline susceptibility studies, when launching a new insecticide or, more precisely, a new insecticidal mode of action. This allows industry to deploy resistance management tactics proactively and region-wide seeking to minimize the likelihood of insecticide control failure within a given timeframe. Resistance monitoring can then target particular areas and contexts where there is greater perceived risk of resistance development.

2.3 Estimating the control failure likelihood of an insecticide

The assessment of insecticide resistance using methods that realistically simulate field exposure is an important initial step in establishing methods for assessing control failure likelihood due to insecticide resistance. The bioassay methods have shifted, evolving from the earlier focus on topical exposure to current surface contact and/or ingestion exposure procedures, which are more realistic. These bioassay methods, like the ones recommended by IRAC,[15] allow for a couple of different approaches to estimate the likelihood of insecticide control failure. Both approaches require the use of commercial insecticide formulations and their respective label rates, besides a minimum required level of insecticide efficacy based on the country's regulatory reference required for insecticide registration. The need for this minimum insecticide efficacy threshold to assess control failure by an insecticide was earlier recognized by ffrench-Constant and Roush,[9] who provide a few sound examples, but which were not subsequently explored.

One method used to assess control failure likelihood was initially applied for the tomato borer *Tuta absoluta*.[18] This approach compares the estimated LC_{80} (i.e. lethal concentration for 80% of the insect population) with the recommended and registered label rate of the insecticide to recognize control failure. If the estimated LC_{80} were higher than the label rate of the commercial formulation, control failure would probably take place. Here we used 80%, as per Silva *et al.*,[18] because this is the minimum efficacy threshold required in Brazil to allow registration of a conventional (organo-synthetic) insecticide.[19] The same criterion was used in a subsequent study with the same pest species, the tomato borer, in the context of the European Union.[20] The same rationale is potentially useful for crop plant expression toxins of *B. thuringiensis* against susceptible insects, but requiring suitable minimum efficacy thresholds for the purpose.[21,22]

The method described above to estimate control failure likelihood exhibits the shortcoming of requiring the establishment of dose/concentration–response curves to estimate the LC_{80} or analogous parameter (e.g. LD_{80}, EC_{80}, etc.). Therefore the method is labor-intensive and exhibits relatively low resolution, limitations similar to the detection of insecticide resistance using dose/concentration–response curves and LC_{50} or LD_{50} estimates.[9] These limitations led to an alternative approach, which was also earlier proposed and recently employed, once again for the tomato borer.[23,24]

The alternative approach – subsequently developed to estimate the risk or likelihood of control failure by an insecticide (i.e. the control failure likelihood) due to insecticide resistance – again uses the recommended label rate of the insecticide and the minimum efficacy threshold for insecticide registration (e.g. 80%). In this case, however, the label rate is used as the discriminating concentration and the mortality achieved in the bioassay is used for comparison with the minimum efficacy threshold; this allows for the recognition of the populations likely to undergo control failure with the tested insecticide.[22]

The actual control failure likelihood can be estimated by multiplying the achieved mortality (%; when subjected to the label rate) by 100, dividing the product by the minimum required efficacy (%) and subtracting the result from 100. Thus control failure likelihood (CFL) = 100 − [achieved mortality (%) × 100]/expected mortality (e.g. 80%), where an achieved mortality equal to the expected mortality (or minimum required efficacy threshold, e.g. 80%) provides a CFL = 0; achieved mortality values above the minimum efficacy threshold will provide CFL values < 0%, indicating a negligible risk of control failure (or control failure likelihood). A desirable next step in such estimations of control failure likelihood, and also for insecticide resistance, is the recognition of the spatially dependent scale of the phenomena allowing for geographically based decision-making in resistance management.

3 SPATIAL PERSPECTIVE

A considerable amount of research effort is devoted to surveys of insecticide resistance among field populations of arthropod pest species. Some of these surveys are comprehensive, involving a few dozen populations, others just a few (<20). A quick survey of the scientific literature indicated 441 papers on the subject (Web of Science, 29 February 2016; search terms: insecticide resistance AND survey). In a few instances, a worldwide effort is involved, as exemplified by the late 1960s survey of insecticide resistance among stored product insect pests,[25] and in particular the ongoing effort with malaria vectors – with mapped records since 1954 – being spatially displayed according the *Anopheles* species and insecticide of interest.[26] There is no denying the importance of these surveys in allowing the recognition of focal areas for priority management. However, the scale and scope of these surveys can and should be improved to enhance the decision-making potential for resistance management. Therefore the recognition of an underlying spatially dependent relationship among sampled populations is an intuitive initial step towards this objective.

3.1 Tobler's First Law of Geography

The idea of spatial proximity favoring resemblance is intuitive and deeply ingrained. Such a notion was later formalized by Prof. Waldo Tobler in 1970 as 'Everything is related to everything else, but near things are more related than distant things',[27] which became known as Tobler's First Law of Geography, or the First Law of Geography for short. This is a fundamental concept of spatial dependence that allows the use of spatial interpolation to estimate the distance of influence between sampling points (and sampled populations). Besides such estimations, spatial mapping of insecticide resistance can be carried out allowing the recognition and determination of the extent of focal areas of concern for appropriate intervention. The same notion is also applicable for control failure likelihood.

The use of spatial surveys to monitor insecticide resistance and/or control failure likelihood imposes important requirements that influence the design of these surveys. An important requirement is the sampling strategy, which should be extensive within a continuous area and distributed in a way that attempts to maximize the spatial interpolation and detection of spatial dependence, which is potentially affected by the pest genetic diversity, agricultural landscape and season, among other factors. While the sample distribution will be affected by the context in which the sampling takes place, the minimum amount of samples required is usually high, above 50 (reaching up to 100 depending on the circumstance). As consequence, the number of sites required for field insect sampling is relatively high, higher than the usual amount used in current surveys of insecticide resistance, regardless of the scale of species and population distribution that are determinants of the spatial scale of sampling. However, the survey is based in diagnostic concentrations and will likely profit from local existing interest and capacity of assistance minimizing its eventual costs and allowing reliable geographical extrapolation of the status of the phenomenon of insecticide resistance, and the concern regarding existing control failure. This provides a proper geographical scale for management intervention to circumscribe, or preferably eliminate, local problems; if this is not possible (owing to political boundaries for instance), this will also be recognized by the survey, and a proper management scale will be recognized.

3.2 Area-wide spatial surveys

Less than 8% of over 440 papers published on surveys of insecticide resistance clearly expressed concern about the spatial scale of the phenomenon (Web of Science, 29 February 2016). Nonetheless, the totality of the papers does exhibit such underlying spatial concerns although none of the research particularly considered and quantified the spatial scale of the phenomenon. Again, the efforts with stored product insects and malarial mosquitoes are a response to these concerns where the focus of insecticide resistance was properly mapped, but the spatial dependence among the samples was not estimated. Therefore extrapolation of the interfering distances involved is not possible, nor is the quantification of the spatial scale of the phenomenon for management purposes. A recent exception was the country-wide survey of temephos resistance in yellow fever mosquitoes carried out in Brazil.[28]

The organophosphate temephos remained the mosquito larvicide of choice for controlling the yellow fever mosquito in Brazil from the 1980s until 2011, and the monitoring of temephos resistance in the country started in the late 1990s.[28] The monitoring was performed using discriminating concentrations to estimate the frequency of resistant individuals in each sampling site, following the standardized procedures of the World Health Organization.[29] The data set was gathered by the then established National Network of Insecticide Resistance Monitoring in *Aedes aegypti* (MoReNAa), under the initiative of the Brazilian National

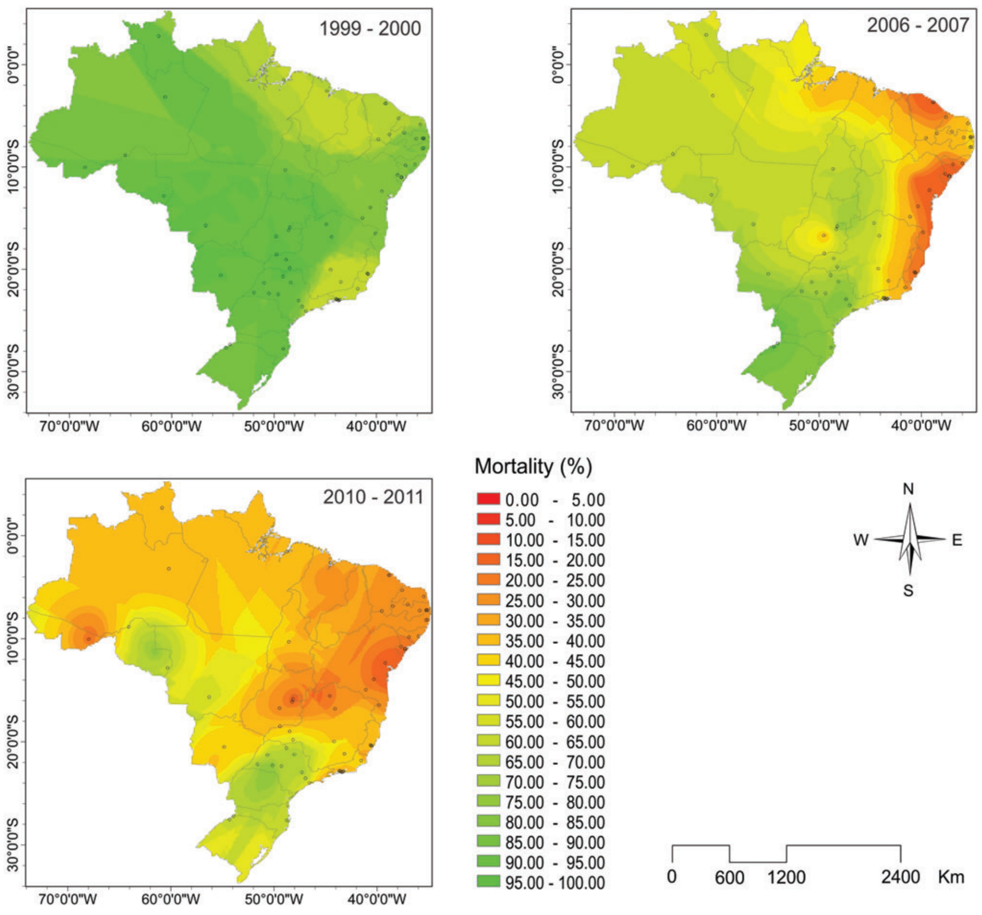

Figure 1. Contour maps and map of sampling sites of temephos resistance in Brazilian populations of the yellow fever mosquito *Aedes aegypti* generated through spatial interpolation. The color legend indicates the represented range of mortality (%) of mosquito larvae obtained in the temephos resistance diagnostic bioassays. The open circles indicate the sampling sites. The sampling sites were sparse in the regions of north and mid-north Brazil with low mosquito occurrence, which prevails in higher-density urban conglomerates characteristic of coastal areas and particularly in northeast and southeast Brazil. (Modified from Chediak *et al.*[30])

Program of Dengue Control (PNCD; Office of Health Surveillance) of the Ministry of Health (Brasília, DF, Brazil). Although not designed nor carried out with the objective of spatial interpolation to generate country-wide mapping of temephos resistance, the data gathered allowed for this procedure. The temporal and spatial spread of temephos resistance in the yellow fever mosquito in the Brazilian territory greatly increased during the 12 year survey, and temephos resistance was prevalent among the mosquitos in the country and, as of 2013, compromised its use against these insects in Brazil (Fig. 1).[30]

A similar effort is under way for whiteflies, also in Brazil, but the surveying area is much smaller, with an interfering distance of less than 10 km, and encompasses parts of two central counties with intensive tomato production under central pivot irrigation. Likely more interesting though is the Brazilian effort in surveying control failure likelihood in the Neotropical brown stink bug *Euschistus heros*. The survey area is the state of Goiás in central Brazil and the vial bioassays are from IRAC (IRAC Susceptibility Test Method 030).[31] The initial results regarding imidacloprid efficacy and control failure likelihood indicate that the problem is rather localized and should be easily dealt with at the focal area (Fig. 2), allowing a better management of the situation. The more limited effort focusing on the tomato borer *T. absoluta* did not allow for the mapping of insecticide resistance and control failure likelihood, but allowed initial estimates of interfering distances for the phenomenon, which is observed in a 300–350 km range in the Brazilian savannah.[23] Nonetheless, even this partial information is useful in delimiting the spatial scale for the implementation of insecticide resistance management tactics.

4 CONCLUDING REMARKS

Surveys of insecticide resistance are a major feature of the scientific literature on the phenomenon with important implications for its management. The Arthropod Resistance Database jointly maintained by Michigan State University and IRAC reinforces this perception.[32] However, the prevailing conceptualizations of insecticide resistance likely prevented the recognition of the importance of distinguishing and surveying not only insecticide resistance but also control failure due to insecticide resistance.

The distinction between insecticide resistance and control failure likelihood is helpful in stimulating the assessment of both, but even when the concern with control failure prevails or merges with that of insecticide resistance, the survey is still possible, but should be performed differently than currently carried out. In this case, control failure estimates should be the focus of attention,

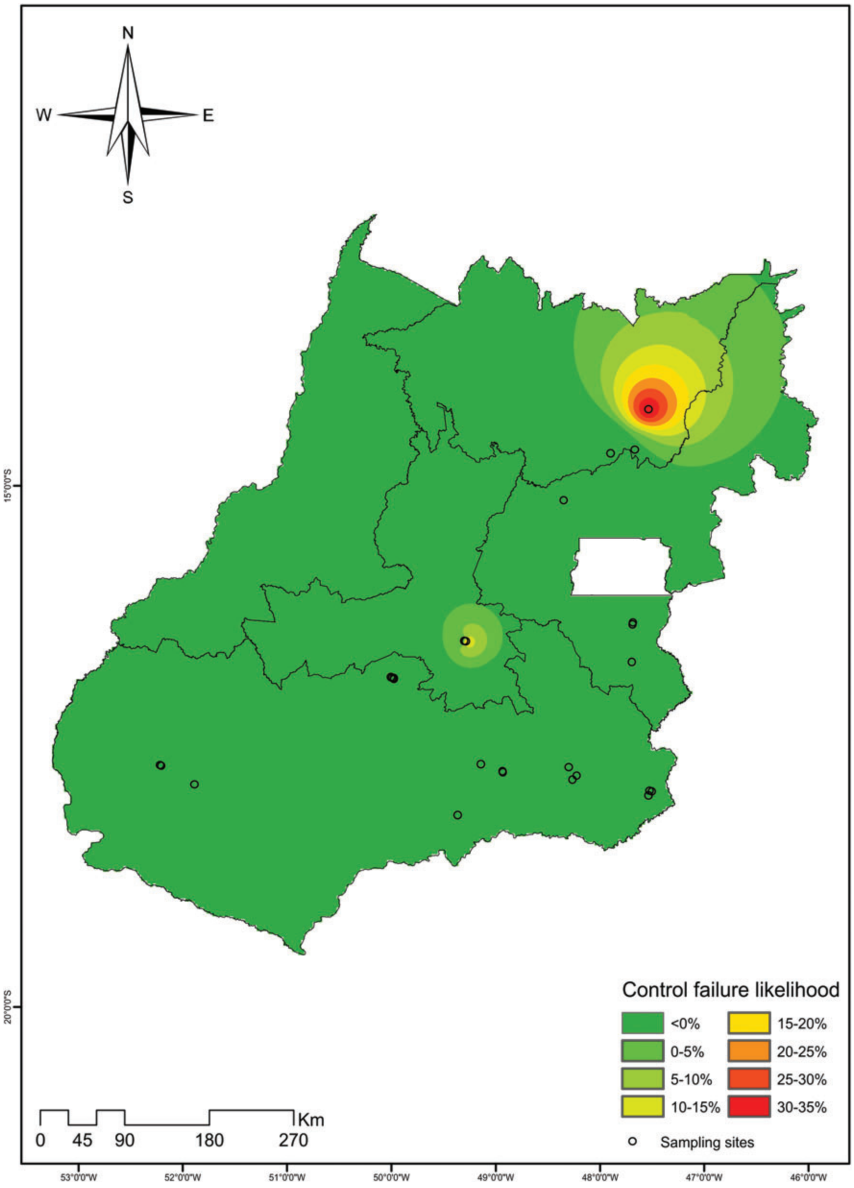

Figure 2. Contour maps and map of sampling sites of imidacloprid control failure in Brazilian populations of the Neotropical brown stink bug *Euschistus heros* from the state of Goiás in central Brazil (2015–2016). The color legend indicates the represented risk of control failure (i.e. control failure likelihood; %) of the neonicotinoid insecticide imidacloprid. The open circles indicate the sampling sites. West Goiás exhibits rare occurrence of the insect species and therefore was not targeted in the sampling effort.

requiring (i) the use of realistic bioassays that simulate insecticide field exposure, (ii) the use of commercial insecticide formulations and their label rates (as discriminating concentration) for the bioassays and (iii) a minimum efficacy threshold for the insecticide (e.g. 80 % mortality). The mortality assessment thus obtained can then be reverted into estimates of control failure likelihood, while time–mortality (survival) curves may be used for estimation of median lethal times (LT_{50}) allowing estimation of existing resistance levels if the species is amenable to such bioassays. Nonetheless, the monitoring of control failure likelihood, although potentially important, is a late effort when compared with insecticide resistance monitoring, limiting the management tools available for the purpose.

Regardless of the monitoring interests and limitations, we need to go beyond the current approaches and incorporate geographical components into the surveys of insecticide resistance and control failure likelihood. The increasingly popular geospatial methods and tools can be used to estimate the spatial dimension of current problems of insecticide resistance and consequent control failures for their proper management. The generation of geo-referenced maps of insecticide resistance and/or control failure likelihood will allow for better decision-making for resistance management, providing much needed support for farmers, research personnel and industry and country officials. Such effort can spatially fine-tune mitigation measures for insecticide resistance, compensating its cost. Otherwise, such measures would be applied in broader areas or contexts at greater expense when based on general baseline studies, market deployment and education strategies. All of these later strategies could be better focused with spatial surveys, whose granularity will profit from cueing on the pest species population structure and bio-ecological traits.

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