Preface

Research, development, and innovations in health risk assessment for Brazilian agriculture

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Introduction

Agriculture and livestock are key sectors of the Brazilian economy, which are essential for the country's economic growth and for the equality between the domestic currency's supply and demand. Agribusiness answered for about 23% of the gross domestic product (GDP) in 2015, according to Confederação Nacional da Agricultura (CNA), and reached 50.3% of total exports in February 2016, according to Secretaria de Relações Internacionais do Agronegócio (SRI) of Ministério da Agricultura, Pecuária e Abastecimento (Mapa) (Brasil, 2016). Currently, this sector is recognized as the most competitive and efficient in Brazil, considering the global scenario.

Despite the high-technology standard adopted by Brazilian producers, in the last ten years, agriculture has suffered considerable economic losses due to attacks of, at least, 35 new pests (Lopes-da-Silva et al., 2014). Furthermore, approximately 500 species of quarantine pests still show potential to cause significant damage to Brazilian crops. According to Sociedade Brasileira de Defesa Agropecuária (SBDA), 150 quarantine pests are already present in South American countries close to Brazilian borders (Lopes-da-Silva et al., 2014). This means that, at any moment, a new pest can reach the country without being noticed, due to the transmission by plants, animals, microorganisms, and their respective propagative organs, as well as through wind or humans, in a more-and-more globalized world. The Brazilian quarantine institution subjects agricultural products to sanitation barriers, which are imposed by commercial partners and may interfere in exports, creating a scenario that is difficult to reverse (Lopes-da-Silva et al., 2014).

It should be noted that propagation by human intervention is quite significant, especially when considering that 90% of global trade is carried out by sea and that air traffic is the most common entry point for pests (Olson, 2006). In the United States, for example, 725 thousand interceptions were reported from 1984 to 2000, of which 73% were through air cargo (Olson, 2006).

This thematic issue of the journal Pesquisa Agropecuária Brasileira (PAB), a special issue on health risk assessment for Brazilian agriculture, carries an important warning about exotic pests that cause economic losses both to Brazilian producers and to the global flow of commodities.

Quarantine pests in Brazil

The propagation of quarantine pests by human intervention is a recurring problem in Brazil. Since the decades of 1980 and 1990, emblematic pests have caused great losses to Brazilian agriculture. One of the most relevant has been the boll weevil (*Anthonomus grandis*), an exotic pest originated from Mexico, that crossed borders and arrived in Brazil in 1983, spreading rapidly through the cotton-producing areas and causing great losses to the crop, since the producers were not prepared to manage it (Praça, 2007). This pest changed the entire geopolitics of fiber production in the country, causing the crop to be transferred to the Cerrado (Brazilian savanna) region.

Another disease, known as witch's broom, caused by *Moniliophthora perniciosa*, was considered by many scientists as a result of an alleged plan to sabotage the competitive, Brazilian cocoa production in the global market. Statistics from 2000 showed that the crop's production reduced from 400 to 210

thousand tons. From 1997 onwards, Brazil had to start importing the product, with a total of 71 thousand tons imported only in 2000 (Alves, 2002).

The fungus *M. perniciosa*, the causal agent of the disease, existed endemically in the North region of the country and somehow spread to the south of the state of Bahia, seriously affecting cocoa production, leading producers to bankruptcy. This problem continues until today and is a challenge to the researches in the area and to the cocoa industry.

It is also important to highlight the impact on agriculture of the emergence, in 2000, of soybean rust, caused by the quarantine organism *Phakopsora* pachyrhizi and currently considered one of the most severe diseases of soybean. Reports show production losses reaching up to 90% in different regions around the world (Hartman et al., 2005). In Brazil, soybean rust caused significant economic, social, and environmental losses (Consórcio Antiferrugem, 2016). In the first years after its introduction in the country, significant losses in soybean production were observed, mainly due to the lack of knowledge of the producers on the types of treatment and management practices for the crop; in addition, few fungicides were registered for the control of the pest (Yorinori & Lazzarotto, 2004). The resulting direct and indirect losses, therefore, can be great. It is estimated that the costs to control the disease are about US\$ 2 billion dollars per year, especially since an average of three herbicide applications are adopted in almost 100% of the entire soybean-producing area in Brazil (Godoy, 2012).

Agriculture defense programs

Quarantine pests are classified as two types – A1 and A2 –, according to the Brazilian public administration. Type A1 refers to the pests that have not yet entered national territory, but that have a great potential to cause economic losses if they are introduced in the country. Type A2 comprises pests that are present in the country and that have already been scientifically reported, are endemic, and have an official control program due to their high risk of causing losses; this is the case of the citrus Hindu mite (*Schizotetranychus hindustanicus*) and of the red palm mite (*Raoiella indica*) (Lopes-da-Silva et al., 2014). Therefore, an effective plant defense program is imperative

to coordinate actions aiming to reduce the risk of the entry of pests or the negative impacts caused by these organisms, regardless of them being insects, nematodes, fungi, bacteria, viruses, invasive plants, among others. This type of prevention is currently one of the strongest governmental policies, since it may represent an economy of billions of dollars in control measures, in maintaining jobs in the field and in the agro-industry, and in reducing environmental impacts.

Mapa created the system of international agricultural surveillance, "Sistema de Vigilância Agropecuária Internacional" (Vigiagro), coordinated by its Department of Agriculture Defense, in order to regulate the inspection of animals, plants, and their products and subproducts in global transit. The importation of products of animal origin is inspected and controlled by this department to safeguard animal and public health, as well as the country's socioeconomic development (Lopes-da-Silva et al., 2014).

The challenge of health surveillance in Brazil is great, mainly when considering the country's extensive border of 23,102 km, of which 15,735 are land and 7,367 km are sea borders (Produção Agrícola Municipal, 2012).

Pests, such as soybean aphid (*Aphis glycines*), parasitic witchweed (*Striga* sp.), rice bacterium (*Xanthomonas oryzae* pv. *oryzae*), palm lethal yellowing phytoplasma, *Maize streak virus*, and *African cassava mosaic virus*, are only a few that roam the Brazilian borders (Embrapa, 2016b). To control these pests, the Brazilian plant defense system includes territorial intelligence services to monitor the country's borders, phytosanitary analysis of the plant material in quarantine, and preventive genetic improvement researches.

Contributions of Embrapa

Embrapa, using its georeferenced database, has been working to identify the probable entry routes of pests in Brazil and the spots where their dissemination is likelier to occur, including federal and state highways, and has also been supporting preventive actions against the entry and establishment of quarantine pests in the country. Until now, 364 possible entry routes by land, from roads and highways close to neighboring countries, have been identified, besides 26 spots

through sea (Spadotto et al., 2014). Also alarming are the 519 landing strips, most of them located in private properties, near Brazil's borders, requiring inspection and control of possible entry routes of pests (Spadotto et al., 2014).

Preventing the entry of potential pests demands an effective action of the Brazilian quarantine services. Quarantine actions have been developed by Embrapa for more than four decades. Aiming at greater biosafety in agriculture, these actions have already led to the identification and eradication of approximately 80 pests. However, the risk of new pests arriving in the country is more and more alarming. In the area of researches, for example, about 85% of the plant propagation material brought to Brazil for this purpose is contaminated with pests (Diniz, 2013). For this reason, Embrapa has a research center exclusively for plant quarantine, where analyses are carried out on seed and other propagation material introduced in the country or exchanged among research institutions (XXI Ciência para a Vida, 2016, p. 2331). Today, cutting-edge technology, such as the identification of molecular markers and DNA sequencing, is associated with traditional methodologies to guarantee that the detection of pests and the treatment of contaminated plants are performed with safety (Margues, 2006, 2007). The plant quarantine laboratory of Embrapa, for example, uses molecular markers to identify fruit flies when they are still larvae inside fruit (Lopesda-Silva et al., 2014). Another used method is DNA barcoding, in which quarantine pests intercepted by the quarantine services have their DNA extracted and sequenced (Barcode of life: identifying species with DNA barcoding, 2016). The standard sequence is then compared to other sequences of the same or of similar species, whose barcoding has already been published. Through phylogenetic analyses, the species is confirmed, reinforcing morphological identification. This type of study allows mapping the many sequences of a quarantine pest and allows a more accurate diagnosis (Lopes-da-Silva et al., 2014).

The case of Helicoverpa armigera

Despite the efforts in the areas of research and public administration to control quarantine pests, Brazil is still stricken by high-relevance phytosanitary threats that cause considerable losses to agribusiness. The species *Helicoverpa armigera* is an example of a recent quarantine pest introduced to Brazilian crops, which causes great losses in cotton, corn, and soybean crops.

Until 2012, this pest had not yet been found in Brazil. However, since then it has been identified in the country. The first register was in the states of Bahia, Mato Grosso, and Paraná, as well as in the Federal District, and the taxonomic and molecular identification of the pest was done by researches from Embrapa Cerrados and Embrapa Soja (Specht et al., 2013). At the same time, the pest was also detected in soybean and cotton crops in the states of Bahia, Goiás, and Mato Grosso by researches from Universidade Federal de Goiás and Fundação Mato Grosso (Czepak et al., 2013). This led to a general concern among producers, encouraged by several sectors that had little knowledge on the species. Due to the voracity of the attack to many commercial crops and to the existence of 180 alternative hosts, it was believed that the insect H. armigera could disseminate Brazilian agriculture.

To prevent greater losses in Brazil, Embrapa organized a "caravan" to warn against phytosanitary threats, called "Caravana Embrapa de alerta às ameaças fitossanitárias". This caravan reached all the producing regions of Brazil, totaling 35 production poles, in 18 states. To exemplify the interaction between Technology Transfer (TT) and Research & Development (R&D), the participants included 34 researchers from the areas of entomology and technology for application to agricultural products, who worked together with TT teams from several research centers of Embrapa. The caravan brought control measures against the larva to the grainproducing areas of Brazil, in terms of integrated pest management (IPM). IPM was structured in 1980, initially by Embrapa soja, located at Londrina, in the state of Paraná, then by Embrapa Milho e Sorgo, located at Sete Lagoas, in the state of Minas Gerais, and by Embrapa Trigo, located at Passo Fundo, in the state of Rio Grande do Sul; however, in other research units, new strategies - such as transgenic crops, refuge areas, restrictions on planting, and technology application - were included in the concept of pest management of regional production systems and of agriculture landscape management.

Phase I of Embrapa's caravan, undertaken in 2013 and 2014, reached all producing regions of Brazil,

thanks to the protagonism of the research units of Embrapa. Phase II was carried out in 2015/2016, continuing previous actions, using the same method to train multipliers. In this phase, the focus was to train producers on decision making for adoption of IPM to control pests, considered controversial.

Preventive genetic breeding

The Brazilian program for preventive genetic breeding, called "Programa Nacional de Melhoramento Preventivo" (Agropreventivo), is an initiative led by Embrapa with the goal to make available to the producers plants resistant to pests (including insects, diseases, and weeds) of quarantine importance (Embrapa, 2016b). This program was signed in February 2014 by the Department of Agriculture Defense. The signed protocol of intentions seeks scientific cooperation and to formalize partnerships among participating institutions. The objective of Agropreventivo is to develop, beforehand, cultivars resistant to quarantine organisms of high risk to Brazilian agriculture, before they reach the country's territory, which could jeopardize the competitiveness of national agribusiness, leading to incalculable consequences in the global market, particularly regarding phytosanitary barriers.

Preventive breeding is a strategic activity, which involves several disciplines, and should be supported by partnerships among different national and international institutions. This is fundamental since tests abroad are expected in countries where the pest already occurs naturally. Resistance genes are monitored through DNA analysis, which allows continuing the assessment in countries free of the disease or pest (XXI Ciência para a Vida, 2016, p. 14-22). In Panama, for instance, rice lineages are being evaluated using genes resistant to the quarantine bacterium *Xanthomonas oryzae* pv. *oryzae*, one of the greatest threats to the rice crop worldwide.

Currently, genomic technology has been used in preventive breeding in quarantine organisms that, besides rice, also attack bean, soybean, coconut, and grapevine (Embrapa, 2016b).

This technological route is supported by similar initiatives that were well succeeded in the past. One example is the casual agent of coffee leaf rust (*Hemileia vastatrix* Berk. & Broome), whose preventive research

helped save the Brazilian coffee crop in the decade of 1970. The strategy of the researchers was to cross the coffee species *Coffea canephora* and *Coffea arabica*, in order to transfer the resistance of the first to the second. The hybrids developed were then tested at the research center on coffee rust, Centro de Investigação das Ferrugens do Cafeeiro, located at Oeiras, in Portugal. In 1970, when coffee leaf rust arrived in Brazil, the producers already had access to a resistant coffee hybrid, which was already being multiplied. At that time, coffee was the main product of Brazil's exporter scope, and, therefore, this anticipation safeguarded the country against a great loss (Mccook, 2008).

Databases on quarantine pests

Public policies for control of new pests are also fundamental, in order to guarantee the competitiveness and the quality of the products of Brazilian agribusiness in the global market. These public initiatives may be supported by information systems, such as the database on pests (BD) (Embrapa, 2016a) and WikiPragas (Embrapa, 2016c), both developed by Embrapa. With them, it is possible to manage worldwide data on pests associated with the main crops of interest of Brazilian agribusiness and to facilitate the risk assessment of the attack to plant products. BD systemizes cataloguing data, such as, the scientific name of the pest, its hosts, the affected plant parts, and the countries where it occurs. WikiPragas is a module that integrates the system and allows detailed files on the pests with quarantine potential, including aspects related to biology, inspection and detection, impacts, control measures, and mitigation.

Organizing research to combat phytosanitary threats

The efforts in organizing research to protect the country against the entry and dissemination of new pests are also done in an orderly and persistent fashion.

Embrapa developed a research structure with the objective to organize and prioritize technical and scientific efforts, in order to improve quarantine procedures and to support public policies developed by Mapa. This structure comprises a group of 40 research projects aiming to prevent the entry of quarantine pests in Brazil and to manage them. These projects involve

25 research centers of Embrapa, besides foreign partners and consultants from the private sector (XXI Ciência para a Vida, 2016, p. 31).

Moreover, Embrapa dedicates itself to study plant and animal protection using two portfolios on research projects, one on plant and another on animal protection. The portfolio on plant protection comprises four areas: guarantine intelligence, using techniques to avoid that a pest spread in a certain region; advanced breeding techniques applied to plant protection; multitrophic vision of the agroecosystems, which considers the complex interaction between plants and the other species of live organisms, as well as all diversity observed in each case; and management of agriculture landscapes (XXI Ciência para a Vida, 2016, p. 31). The portfolio on animal protection seeks to generate knowledge and technologies that allow amplifying the protection, production, and competitiveness of the cattle, swine, poultry, eggs and derivatives, caprine, ovine, aquaculture (fish, shrimp, mollusk, and bivalves), equine, and bubaline production chains. Transversal studies support these researches, focusing on veterinary epidemiology, animal immunology and vaccinology, biotechnology applied to animal health, comparative pathology, economic studies on animal diseases, genetic resistance of the host to animal pathogens, veterinary public health, and animal wellbeing.

The research structure and the portfolios on projects on animal and plant protection are in alignment with the public policies and support scientifically the program of Mapa through Vigiagro.

Highlighting the contents of the papers published in this issue

In this thematic issue, 72% of the articles are contributions of authors from different institutions, showing an important research network on the studied theme. Of the 36 published papers, 6 included the participation of institutions from Argentina, the United States, France, England, and Mexico. Regarding national institutions, the following ones took part in this issue: 31 research units of Embrapa, 20 federal universities, 8 state research institutions, 6 state universities, 6 private universities, and five other institutions.

The results of the researches of these institutions are presented in 18 scientific articles, 7 scientific notes, 5 reviews, and 6 special collaborations. Among these collaborations, is the review (p. 422) on the decline and collapse of bee (*Apis mellifera*) colonies, which compiles the most current studies on the possible causes of a problem that worries global agriculture. Aspects, such as nutrition, management of bee hives, parasites, and effects of agro-toxics, are presented and discussed. Other papers explore the biological fundamentals and innovative aspects related to plant quarantine improvement in Brazil, report advances on etiological and epidemiological knowledge on pests, and show the applicability of biological and genetic control measures.

Soybean rust, the most severe soybean crop disease, is discussed in the review "Asian soybean rust in Brazil: past, present, and future" (p. 407). Losses that reach 90% of the Brazilian soybean production make this disease a challenge to researchers that seek its efficient control, which includes obtaining tolerant varieties to resistant transgenic plants, and also emphasize the need for the development of management systems that are more adequate to soybean, depending on the region where the crop is established.

The review on introduced forests pests in Brazil (p. 397) shows that monitoring actions, sentinel plants, and genetic improvement are priorities for the sustainability of the 7.6 million hectares planted in the country. Still in relation to forest areas, the paper on the susceptibility of cedar (*Cedrela fissilis*) to the attack of pests in seasonally deciduous forest (p. 607) showed that the caterpillar *Hypsipyla grandella* and the larvae of the sawyer beetle *Oncideres* sp. deform cedar, which makes the commercial exploitation of this wood unfeasible.

Regarding animal production, the main health threats that affect the production chain of swine in Brazil are pointed out in another review paper (p. 443). Among the papers on this theme, the following subjects were also discussed: results of researches on fish, mainly associated with the prevention of photobacteriosis in cobaia culture (*Rachycentron canadum*) (p. 465) and with the host-parasite relation during infestation by *Epistylis* sp. in farmed cichlid and pimelodid fish (p. 520). The map on the likelihood of the introduction of the foot-and-mouth disease in Brazil (p. 661), as well as the recommendations for

the development of public policies to reduce the risk of entry of this disease through Brazilian borders, shows the amplitude of the devastating impact on the global market that may be caused by the occurrence of this disease in the country. Regarding sheep and goats, the risks factors in the transmission of the bacterium *Chlamydophila abortus* (p. 654) were emphasized in a study carried out in over 100 farms. This bacterium is linked to reproductive disturbances, especially enzootic abortion of sheep and goats.

Phytosanitary threats become even more critical when agriculture is competitive and has intense global exchange, which makes protection through efficient control mechanisms necessary. In this sense, several papers (pgs. 473, 502, 494, 483, and 623) on the process of plant quarantine showed that: i) fungi, viruses, and mites are the most intercepted in the country and that the infestation rate of the analyzed material is approximately 2%; ii) there are legal issues related to the importation of biological control agents by Brazil and that quarantine facilities are equipped with adequate and safe procedures to asses these introductions, aiming at the complete safety in the importation of exotic organisms; iii) Brazilian agriculture needs control measures for the interchange of materials and that facilities for plant quarantine should regulate the entry of new materials, in order to protect the entire production system and, at the same time, mostly protect commercial relations, avoiding nontariff barriers; iv) through the eradication of exotic fungi in imported materials, it was possible to identify, from 1977 to 2013, 37 exotic fungi, of which 4 are classified as absent quarantine; and v) due to its population dynamics, the bacterium Pseudomonas savastanoi pv. phaseolicola should be categorized as A2, i.e., a present quarantine pest.

The presence of the caterpillar *H. armigera* was detected in Brazil at the beginning of 2013 and was considered a great threat to the Brazilian production system; this event showed the need of effectively reinstating and spreading IPM as a routine practice by the country's producers. Among the adopted practices, stand out new transgenic crops, restrictions on planting, refuge areas, pest management in the regional production system, and the concept of landscape management, not only of one crop. Of the IPM tactics, the use of insecticides is an important tool, and the evaluation of products available in the

market has become an emergence procedure for pest control. In this sense, seven chemical and biological insecticides (baculoviruses and *Bacillus thuringiensis*) were tested, and the results are a key contribution to the fight against this threat (p. 527). The biological aspects and the survival of *H. armigera* and *Helicoverpa zea*, when exposed to Bt proteins, were also highlighted (p. 537).

Since mycotoxins may cause severe damages to the flours of several products, it is important to detect their presence in wheat and soybean flours, in which deoxynivalenol and zearalenone were identified by the Elisa test (p. 647).

Future climate change scenarios were considered to predict the occurrence of the red palm mite (p. 586). The used models revealed that the impact of the pest will probably worsen with global warming. The papers on the spotted wing drosophila (Drosophila suzukii) showed that there was considerable damage, especially in grape (p. 599), and that apple, peach, persimmon, fig, and pear are possible hosts. The obtained results also indicated that the South region of Brazil is climatically favorable for its spread, where there can be great losses (p. 571). However, temperatures above 30°C make the dispersion of D. suzukii difficult (p. 571). A total of 18 grapevine genotypes were evaluated, and the most resistant and susceptible to the attack of D. suzukii were informed (p. 599).

The sustainable intensification of production systems, such as crops, livestock, and forestry, becomes feasible as important studies on the identification of grass species, used as pasture, compose these systems. Brachiaria (*Urochloa ruziziensis*) clones resistant to the fall armyworm (*Spodoptera frugiperda*) were presented (p. 579) as a way to make these systems viable in several climatic conditions. This study showed differences among the brachiaria clones and that the fall armyworm did not develop in 20 of them.

The "Q biotype" whitefly is one of the phytosanitary threats identified as highly resistant to insecticides and with high capacity of transmitting viral diseases. The Q biotype is found in South America, in neighboring countries, and has high a probability of entering Brazil. The "B biotype" whitefly has already been reported in the country, as well as the native biotype. The development of molecular markers for the

identification of the Q biotype was also discussed in another paper (p. 555).

Furthermore, this thematic issue of PAB presents other works on the management of crops, regarding the application of fungicides to reduce kernel rot in corn (p. 638) and the effect of wheat sowing date on the prevention of wheat blast disease (*Magnaporthe oryzae*), currently one of the most important diseases of this crop (p. 631). Several key themes were discussed in the scientific notes, including: i) monitoring of *H. armigera* in the Cerrado (p. 697); ii) presence of *Wheat mosaic virus* (WMoV) in wheat samples from the United States (p. 688); iii) biological control of the red palm mite by populations of the mite *Amblyseius largoensis* in the field (p. 671); and iv) mitochondrial markers of *S. frugiperda* populations associated with corn and cotton crops (p. 692).

This way, PAB innovates by releasing this thematic issue, which will contribute to the recognition of the importance of health risk assessments for Brazil and to the need to promote scientific advances in order to protect the country against these threats, which put into risk the stability of agriculture production, as well as the competitiveness and sustainability of Brazilian agribusiness.

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