Number 4 Issue 2 June 2014

International Biotechnology Color Journal

A Scientific Peer Reviewed Journal with Focus on BIOTECHNOLOGY
and Covering Its Many Hues, Tints, Tones & Shades

Cobs of some maize varieties from Oaxaca, Mexico
(taken form http://www.mexicocampoadentro.org/maices_oaxaca.php)

Special issue:
Abstracts to selected oral presentations from the From the Biotechnology Summit 2014

From october 8 to october 10, 2014 at Huatulco, Oaxaca, Mexico

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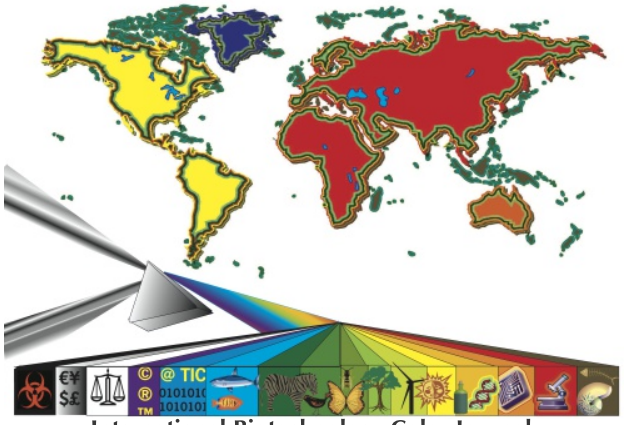
The IFBR&ESCHNSAST&S is a civil association and nonprofit organization.

Its main goal is to help bringing together efficient biotechnological solutions for many human problems, with the need for environmentally friendly and sustainable processes.

As means to such goal, the Foundation counts on education to create social awareness to biotechnology's benefits and risks, and to promote the formation of highly qualified professionals and research scientists.

The constitutive act of the foundation was signed in the Heroic City of *Huajuapan de León, Oaxaca*, on September 14th, 2009, at the *NOTARÍA PÚBLICA No. 61 de los ESTADOS UNIDOS MEXICANOS*. As .



International Biotechnology Color Journal

Susana Lozano Muñiz
President of the Foundation

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Director: Rogelio Rodríguez Sotres, Faculty of Chemistry, Universidad Nacional Autónoma de México. Cd. Universitaria. Ave. Universidad 3000, C.P. 04510, Coyoacán, D.F. México. E-mail: sotres@servidor.unam.mx
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International Biotechnology Color Journal

A Scientific Peer Reviewed Journal with Focus on BIOTECHNOLOGY and Covering Its Many Hues, Tints, Tones & Shades

Editorial section

International Biotechnology Color Journal (IBCJ) is an electronic Open Access journal, devoted to rapidly publishing full peer-reviewed articles covering all the fields of biotechnology. The central focus of IBCJ is to publish scientific reports, though it also provides a forum for reviews, short notes on relevant findings, essays on novel technical advances or relevant updates, book reviews, scientific meeting reports, and letters to the editor. Instructions for every type of contribution are presented in the journal's Homepage and in PDF format in the last issue of each year.

The Editorial Board of IBCJ is fully committed to publish articles innovating in all areas of biotechnology. Contributions are reviewed from a rigorous optic of scientific criticism; thus, any original contribution that fits within the scope of the journal and promotes the advancement of biotechnology are particularly welcome.

Editorial comments to the contents of this issue

By José Juan Zúñiga-Aguilar, Chief editor.

In this special issue of the International Biotechnology Color Journal, we are particularly glad to publish the abstracts to the oral sessions from the Biotechnology summit 2015, held in Huatulco, Oax. Mexico, from the 8th to the 10th of october, 2014.

Huatulco is a peaceful Beach Resort with relaxing environment. An excelent place to host the conference Biotechnology Summit 2014, organized by an international committee with the support of the "International Foundation for Biotechnology Research & Early Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society". The meeting offered an international forum for scientists devoted to a wide range of subjects around Biotechnology.

To enhance the visibility of the works presented at the meeting, IBCJ now publiches selected abstracts to those oral presentations were the authors gave their consent.

The impact of new scientific advances in a number of fields of Biological Sciences, Chemistry and Engineering of materials, Electronics and solid state Physics, amongst others, has provided Biotechnology with novel powerful tools. Scientific meetings of high standards, such as this one, are relevant to bring toghether scientest of different areas to discuss their work toghether and generate new ideas for the future.

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ABSTRACTS TO ORAL PRESENTATIONS OF THE BIOTECHNOLOGY SUMMIT 2014

The Meeting was organized by an International comitte and hosted by the "International Foundation for Biotechnology Research & Early Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society*", the "Universidad del Papaloapan" and the "Universidad del Mar, (UMAR) campus Huatulco".*

The Biotechnology Summit 2014 was held in Huatulco, Oaxaca, Mexico, on October 8 to 10, 2014.

Organizing Scientific comittee:

Susana Lozano Carlos Blanco Víctor Toledo López

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Cristina Rodriguez-Padilla José Juan Zúñiga Aguilar

(1) The abstracts presented here were not peer reviewed

Tuning optimal-robust linear MIMO controllers of bioreactors by using Pareto optimality.

Jesús Carrillo Ahumada

Profesor-Investigador, Instituto de Biotecnología, Universidad del Papaloapan, Tuxtepec, Oaxaca, Circuito Central #200, colonia Parque Industrial, Tuxtepec, Oax., México C.P. 68301, Phone 01 287 87 59240 ext. 220, e-mail: jcarrillo@unpa.edu.mx

Pareto optimality was introduced in order to find the better equilibrium between performance and robustness of linear controllers by the simultaneous minimization of the quadratic-error and quadratic-control functions integrals. The Pareto optimization problem was solved setting the characteristic matrix eigenvalues in the region of left complex semi plane where |Im/Re|<1 as constraint. *2D* Pareto fronts were built with the quadratic-error function integral *vs.* quadratic-control function integral. The proposed method was applied for tuning linear controllers of two bioreactors one of them unstable SISO and the other one stable MIMO.

Theranostic nanoplatforms in cancer: an overview

Aracely Angulo Molina[1] and Lizette Alejandra Minjares Espinoza[2]

[1]Universidad de Sonora, [2]Universidad de las Americas Puebla, e-mail: aracelyam@hotmail.com.

Nanobiotechnology in medicine is an area of emerging interest and opens up a whole universe of new possibilities for the early stage diagnosis and treatment in cancer. Developing countries are also taking part in the development of this technological revolution in medicine. In Latin America, the leading nations in this area are Brazil, Mexico and Argentina. The National Nanotechnology Initiative (Arlington, VA, USA) defines nanotechnology as 'the understanding and control of matter at dimensions of roughly 1-100 nm, where unique phenomena enable novel applications'. In fact, nanomaterials are designed to have at least one dimension (length, width, height) at the nanoscale of 1-100 nm. Nanoscience is defined as the study of phenomena and the manipulation of materials at the atomic, molecular and macromolecular scales, where the properties differ from those at a larger scale. Nanomaterials that have a nanoscale length, width and height are known as nanoparticles. As the size of the particles gets reduced to nanoscale range, there is an immense increase in the surface to volume ratio which increases reactivity and change the mechanical, electrical, and optical properties of the particles. The nanoparticles, having much larger specific surface area than their coarse analogs, exhibit enhanced biological activity and present undeniable interest as carriers or in drug delivery. Many types of nanoparticles exist with respect to their size, shape, material, and coatings. The specific properties of the core materials provide distinct monitoring and therapeutic applications. For example, nanoliposomes and nanocasules have been evaluated over the years, and a significant amount of evidence has been obtained showing that these carrier materials are able to improve the balance between the efficacy and the toxicity of therapeutic interventions. Besides for therapeutic purposes, nanomedicine formulations have in recent years also been increasingly employed for imaging applications. Theranostics is a concept which refers to the integration of imaging and therapy.

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Red: Medical Biotechnology

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Impact of transgenic maize in Oaxaca, the maize origin land.

Ma. del Carmen Urzúa-Hernández[1], Teresa Ivonne Castillo Diego[2] and Susana Lozano Muñiz[3,4]

[1]Universidad Nacional Autónoma de México, Facultad de Química

[2]Universidad Tecnológica de la Mixteca, 3Universidad del Papaloapan,

[4]International Foundation for Biotechnology Research & Early Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society A.C.

The world needs producing Maize sustainably. It can grow at all altitudes. In rural places it is produced mainly with traditional technologies, since the use of improved varieties is scarce. In Mexico the corn is the main food as: esquites & corn on the cob, masa for tortillas & antojitos (typical food) as: garnachas, molotes, tamales, atole, tejate, tlayudas, dobladas, quesadillas, memelas, tostadas, tortas de tamal, champurrado, etc. Oaxaca is know as "cradle of corn", proof of that is the breadth and variety of native corn color as white, yellow, red, and blue. Flavio Aragón Cuevas, et al in 2006, made a study where they find different color of maize in Oaxaca Mexico as yellow, blue, white, orange, red & black. They collected maize in Oaxaca and they found 35 landraces: Ancho, Arrocillo, Bolita, Celaya, Chalqueño, Chiquito, Comiteco, Conejo, Cónico, Cónico Norteño, Elotes Cónicos, Elotes, Occidentales Mixeño, Mixteco, Mushito, Nal-Tel, Nal-Tel de altura, Negro de Tierra Fría, Negro Mixteco, Olotillo, Olotón, Olotón imbricado, Palomero Toluqueño, Pepitilla, Serrano, Serrano de Oaxaca, Serrano Mixe, Tabloncillo, Tehua, Tepecintle, Tuxpeño, Vandeño, Zamorano, Zapalote Chico, Zapalote Grande. It is this diversity of maize, which could risk to the possibility of interbreeding with genetically modified maize extensively studied in the world. Independently it is a tool to free hunger from other communities in the world, or to be used as an alternative source for biofuels. The impact of introducing transgenic maize in Oaxaca is a risk to the diversity of the center of origin of maize. Although it is a benefit to other communities that require more maize production for various needs such as feeding or biofuel production.

Figure 1. Cobs of some maize varieties from Oaxaca, Mexico (taken form http://www.mexicocampoadentro.org/maices_oaxaca.php).

Green: Wildlife, Ecology & Environment Biotechnology

Chemistry Student's interest on Biotechnology.

María del Carmen Urzúa-Hernández[1] and Susana Lozano-Muñiz[2,3]

[1]Universidad Nacional Autonóma de México. Edificio A, Facultad de Química, Circuito Escolar s/n, Ciudad Universitaria, 04510, México, DF. mcurzua@hotmail.com

[2]Universidad del Papaloapan, campus Tuxtepec Oaxaca

[3]International Foundation for Biotechnology Research & Early Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society A.C.

The training of professionals dedicated to biotechnology in Mexico has failed to have the expected impact on related areas. To investigate what happens to students, we validated a questionnaire and a semi-structured interview and then applied it to students from two careers in the chemistry area. The results allowed us to detect a serious lack of information about what is involved in biotechnology careers. Clearly their background information or the students' taste, developed in childhood, has a positive influence in the interest for certain aspects of the career. Thus, a wider dissemination of truthful information about what biotechnology is and its potential uses for the progress of humanity is considered desirable. It is also important that information about biotechnology is viewed from a historical point of view, including the period where there was no direct genetic manipulation, so students know exactly what the differences are in the impact of biotechnology, as a whole, and genetic engineering, in particular.

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Indigo: Biotechnology education

Attitudes toward animal research among Mexican undergraduate science students

Diana Caballero Hernández[1], Susana Lozano-Muñiz [2,3], Cristina Rodrlguez-Padllla[1]

[1]Laboratorio de lnmunología y Virología,Facultad de Ciencias Biológicas, Univensidad Autónoma de Nuevo León,N.L. México. E-mail: decaballero@outlook.es

[2]Universidad del Papaloapan, campus Tuxtepec, Oaxaca, México.

[3]International Foundation for Biotechnology Research & Earty Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society A.C.

Teaching of bioethics has become an important part of scientific careers' curricula offered by universities. One of the many topics covered during such courses is the use of animals in research, an issue that for the general public is very controversial, given the growing concerns for animal welfare. This ultimately leas to a poor support for the use of animals in research, testing and entertainment. Attitudes regarding this topic exhibited by students from scientific careers are of particular interest, since these will likely be challenged by real life situations during their training or as professionals. We have applied the Gallup & Beckstead questionnaire to a group of undergraduate Mexican students to explore their attitudes toward animal experimentation. The survey sample consisted of 127 students currently enrolled in two Mexican universities: *Universidad Autóma de Nuevo León* and *Universidad del Papaloapan*, offering majors in diverse disciplines including, biology, chemistry, food science, nursing and biotechnology. Student participation was anonymous and voluntary. To participate, they were provided a link to the online questionnaire to be completed on a web browser. The questionnaire also asked the next basic personal background from the participants: group age, gender, academic year, and career. According to the results, the students showed a strong concern for animal well-being and a strong agreement with the need for more regulation of animal research, but at the same time, the surveyed students recognized the importance of animal research for the advancement of knowledge. This is a small-scale study, results are preliminary,and a full-scale study is needed before solid conclusions can be reached.

Current trends in functional foods: dietary fiber

Laura Inés Elvira Torales, Eugenio Rangel León

Instituto Tecnológico Superior de Tierra Blanca, Av. Veracruz s/n esq. Héroes de Puebla, Col. Pemex, Tierra Blanca, Ver. C.P. 95180. e-mail: liet82@hotmail.com

Nowadays the importance of dietary fiber (DF) in health is well recognized. Since the 90's the WHO and many mexican government bodies have produced guidelines aimed to increase the daily intake of foods rich in dietary fiber; but, the formulation of new high dietary fiber products is still a challenge for the food industry. Dietary fiber fruit concentrates have a better nutritional quality than those from cereals due to the amount of associated antioxidant compounds and their balanced properties, such as: higher fiber content, soluble/insoluble DF ratio, water and fat holding capacities, lower energy value, and phytic acid content. On other hand, the food industry has large amounts of waste and byproducts that are a good source of dietary fiber and antioxidants and could be used as food. In recent times, dietary fiber has been used as a functional ingredient in many commercial food products, either to increase the intake of dietary fiber and antioxidants, or as a fat replacer in products with a high content of fat. This paper presents current research on dietary fiber, recent applications and its functional properties in different food products.

Starter cultures in the meat industry

Victor M. Toledo Lopez

Instituto Tecnológico de Mérida., Av. Tecnológico Km 4.5, Plan de Ayala, 97118 Mérida, Yuc. e-mail: vtoledo08@yahoo.com.mx

For many years ago, the agro-food industry has been using microorganisms and their enzymes to improve and modify the properties of food products. Different types of microorganisms (bacteria, yeasts and fungi) called "starter cultures" are used in the meat industry, to speed up some processes of maturation, metabolizing proteins and lipids, as well as synthesize compounds that give this product aroma and flavor. Their use presents four main functions: (i) decrease the value of the pH, at a given rate, and within certain limits, (ii) act against native germs and germs that colonize food and are considered microbial contamination, (iii) produce aroma as a consequence of the microbial metabolism in foods (in certain species) and (iv) generate certain qualities through the production of enzymes to induce hydrolytic and/or catalytic processes in the food. The mostly used genera are *Lactobacillus*, *Leuconostoc*, *Pediococcus, Streptococcus* (bacteria), and *Penicillium* (fungi), mainly *P. candidum*, *P. camemberti* and *P. roqueforti*. These species are added as initiators to ensure a flavor and aroma uniformty and good conservation. Used species of mould must not have toxic properties, so its use does not convey any risk to the final consumer. The main advantages of the starter cultures, when added to matured sausages can be summarized in the following points: control of the maturation process, inhibition of undesirable microorganisms, reduction of health risks, increase of quality and standardization, control of specific taste and aroma.

Artificial intelligence applied to biotechnology

Larissa Jeanette Peniche Ruiz

Instituto Tecnológico de Mérida. Av. Tecnológico Km 4.5, Plan de Ayala, 97118 Mérida, Yuc. e-mail: larissa.peniche@itmerida.mx, larissajeanette@itmerida.mx

Sciences such as artificial intelligence and biotechnology do not seem to have much in common, but they indeed have crossed their paths and put their efforts together to come a long way in helping each other by means of artificial neural networks, genetic algorithms and multi agent systems applied to food technology, agricultural and livestock production and renewable energy generation. Artificial intelligence deals with the study of computations to aid perception, reason and act (Winston, 1992) and it seeks to understand and mimic the functioning of the brain, in order to make machines more useful to humankind. On the other hand, biotechnology is defined as a set of molecular technologies such as manipulation and gene transfer, DNA typing and cloning of plants and animals. It is not unreasonable to say that both disciplines have very different objectives and functions: machines in opposition to living organisms, algorithms in opposition to production processes, field experiments in opposition to simulators, reality (the real world) in opposition to virtual reality. In opposite corners and yet... is there some point of convergence? Apparently, there is. In principle, they both seek human well-being and they are both based on the processes of life, which they try to imitate and improve. Most importantly, they both require the power of technology: computers, sensors, circuits, etc. There is no techno-biologist without a computer! It is easy to see that these disciplines can, in fact, merge and even complement each other in such a way that by joining their efforts they can come up with projects like an artificial nose to perform sensory analysis in the tasting of food; the successful imitation of skin with bioengineering and artificial cloning to implement a prototype of an intelligent limb prosthesis; a farming multi agent system capable of simulating livestock variables in order to maintain control over the use of the soils to prevent its deterioration and to ensure the economic sustainability of people; and finally the use of artificial neural networks and genetic algorithms to optimize the use of renewable energy in residential buildings in macro cities such as Mexico city. Artificial intelligence extends very long arms (and not exactly of the robotic kind) to all the sciences and today I am very pleased to chat with you all, specialists and scholars of biotechnology, to thank you all for the great opportunity offered to us engineers in computer systems, both of this burgeoning field of artificial intelligence and many other computer specialties such as the BD and computer networks to name a few. I would also like to invite you to talk to your closer peers and ask us, dare us to support you on the wonders that biotechnology works. Let us give the opportunity to future professionals in these areas to work together and design promising projects that carry aloft the well-being of us all.

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Entrepreneurship: A choice for life?

Ricardo Gómez Flores.

Centro de Incubación de Empresas y Transferencia de Tecnología (CIETT), UANL, Monterrey, Nuevo León, México E-mail: rgomez60@hotmail.com

The word "entrepreneur" comes from the French word *entreprendre*, 19th century, meaning "one who undertakes". According to the Oxford dictionary, an entrepreneur is someone "who establishes a business, taking on financial risks in the hope of profit." Entrepreneurship is a habit and an attitude; a set of skills applied to the search for innovation in the business, personal, and professional contexts, and an approach to life around innovative thinking, calculated boldness, and proactive behavior. Students who have an entrepreneurial mindset have developed strong communication skills, recognize how to take the initiative, and know how to execute it. Certainly, education is critical to every entrepreneur, it means, such people are made, not born, but being born into a family of entrepreneurs can be very motivating and influential, although this does not guarantee a future as an entrepreneur. The financial support to entrepreneurs in Mexico today is very exciting and should be taken; incubators of higher education institutions or governments, facilitate the process of entrepreneurship. It is important to understand that SMEs generate most of the country's GDP, therefore Mexico's economic future depends on them. Although entrepreneurship is essentially the creation of a company, by finding and taking advantage of an opportunity, and by gathering resources to make that opportunity to become a business, it is actually also a way of life. Creativity, critical thinking, resourcing, searching and seizing opportunities, time management, persuasion, negotiation, oral and written communication, leadership, and decision making are essential skills that every entrepreneur should develop. Leadership reflects the personality of the entrepreneur who must be someone who knows the market to take advantage of opportunities that arise to add value to his business, which fulfills his ambitions. The entrepreneur must be tolerant to risk, stress and failure, because the environment in which he operates is characterized by financial or personal potential risk; in addition, he must be tenacious, determined, disciplined and persistent, and have a great willingness to work to make a commitment. He must also have confidence in himself and have a strong passion to achieve goals, as well as having the ability to create and innovate to provide a fast and effective solution to the problems of invention and business development that are encountered.

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Bio-businesses and their economic value added

Teresa Ivonne Castillo Diego[1], Jose Luis Hernandez Motera[2], Susana Lozano Muñiz[3,4]

[1]Academic: Natural resources, social needs and strategies of business. Universidad Tecnológica de la Mixteca, Huajuapan de León, Oaxaca. México. Phone number: +01 953 53 203 99. E-mail: ivonne@mixteco.utm.mx

[2]Instituto Tecnológico de Orizaba

[3]Universidad del Papaloapan, campus Tuxtepec, Oaxaca, México.

[4]International Foundation for Biotechnology Research & Earty Stimulation in the Culture of Health, Nutrition, Sport, Art, Science, Technology & Society A.C.

Bio-businesses are organizations that are more focused on research and development, which are the primary and secondary activities in a value chain; herein lies the importance of analyzing how technology and science, properly work within these companies. Some businesses may be mistaken as bio-enterprises, when we are really talking about traditional businesses, which sell their products to the industries that use biotechnology. The difference between a bio-company and a traditional company, is that the bio-company is mainly dedicated to the research and its essential goal is to provide innovative solutions to the market. For a bio-company, the trends of the market will value your product, however, there is a downside to this type of organizations, the risk is in the market; normally these bio-enterprise are consolidated industries, profitable, competitive, with economic added value, but with a future growth linked to innovation. This research carries out a comparative financial analysis between a company and a traditional bio-business devoted to the paper industry, to assess the financial impact through the generation of economic added value in both businesses. The bio-business object of this research generates an EBIT (Earnings before interest and taxes) of twenty million pesos, Its net operating profit, after tax, amounts to about nineteen million and its economic added value is positive; in contrast, the traditional paper industry generates a positive EBIT, but its economic added value is negative. This bio-business pretends to pertain to the green market, composed of those markets emerging in the medium and long-term due to the trends in demand for organic products, and to education and environmental culture expansion, which will have an impact on its growth and development.

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Towards resource mobilization from global wheat gene bank to the farmer's field

Sukhwinder-Singh[1], Parshant Vikram[1*], Carolina Saint Pierre[1], Burgueño ferreira[1], Juan Andres[1], Huihui Li[1], Sergio Cortez Gamboa[2], Gaspar Estrada Campuzano[3], Pedro Figueroa López[4], Guillermo Fuentes Dávila[4], Víctor Manuel Hernández Muela[4], Javier Ireta Moreno[4], Martínez Rueda[3], Leodegario Osorio Alcalá[4], Ruairidh Sawers[5], Ernesto Solís Moya[4], Héctor Eduardo Villaseñor Mir[4], Víctor Zamora Villa[6], Ivan Ortiz-Monasterio[1], Thomas Payne[1], Javier Peña[1], Matthew Reynolds[1], Pawan Singh[1], Andrzej Kilian, Peter Wenzl[1]*

[,1]International Maize and Wheat Improvement Centre, El Batan, Mexico. [2]Universidad Politécnica de Francisco I. Madero, Hidalgo, Mexico. [3]Universidad Autónoma del, Estado de México, México, Mexico. 4 INIFAP, México, Mexico. [5]Laboratorio Nacional de genómica para la Biodiversidad, México, Mexico. [6]Universidad Autónoma Agraria Antonio Narro, Saltillo, Coah. Mexico.*

Wheat, world's most important calorie source is a staple food crop and in coming decades, its demand is projected to increase by 60%. Increasing population pressure, climate change, shrinking farm resources and reduced genetic diversity are the major challenges for ensuring global food security. Broadening of the genetic base is very important step in accelerating the progress of breeding programs. One of the ambitious initiatives of CIMMYT, Seeds of Discovery (SeeD) has made efforts to characterize and mobilize novel, useful genetic variation from landraces, wild relatives and other underutilized sources into adapted elite genotypes. (1) Genome-profiling of more than 50,000 wheat gene bank accessions were carried out using a genotype by sequencing (GBS) platform. (2) In way to identify trait donors approximately 15,000 wheat landraces were characterized for heat and drought, ~20,000 gene bank accessions for grain quality and 6000 for diseases. (3) Wheat landrace core sets developed using GBS and phenotype information represent a unique resource. (4) Bridging germplasm was also developed for mobilization of useful phenotypic variations from exotics to elite germplasm. The linked top cross population panels (LTP) were developed in which exotic alleles were mobilized from 200 diverse accessions (landraces and primary synthetics) to farmer adapted elite cultivars. Within panels populations were linked through common elite parents. These resources will be used for Genomic selection (GS) and nested association mapping (NAM). The SeeD-wheat project has a unique balance between conventional and advanced breeding methods as well as between germplasm characterization and product delivery. Wheat researchers world-wide can utilize these resources for targeted wheat genetic improvement.

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Development of biofertilizers in Oaxaca

Ma. Nieves Trujillo Tapia[1] and Eustacio Ramírez Fuentes[2]

Instituto de Ecología[1], Instituto de Recursos[2]

Universidad del Mar, Puerto Ángel, Oaxaca. México.e-mail: nieves@angel.umar.mx

The indiscriminate use of inorganic fertilizers has reduced the fertility of agricultural soils in general and in particular in Oaxaca, reducing production and profits for farmers. Seeking for Biotechnological alternatives to bring sustainability to the Mexican countryside, and reduce the negative effects of inorganic agrochemicals, without sacrificing crop yield, a search for various sources of organic fertilizers and biological fertilizers, set on the coast of Oaxaca, developed a biological fertilizer based on blue-green algae (cyanobacteria) to fix nitrogen in costal agricultural soils of Oaxaca. Cyanobacteria are photoautotrophic free-living microorganisms found in a variety of habitats, but mainly in those who are poor in nutrients, or have suffered anthropogenic disturbances. So the aim of this study was to isolate, identify and purify cyanobacteria. Three strains of filamentous cyanobacteria (*Fischerella sp. Anabaena sp.* and *Nostoc sp.*) were isolated. The three strains were grown in liquid medium with constant aeration BG110 and then the culture was scaled up to 240 liters. Once scaling parameters were optimized, growing cyanobacteria were filtered and tested in a corn (H-520) crop, rain-fed, in three experimental plots and compared with three commercial organic products. The cost-benefit ratio in rain-fed maize production using cyanobacteria was better (1.6) in comparison to fertilization with biological commercial products offered by *Quimcasa*, *CAMEX* and *Nocon* companies. Also, the amount of applied chemical fertilizers (nitrogen) could be reduced by more than 50%, so reducing the negative impact on the environment by the application of inorganic salts. The use of nitrogen-fixing filamentous cyanobacteria may represent a good alternative to organic fertilization for maize cultivation in the coastal region of the state of Oaxaca.

Green: Wildlife, Ecology & Environment Biotechnology

International Biotechnology Color Journal

Evaluation of antimicrobial and insecticidal proteins in arachnid venoms

Elba Villegas[1,], Patricia Olivos[2], Pavel Padilla[2], Pamela Cordero[1], Verónica Obregón[1], Laura Lina[1], Ninfa Rosas[3], Nayely Espinoza[2], Alexis Rodríguez[1], Gerardo Corzo[4]*

*[1] Centro de Investigación en Biotecnología (CEIB), [2]Facultad de Ciencias Biológicas, Universidad Autónoma del Estado de México (UAEM), México. [3]Centro de Biotecnología Genómica, Instituto Politécnico Nacional. [4]Instituto de Biotecnología, Universidad Nacional Autónoma de México, *E-mail:*.* elbav@uaem.mx

Spider venoms are complex mixtures of biologically active compounds such as, proteins, antimicrobial peptides, enzymes, nucleotides, amino acids, lipids, and biogenic amines, among other compounds. Antimicrobial peptides (AMPs) and insecticidal specific toxins (IST), once purified and characterized can be chemically synthesized for further research. L-Pin2 is an antimicrobial peptide isolated from the venom of scorpion and interacts with the outer and cytoplasmic membranes of bacteria. PaluIT1 is an insecticidal peptide isolated from the venom of spider. It binds the para-type voltage-gated sodium channel of insects altering its inactivation properties. In this work, we present the potential use of an enantiomer of L-Pin2, D-Pin2, as therapeutic agent and the insecticidal properties of PaluIT1 towards Lepidoptera species. D-Pin2 was more stable to human serum and to *Pseudomonas sp*. proteases than that of L-Pin2. Furthermore, D-Pin2 was less toxic towards human erythrocytes than L-Pin2. On the other hand, PaluIT1 median lethal dose (LD50) was determined towards five different Lepidoptera larvae, interestingly PaluIT1 is very effective towards *Spodoptera exigua* but it was not towards *Heliothis viriscens*.

Interaction ruminal bacteria and food particles: usefulness as symbiotic

Mónica Marcela Galicia-Jiménez

Institute of Genetics. Universidad del Mar, Campus Puerto Escondido, Mexico.

The use of different diets to ruminants has focused on finding higher production through diets for the animal, with an effect on rumen microbial population. However, the production efficiency has been reported to vary inconsistently. One option could be daidzein, which have a role in plant-microbe interaction. Our objective is to integrate knowledge of this interaction, understanding the signaling pathways used by bacteria in the rumen for the colonization of food particles and its degradation, to then manipulate rumen fermentation, and finally creating the symbiotic cultures for cattle. By means of *in silico* studies putative genes involved in *Ruminococcus albus* chemotaxis were identified. Also, daidzein was found useful in 4 plants with forage potential. A chemotaxis assay for rumen bacteria was standardized by modifying the known aerobic capillary method in combination with the technology used for the measurement of gas production *in vitro*, which demonstrated a chemoattractant effect of daidzein.

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Design of short alpha-helical peptides and their activity against pathogenic bacteria *Mycobacterium tuberculosis*

Alexis J. Rodríguez-Solís[1,2], Elba C. Villegas-Villarreal[1], Alejandra Montoya-Rosales[3], Bruno Rivas-Santiago[3], Gerardo A. Corzo-Burguete[1]

[1]Departamento de Medicina Molecular y Bioprocesos, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca Morelos, México, [2]Centro de Investigación en Biotecnología, Universidad Autónoma del Estado de Morelos, Cuernavaca, Morelos, México, [3]Medical Research Unit-Zacatecas, Mexican Institute of Social Security, UIMZ-IMSS, Zacatecas, Mexico.e-mail: corzo@ibt.unam.mx

The contention of multidrug-resistant bacteria requires the use of new antibiotics. Pandinin 2 (Pin2) is a highly hemolytic antimicrobial peptide that has a central proline residue. Proline forms a structural ‘‘kink’’ linked to its pore-forming activity towards human erythrocytes. In this work, the residue Pro14 of Pin2 was both substituted and flanked by Glycine residues pursuing the low hemolytic activities of antimicrobial peptides as Magainins and Ponericins, respectively. Both Pin2 variants showed antimicrobial activity against *E. coli, S. aureus*, and *M. tuberculosis*. However, Pin2 [GPG] was 30% less hemolytic. To avoid the drawback associated to the cost of synthesis of large peptides, two short peptides were designed and synthesized based on Pin2 [G] and Pin2 [GPG]. Both Pin2 [G] and Pin2 [GPG] short variants, showed antibiotic activities against *E. coli* and *M. tuberculosis*. Besides, short Pin2 [G] presented only 25% of hemolysis at 100 μM, while the peptide short Pin2 [GPG] did not show any hemolytic effect at the same concentration. Furthermore, these short antimicrobial peptides showed better activity at molar concentrations against multidrug resistant *M. tuberculosis* than conventional antibiotics. Pin2 [G] and Pin2 [GPG] short variants have the potential to be used as alternative antibiotics with reduced hemolytic effects.

Use of biotechnology in controlling preharvest aflatoxin contamination, a major agricultural problem

Deepak Bhatnagar

USDA/Agricultural Research Service, New Orleans, Louisiana 70124, USA. deepak.bhatnagar@ars.usda.gov

Aflatoxin is a human carcinogen and acutely toxic to humans, pets, livestock, and wildlife. It is produced by the fungus *Aspergillus flavus* prior to harvest when it invades crops such as maize, peanuts, treenuts, and cottonseed, thereby greatly reducing their value and marketability. This contamination is a worldwide problem, especially in the warmer parts of the world. Researchers from around the world have attempted to control this contamination using traditional methods such as crop management and breeding resistant crops; these approaches have found limited success. The use of biotechnology has enabled researchers worldwide to decipher and find solutions much more rapidly too many health and agriculture problems. These technologies are now being applied to understand the aflatoxin contamination problem, and to develop strategies to control this significant agricultural issue. The methods used are the study of the genomics of the fungus, proteomics of the crops, metabolomics and transcriptomics to decipher the host-plant fungal interactions.

Exploring gut microbiota alterations associated with HIV infection using 16S rRNA sequencing and the Ion Torrent PGM

Sandra María Pinto-Cardoso

Centro de Investigación en Enfermedades Infecciosas (CIENI) & Instituto Nacional de Enfermedades Respiratorias (INER). E-mail: sandra.pinto@cieni.org.mx

HIV-1 infection induces a rapid and massive depletion of gut TCD4+ cells. Loss of these cells in the gut-associated lymphoid tissues (GALT) allows for the translocation of bacterial products to the systemic circulation. Microbial translocation is a contributing factor to systemic immune activation and disease progression. As the immune system in the GALT plays such an important role in shaping the gut microbiota composition, and considering the prevalence of gut-linked diseases in HIV infection, it is important to understanding how alterations in the gut microbiota composition may influence the progression of the infection. I will share our experience at the CIENI, the center for research in Infectious Diseases, on analyzing and comparing diversity and composition of microbial communities in fecal samples from HIV-positive samples (people living with HIV) with or without combined antiretroviral treatment (cART) and HIV negative samples using both an "in house" research method for amplifying and sequencing the V3 region of the 16S rRNA gene, as well as the Ion Torrent Metagenomics 16S kit (as part of the early access program, with CIENI as participant).

International Biotechnology Color Journal

SAGA – A high-throughput genotyping platform for Mexican agriculture

Petroli César D[1], Sansaloni Carolina P[1], Galvez Hector J, Martinez Manuel H, Sierra Aleyda A, Campos Jaime A, Hearn Sarah1, Soca Giovanny[1], Kilian Andrzej[2], Wenzl Peter[1]

[1]CIMMYT – International Maize and Wheat Improvement Centre, Mexico.

[2]DArT PL – Diversity Arrays Technology, Australia. e-mail: cesar@cenargen.embrapa.br

Human population is growing fast, natural resources are decreasing and food security of coming generations is at risk. Crop genetic resources are one of few options left to continue raising food production to feed humanity by 2050. The MasAgro-Biodiversidad project, the most upstream component of the Mexican Government-funded MasAgro program, systematically characterizes and mobilizes novel genetic variation from maize and wheat genebanks into breeding programs. A strategic partnership was established with DArT PL (www.DiversityArrays.com) to create , in Mexico, a "Genetic Analysis Service for Agriculture" (SAGA – Servicio de Análisis Genético para la Agricultura). SAGA is on its way to generate tens of thousands of high-density genome profiles from DNA samples using the “DArTseq” Genotyping by Sequencing (GBS) method developed by DArT PL. Each genome profile typically generates several tens of thousands of both Single Nucleotide Polymorphism (SNP) and Presence/Absence Variation (PAV) molecular markers. SAGA uses a HiSeq2500 next-generation sequencer (Illumina) and DNA-barcode-enabled multiplexing of individual samples. The principal goal of SAGA is to genotype more than 100,000 maize and wheat samples from genebanks at CIMMYT, INIFAP and others organizations. SAGA also offers advice and support in the interpretation and application of GBS data for a variety of applications. All data generated will be made available to the public via an internet portal. This data will contribute to diversifying the genetic base of maize and wheat breeding programs in order to accelerate the development of high-yielding and climate-read cultivars of two of the three most important crops of humanity. As the number of samples analyzed for MasAgro-Biodiversidad decreases, and subject to sufficient demand, SAGA will seek to offer its GBS-based genome-profiling services to the Mexican research community at large.

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Introduction - status and global networks to address this issue

William Moar.

Corn IRM Technical Lead. Tel. (314) 694-7793. . E-mail: william.moar@monsanto.com

Maintaining the durability of Bt crops against targeted pests globally presents challenges due to differences in pest biology, product performance, and grower needs and capabilities. Recent initiatives to maintain or increase durability of Bt crops include gene pyramiding, enhanced refuge options such as refuge-in-a-bag (RIB), and industry alignment through ETS (Excellence through Stewardship) that will improve the effectiveness and consistency of IRM programs. This presentation will expand on these topics and discuss how industry is moving forward to provide more tools for insect control globally.

Alternative splicing and highly variable cadherin transcripts are associated with field-evolved resistance of pink bollworm to Bt cotton in India

Jeffrey A. Fabrick[1], Jeyakumar Ponnuraj[2], Xianchun Li[3], Yves Carrière[3], Bruce E. Tabashnik[3]

1 USDA-ARS, U.S. Arid Land Agricultural Research Center, Maricopa, Arizona, United States of America. E-mail: Jeff.Fabrick@ARS.USDA.GOV .

2 National Institute of Plant Health Management, Rajendranagar, Hyderabad, Andhra Pradesh, India.

3 Department of Entomology, University of Arizona, Tucson, Arizona, United States of America.

Evolution of resistance by insect pests can reduce the benefits of insecticidal proteins from *Bacillus thuringiensis* (Bt) that are used extensively in sprays and transgenic crops. Despite considerable knowledge of the genes conferring insect resistance to Bt toxins in laboratory-selected strains and in field populations exposed to Bt sprays, understanding of the genetic basis of field-evolved resistance to Bt crops remains limited. In particular, previous work has not identified the genes conferring resistance in any cases where field-evolved resistance has reduced the efficacy of a Bt crop. Here we report that mutations in a gene encoding a cadherin protein that binds Bt toxin Cry1Ac are associated with field-evolved resistance of pink bollworm (*Pectinophora gossypiella*) in India to Cry1Ac produced by transgenic cotton. In laboratory diet bioassays we confirmed previously reported resistance to Cry1Ac in pink bollworm from the state of Gujarat, where Bt cotton producing Cry1Ac has been grown extensively. Analysis of DNA from 436 pink bollworm from seven populations in India detected none of the four cadherin resistance alleles previously reported to be linked with resistance to Cry1Ac in laboratory-selected strains of pink bollworm from Arizona. However, DNA sequencing of pink bollworm derived from resistant and susceptible field populations in India revealed eight novel, severely disrupted cadherin alleles associated with resistance to Cry1Ac. For these eight alleles, analysis of complementary DNA (cDNA) revealed a total of 19 transcript isoforms, each containing a premature stop codon, a deletion of at least 99 base pairs, or both. Seven of the eight disrupted alleles each produced two or more different transcript isoforms, which implicates alternative splicing of messenger RNA (mRNA). This represents the first example of alternative splicing associated with field-evolved resistance that reduced the efficacy of a Bt crop.

Insect nutritional ecology and environmentally-mediated variation in Bt susceptibility...

G. A. Sword[1], C. Deans[1], P. Tamez-Guerra[2], S.T. Behmer[1] .. .

1 Department of Entomology, Texas A&M University, College Station, TX, USA 77843-2475.

2 Fac. Ciencias Biológicas, UANL. AP 46-F. San Nicolás de los Garza N.L. E-mail: patricia.tamezgr@uanl.edu.mx

Insecticide resistance in agricultural pest species represents a costly and ever-increasing problem that poses significant challenges for meeting the nutritional demands of our growing global population. The evolution and spread of genetic mutations conferring resistance is the primary explanation for observations of low pesticide efficacy and resulting poor pest population control in agricultural systems. Unfortunately, in many cases it takes years to identify the mutations and alleles responsible for these resistant phenotypes, with most incidents assumed to be the result of mutations without any substantiation. As a result, the potential for gene-by-environment interactions to play a dominant role in mediating pesticide resistance is often overlooked. One environmental factor that has been shown to be highly variable is plant macronutrient content. Macronutrients such as protein (P) and carbohydrates (C) have also been shown to strongly affect insect behavior, physiology, and performance, including detoxification potential. Studies have shown that dietary protein-to-carbohydrate ratios (P:C) can influence feeding behavior and detoxification ability in locusts, as well as immune function in caterpillars, indicating that the impact of nutritional variability on insecticide resistance is probative. In this study we explored the potential for plant macronutrient content to impact susceptibility to Cry1Ac endotoxin, found in transgenic cotton, in the generalist herbivore *Helicoverpa zea*. Using artificial diets, we mimicked the macronutrient content of different cotton tissues and reared larvae on diets either without Cry1Ac, with a low concentration of Cry1Ac, or with a high concentration of Cry1Ac. We then measured the main and interactive effects of Cry1Ac and diet macronutrient content on larval survival, pupal mass, and hatching success. Our results clearly demonstrate an effect of specific diet macronutrient content on Cry1Ac susceptibility and suggest that variation in insect performance in both the field and laboratory assays may be at least partially attributed to variation in diet nutritional quality.

Analysis of the immune response in wild Helicoverpa zea (Lepidoptera: Noctuidae) populations and host relationship

P. Tamez Guerra[*,1], *M. Martínez-Valenzuela*[1], *J. A. Valadez-Lira*[1], *P. Pietrantonio*[2], *R. A. Gómez-Flores*[1], *C. Rodríguez-Padilla*[1]

1 Fac. Ciencias Biológicas, UANL. AP 46-F. San Nicolás de los Garza N.L. E-mail: patricia.tamezgr@uanl.edu.mx

2 Insect Toxicology, Dep. Entomology, Texas A&M Univ. 2475 TAMU. College Station, TX-USA, 77843.

The expression of genes encoding to lysozyme (lis), three antimicrobial peptides (AMPs) (galiomicin (gal), cecropin (cec) and gloverina (glo) and peptidoglycan receptor protein (pgrp c), were evaluated by real-time RT-PCR, using mRNA sequences reported in H. armigera to assess differences related to larvae growth instar, exposure to host plant, and Bt as a commercial product or as Cry toxins in transgenic crops. Transcripts amplification was analyzed in 2nd, 4th and 5th instars: i) collected from conventional (C) or transgenic crops (Ct) in field conditions; ii) collected among different geographical areas from Mexico and USA; iii) exposed under three laboratory conditions (LB): a) after exposure to conventional cotton plants (LBA), b) exposed to Bt-cotton (LBAt), or c) to Bt commercial product Bactospeine® (LBBt) on artificial diet. Identified sequences were reported in the GenBank. Results found in larvae reared on artificial diet with Bt-cotton (LBAt), cec expression was repressed in 2nd instar, whereas Bactospeine® (LBBt) repressed gal in 4th instars. By comparing the expression in larvae fed on C and LBA, pgrp c, gal and lis were suppressed in the 2nd instar fed on C, whereas glo was suppressed in the 4th instar, and gal was suppressed in the 5th instar. By increasing exposure time, in LBA and LBAt expression of LIS in the 5th instar was stimulated. Larvae collected form field crops resulted in AMPs expression upon the variety and crop conditions. In fact, the highest expression of AMPs and lis was among larvae collected from crops subjected to drought stress and Bt-transgenic crops in Sinaloa state.

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Cry1Ac protoxin from Bacillus thuringiensis affects the fitness of Helicoverpa zea B. (Lepidoptera, Noctuidae)

B. Pereyra-Alférez, J.O. Medina-López, S.A. Martínez-Morales, F. Molina-Estrada, L.J. Galán-Wong

Instituto de Biotecnología. Facultad de Ciencias Biológicas - UANL. Pedro de Alba y Manuel L. Barragán s/n. Cd. Universitaria. San Nicolás de los Garza, NL. CP. 66451. México. E-mail: bpereyra@gmail.com

Biotechnological cotton known as Bollgard II™ express Cry1Ac and Cry2Ab *B. thuringiensis* toxins, which control two of main lepidopteran cotton pests: *Pectinophora gossypiella* (pink worm) and *Helicoverpa zea* (corn earworm). However, the constant pressure of selection could to drive the apparition and evolution of resistance. At laboratory level there is evidence of resistance to Cry1Ac for more than 500 cases. Our goal was to obtain colonies of *H. zea* with resistance to the protoxin of Cry1Ac and to know the how this trait is transmitted to offspring. Wild type insects (eggs, pupae and adults) were collected and established in our laboratory. Laboratory diet bioassays demonstrated a baseline (LC50) of 1.3 µg g-1 diet for Cry1Ac protoxin. From this susceptible colony, we obtained resistant colonies to 10, 20 and 50 µg g-1 diet. Resistant colonies showed a lower fitness, high mortality at pupae phase, a progressive decay of amount of progeny, and an unusual male:female rate (2:1). Last result is controversy, because currently female amount is higher in male comparison. Another important result was the observation of very lower egg fertility (< 5%) and after 4th matting generation, fully eggs were no fertile. Interesting was to note that when resistant female was matting with wildtype males, the eggs were no fertile. Nowadays, we are trying to demonstrate changes in genetic expression of several important genes involved in resistance such as cadherin, aminopeptidades and alkaline phosphatase, putative receptor(s) of Cry1Ac in *H. zea.*

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What could be the benefits of planting genetically-engineered maize in Mexico?

Carlos A. Blanco[1], José Guadalupe Pellegaud[2], Urbano Nava-Camberos[3], David Lugo-Barrera[4], Paulina Vega-Aquino[2], Jesús Coello[5], Antonio P. Terán-Vargas[6], Jesús Vargas-Camplis[6].

1 Biology Department. The University of New Mexico. E-mail: cblancom@unm.edu.

2 Los Mochis, Sinaloa, México.

3 Universidad Juárez del Estado de Durango, México.

4 Dow AgroSciences, México.

5 Grupo MASECA, México.

6 INIFAP, México.

Mexico is the fifth largest maize producer of the world, representing close to a third of its internal agricultural production. Paradoxically, Mexico imports a third of the maize that it consumes, and its internal production yields 40% below of the world's average. Arthropod pests are partially responsible for lowering Mexican pest production. A group of Mexican scientists and crop advisors conducted a census of the impact of arthropod pests throughout the country. Results indicate that *Spodoptera frugiperda*, the most problematic pest, requires up to 3 sprays per crop season, representing 3,000 tons of insecticidal active ingredients (a.i.) for its control, followed by other destructive Lepidoptera such as *Agrotis ipsilon* and *Helicoverpa zea*, requiring also 1-3 insecticide applications per crop cycle, amounting to 2.8 and 0.8 tons of insecticidal a.i. for its control, respectively. Coleopteran pests require less insecticide than the previous pests and have narrower special distribution. The white grub complex (*Phyllophaga spp.*) is controlled in only a few Mexican regions with 1-2 insecticide applications, representing up to 1.6 tons of a.i., while *Diabrotica spp.* only requires an average of 1.5 applications but in a larger area of the country, amounting to 0.4 tons of a.i. Since genetically engineered (GE) maize hybrids have proven to be effective against these pests, and because these arthropods occur at different times of the maize development, in some regions of Mexico this crop is sprayed multiple times to control Lepidoptera and Coleoptera that otherwise could be effectively managed with certain GE maize hybrids, with the potential of reducing thousands of insecticidal active ingredient.

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Susceptibility of the fall armyworm, Spodoptera frugiperda to Bt toxins and conventional pesticides among different maize production systems in the US and Mexico

Rebeca Gutiérrez-Moreno[1], David Mota-Sánchez[1], J. C. Rodríguez-Maciel[2], Carlos A. Blanco[3], Mark E. Whalon[1].

1 Department of Entomology, Michigan State University, 2COLPOS,

3 Biology Department. The University of New Mexico. Email: carlos.a.blanco@aphis.usda.gov

The fall armyworm (FAW) *Spodoptera frugiperda* (J.E. Smith) is one of the main pests of corn in many areas of the American continent. If uncontrolled this insect causes severe damage to the crop. Genetically modified (GM) corn with the ability to produce Bt proteins is regarded as a major tool for pest control in modern and high-tech agriculture, but recent isolated cases of resistance of targeted pests threatens the useful life span of this technology. While this technology is a major method of controlling corn insect pests in the U.S., in Mexico pest control strategies are based almost entirely in the use of conventional pesticides. For instance, in the past few years in some areas of Mexico it has been necessary to spray up to six times during the season to control FAW. The overuse of pesticides for FAW control may lead to development of insect resistance more quickly than with use of GM Bt corn. Despite the economic important of FAW in the Americas there are few studies that provide information about the adaptation of this insect to insecticides whether applied as a spray or deployed through genetic modification of the plant. Our hypothesis is that FAW from Mexico would exhibit less susceptibility to conventional insecticides whereas FAW from the US would exhibit less susceptibility to Bt proteins. We are conducting bioassays to determine the susceptibility of FAW to Bt proteins and five conventional pesticides (chlorpyriphos, spinetoram, permethrin, flubendiamide and methomyl) in FAW populations from Mexico and the US.

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Susceptibilities of geographic populations of *Helicoverpa zea* (Boddie) in Mexico to Bt ∂-endotoxins Cry1Ac and Cry2Ab; a 13 year study

Sotero Aguilar-Medel[1], J. C. Rodríguez[2,], J. L. Martínez-Carrillo[3], Ovidio Díaz-Gómez[4], Gonzalo Silva-Aguayo[5]*

1 Universidad Autónoma del Estado de México, Centro Universitario Tenancingo, Tenancingo, Estado de México. México.

2 Colegio de Postgraduados, Instituto de Fitosanidad. Montecillo, Estado de México. México. * concho@colpos.mx.

3 Instituto Tecnológico de Sonora. Cd. Obregón, Sonora, México.

4 Facultad de Agronomía. Universidad Autónoma de San Luis Potosí. San Luis Potosí, México.

5 Departamento de Producción Vegetal, Facultad de Agronomía, Universidad de Concepción. Chillán. Chile.

An insect resistance monitoring program was developed for Mexico to accommodate the commercial introduction and stewardship of Bt cotton. In 1998 and 2003 field-collected geographic populations of the cotton bollworm *Helicoverpa zea* (Boddie) were evaluated against *Bacillus thuringiensis* Berliner crystalline δ-endotoxins Cry1Ac, and Cry2Ab, respectively, to establish baseline susceptibility data in preparation for the commercial introductions of Bollgard® (expressing Cry1Ac) and Bollgard® II (expressing Cry1Ac and Cry2Ab) cottons, respectively. The sub-lethal response of growth inhibition was evaluated as measured by larval weight reduction (Inhibition of Weight: IW) and inhibition of normal progression through developmental stadia (Inhibition of Development: ID). Informed by the baseline susceptibility data, an annual monitoring program was subsequently established under which a single diagnostic concentration of Cry1Ac and Cry2Ab was used in diet bioassays to test for continued "normal" susceptibility to these insecticidal proteins among geographic populations. The diagnostic concentration selected was suitable since it aligned with previous work and represented the concentration under which larvae evaluated in baseline studies were reduced in weight by 98% or more (362pprox. IW98) relative to untreated controls, or under which 97% or more failed to molt to 3rd instar (362pprox. ID97), after 5 days of exposure. In the monitoring study, populations were tested against Cry1Ac from 1998 through 2010, and against Cry2Ab from 2002 through 2004, and again from 2007 through 2010. None of the Cry1Ac-exposed larvae tested during the 13 year period reached the third larval instar by five days and mass reduction relative to untreated control larvae was uniform at 98 to 99%. For the seven years of Cry2Ab monitoring, no treated larvae reached third instar, and mass reduction was uniform at over 98% relative to controls in five-day assays. These results illustrate the value of a single diagnostic concentration and sub-lethal response criteria to monitor susceptibility of target pests to Bt proteins expressed in crops over time and suggest that susceptibilities to the Cry proteins expressed in Bollgard (Cry1Ac) and Bollgard II (Cry1Ac; Cry2Ab) cottons have not changed during the period these technologies have been grown in Mexico. This report is also unique since it is one of relatively few internally consistent long-term studies on resistance monitoring for this or any other geography.

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What has happened in Mexico after nineteen years of releasing pest resistant GM cotton? An efficacy analysis related to Bt crops based on environmental release reports

M. Andrea Orjuela-R., Francisca Acevedo Gasman

Coordinación de Análisis de Riesgo y Bioseguridad, Comisión Nacional para el Conocimiento y Uso de la Biodiversidad. Liga Periférico-Insurgentes Sur 4903, Parques del Pedregal, Tlalpan 14010, México D.F. E-mail: morjuela@conabio.gob.mx, facevedo@conabio.gob.mx

The first GM resistant cotton was probably released in Mexico during 1995 in the State of Tamaulipas, in a permitted area comprising 35 ha in the municipios of Altamira and Aldama. From this moment on, eight GM cotton events containing at least one *Bacillus thuringiensis* (Bt) gene have been given release permits in different regions of the North of the country. Out of these eight GM cotton events, six have been developed by Monsanto, one by Bayer and one by Dow AgroScience. Three GM cotton events accumulate the highest number of release permits granted by the Mexican authorities, these are: Bollgard cotton (MON-ØØ531-6), requested to be released in the States of Baja California, Sonora, Chihuahua, Coahuila, Durango, Colima, Sinaloa, San Luis Potosí, Tamaulipas and Veracruz between 1995 and 2010; Roundup Ready™ Bollgard™ cotton (MON-ØØ531-6 x MON-Ø1445-2), which accounts for release solicitations in the States previously mentioned as well as Baja California Sur, and released between 1997 and 2010; and Roundup ready™ Flex™ Bollgard II™ cotton (MON-88913-8 x MON-15985-7) which started being released in the year of 2003 and has been solicited for different localities in the states of Baja California, Sonora, Chihuahua, Coahuila, Durango, Sinaloa, San Luis Potosí, Tamaulipas, Veracruz and Zacatecas. This last GM cotton event is still being released nowadays. Only the stacked GM events Roundup Ready™ Bollgard™ and Roundup Ready™ Flex™ Bollgard II include experimental, pilot and commercial phase releases as well as reports of their performance during several of these releases. The most important cotton pests in Mexico are the pink bollworm (*Pectinophora gossypiella*) and the boll weevil (*Anthonomus grandis*) while others are of a secondary nature including the tabacco budworm and corn earworm complex (*Heliothis virescens* y *H. zea*). The eight Bt cotton events that have been released in the country have at least one or several of the following Cry genes: Cry1Ab, Cry1Ac, Cry2Ae y Cry1F. A recent permit was also given to a stacked VIP3(a) containing GM event. Cry1Ab, Cry1Ac, Cry1F and Cry2Ae have shown efficacy, among other insects, to *Heliothis virescens*, *H. zea*, a lepidopteran complex identified as a secondary plague in the different cotton producing States of the country. The Cry1Ac gene is part of seven GM cotton events released in Mexico; the Cry1Ab and Cry1F genes are present each in one GM cotton event released in Mexico, while Cry2Ae is only present in Roundup ready™ Flex™ Bollgard II (MON-88913-8 x MON-15985) together with Cry1Ab. Given this context, it is of our interest to contrast, by revising and analyzing the data included in the environmental release reports generated in Mexico, how these GM cotton events have shown to be effective in the control of cotton plagues, in the States in which the permits have been given by the competent authority. It is because of this reason that in this presentation we will address the following question: Do GM cotton events show efficacy in relation to the present cotton plagues in the areas of release? We show a preliminary analysis based on report data generated during releases in the Mexican territory.

Studies of Cry3Aa-intoxication identify strategies to increase potency

Brenda Oppert[1], Elena N. Elpidina[2]

1 USDA ARS Center for Grain and Animal Health Research, Manhattan, KS, USA. E-mail: bso@ksu.edu

2 A.N. Belozersky Institute of Physico-Chemical Biology, Moscow State University, Moscow, Russia.

The yellow mealworm, *Tenebrio molitor*, is sensitive to the Cry3Aa coleopteran-specific toxin from *Bacillus thuringiensis* (Bt) but is insensitive to the lepidopteran-specific toxin Cry1Ac. These two toxins were used to evaluate differences in gene expression in *T. molitor* larvae exposed to Cry toxin for 12 hours, a time point that was previously determined to provide the greatest difference in gene expression. Approximately 237 million paired-end sequence reads (250 bp insert size) were obtained from control (no toxin), Cry1Ac-fed (negative control), or Cry3Aa-fed larvae, with triplicate biological replicates for each treatment group (nine samples total). Statistically significant changes in gene expression were found in Cry-treated compared to control larvae (Student t-test, ≥ 90 C.I. and >8-fold change). However, there were only 23 genes that were differentially-expressed in Cry1Ac-challenged larvae, whereas there were 438 genes (428 specific to Cry3Aa) differentially expressed genes in Cry3Aa-challenged larvae, reflecting the relative sensitivity of *T. molitor* larvae to these toxins. Using Blast2GO, we determined that most of the differentially expressed genes are involved in metabolic and cellular processes, binding, and catalytic activity. These changes are likely related to the cessation in feeding that occurs in intoxicated larvae and induction of immune-like defenses that we have observed in previous studies. The most severely repressed transcripts (<0.02-fold) in Cry3Aa-intoxicated larvae included lipase, serine proteases (8), dipeptidyl peptidase, and hexamerin (2); the most highly increased transcripts (>50-fold) were mostly unidentified or hypothetical proteins (9), serine and cysteine proteases, lysosomal proteins (2), lipase, and transport-related proteins (2). Bioassays indicate that protease inhibitors can reduce the time to kill and increase mortality in *T. molitor* larvae exposed to sublethal doses of Cry3Aa. These data demonstrate that understanding insect responses to Bt toxins is valuable, and can be used to increase the potency of coleopteran specific Bt toxins.

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Effect of Bacillus thuringiensis Cry3Aa toxin on the expression of gut peptidases in *Tenebrio molitor* larvae

Elena N. Elpidina[1], Tural Y.O. Yarachmedov[2], Brenda Oppert[3]

1 A.N. Belozersky Institute of Physico-Chemical Biology, Moscow State University, Moscow, Russia, elp@belozersky.msu.ru.

2 Faculty of Bioengineering and Bioinformatics, Moscow State University, Moscow, Russia.

3 USDA Agricultural Research Service, Center for Grain and Animal Health Research, Manhattan, KS, USA.

We studied *Bacillus thuringiensis* Cry3Aa intoxication and the expression of insect gut peptidases in *Tenebrio molitor* larvae. Larvae digest protein by cysteine (papain family) peptidases in the anterior midgut and serine peptidases (chymotrypsin family) in the posterior midgut. The data demonstrate that after 12 h of intoxication, 11/26 cysteine peptidase transcripts were significantly increased in expression (2-4-fold), including the main digestive cathepsin B, but after 24 h these transcripts mostly were decreased. Serine peptidase transcripts included 88 presumably active peptidases and 104 inactive homologs. Thirty two transcripts of 88 significantly changed expression after 12 h intoxication, and changes ranged from approximately 6-fold increase to 20-fold decrease. Expression of the main digestive trypsin and chymotrypsin mRNAs decreased. Among different groups of serine peptidases with altered expression levels, only chymotrypsin-like peptidases were significantly increased after 12 h intoxication. By 24 h, the changes in expression were similar to the 12 h profile. The majority of serine peptidase homolog mRNAs were decreased in expression. The severe change in expression of serine peptidases induced by intoxication may be associated with the close proximity of secreting cells to the lesion focus in the posterior midgut. At the same time, the insect maintained the production of critical digestive cysteine peptidases synthesized and located in the anterior midgut. The data on suppression of insect digestive system after 24 h intoxication correlate to observations of intoxicated larvae and cessation of feeding.

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Countering multiple resistance with modified Bt toxins

Bruce E. Tabashnik[1], Jeffrey A. Fabrick[2], Alejandra Bravo[3], Mario Soberón[3]

1 Department of Entomology, University of Arizona, Tucson, Arizona, United States of America. E-mail: brucet@cals.arizona.edu

2 USDA-ARS, U.S. Arid Land Agricultural Research Center, Maricopa, Arizona, United States of America.

3 Instituto de Biotecnología, Universidad Nacional Autónoma de México, Cuernavaca, Morelos, México.

Evolution of resistance in pests threatens the long-term efficacy of insecticidal proteins from *Bacillus thuringiensis* (Bt) used in sprays and transgenic crops. For example, field-evolved resistance to Bt cotton producing the single Bt toxin Cry1Ac has been documented for pink bollworm (*Pectinophora gossypiella*) in western India. In laboratory diet bioassays, we found that the genetically modified Bt toxins Cry1AbMod and Cry1AcMod effectively countered pink bollworm resistance to the native Bt toxins Cry2Ab, Cry1Ab and Cry1Ac. Resistance ratios based on the concentration of toxin killing 50% of larvae for a lab-selected resistant strain relative to a susceptible strain were 210 for Cry2Ab, 270 for Cry1Ab, and 310 for Cry1Ac, but only 1.6 for Cry1AbMod and 2.1 for Cry1AcMod. For both the resistant and susceptible strains, the results show slight but significant synergism between Cry1AbMod and Cry2Ab, whereas the other combinations of toxins tested did not show consistent synergism or antagonism. The results suggest that the modified toxins may be useful for managing populations of pink bollworm resistant to Cry1Ac, Cry2Ab, or both.

Competing interests: AB, MS and BET are coauthors of a patent "Suppression of Resistance in Insects to Bacillus thuringiensis Cry Toxins, Using Toxins that do not Require the Cadherin Receptor" (patent numbers: CA2690188A1, CN101730712A, EP2184293A2,EP2184293A4, EP2184293B1, WO2008150150A2, WO2008150150A3). JAF is coauthor of a patent "Cadherin Receptor Peptide for Potentiating Bt Biopesticides" (patent numbers: US20090175974A1, US8354371, WO2009067487A2, WO2009067487A3). Pioneer, Dow AgroSciences, Monsanto and Bayer. CropScience did not provide funding to support this work, but may be affected financially by publication of this paper and have funded other work by AB, JAF, MS and BET.

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Resistance management for Bt crops: successes and failures

Bruce Tabashnik

Department of Entomology, University of Arizona. E-mail: brucet@cals.arizona.edu

Transgenic crops producing insecticidal proteins from *Bacillus thuringiensis* (Bt) decrease reliance on insecticide sprays, but evolution of resistance by pests can reduce the efficacy of these crops. A review of data from 77 studies conducted in eight countries reveals that reduced efficacy of Bt crops with practical consequences for pest control was associated with field-evolved resistance in some populations of 5 of 13 species of major pests by 2010, compared with only one such species in 2005. Factors contributing to this surge in documented cases of resistance include more extensive monitoring as well as increases in the area planted to Bt crops, the number of pest populations exposed to Bt crops, and the cumulative duration of exposure. Whereas most previous assessments characterized pest populations only as resistant or not, the new analysis introduces a series of five color-coded levels ranging from strong evidence of sustained susceptibility to the most serious cases of resistance. Field outcomes support theoretical predictions that factors delaying resistance include recessive inheritance of resistance, low initial frequency of resistance alleles, and abundant refuges of non-Bt host plants. The results imply that proactive evaluation of the inheritance and initial frequency of resistance are useful for predicting the risk of resistance and improving strategies to sustain the effectiveness of Bt crops.

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Vip3A proteins for the control of caterpillars

Juan Ferré, Maissa Chakroun, Yolanda Bel, Núria Banyuls, Baltasar Escriche

University of Valencia, Faculty of Biology, Department of Genetics, Valencia (SPAIN). E-mail: juan.ferre@uv.es

Vip3 toxins are *Bacillus thuringiensis* (Bt) proteins which are secreted to the medium during the vegetative growth and, for this reason they do not contribute to the insecticidal activity of bioinsectices prepared from Bt spore/crystal mixtures. Nevertheless, some of the Vip3 proteins are highly active against Lepidoptera and show a relatively broad insecticidal spectrum. Because Vip3 proteins share no homology with the Bt crystal (Cry) proteins and because they have different modes of action, Vip3A proteins have been combined with Cry1 proteins in transgenic crops (Bt crops). The mode of action of Vip3 proteins is not well understood. It is known that there are some insect species susceptible to Cry1 proteins which are practically completely tolerant to Vip3 proteins and vice versa. Although Vip3 proteins and Cry1 proteins share general features in their modes of action, they differ notably when the different steps are studied in detail. For example, we have found that, contrarily to the Cry1 proteins, Vip3 proteins do not have a protease-resistant core, or at least not so resistant as in the case of Cry1 proteins. This implies that the activation step within the insect's midgut is more critical in the case of Vip3 proteins because there is a dynamic equilibrium between protoxin activation and toxin degradation. A second differential feature between Cry1 and Vip3 proteins is that they bind to different membrane binding sites. This has been shown with different insect species using either labeled Cry1 proteins or labeled Vip3 proteins, and performing competition studies with unlabeled heterologous competitors. This feature means that, for resistant management purposes, it is a good strategy to combine cry1 and vip3 genes in the same plant because mutations in insect populations conferring resistance to one toxin (due to alteration of its receptors) would not confer cross-resistance to the other toxin.

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Manipulating secondary plant compounds for enhance crop protection

R.W. Kurtz

Global Lead Insect Resistance Management, Syngenta Biotechnology, Inc. E-mail: rkurts@cottoninc.com

Traditional plant breeding methods to improve a crop's ability to tolerate or reduce insect damage are well established; however, relatively little research has been done on major commodity crops exploring cisgenic or transgenic approaches for improving host plant resistance by manipulating secondary plant compounds. The purpose of this talk is to review current research in this area, discuss potential hurdles, and explore future opportunities for crop protection and resistance management.

Effects of endophytic fungi on fall armyworm, Spodoptera frugiperda Smith, and host plant damage

Ek-Ramos M.J[1]., Sword G.A.[2], Valencia C.U.2, Tamez-Guerra P.[1], Galindo-Campos A.A.[1], Terrazas-Castelán E.[1], Castillo-Velázquez U.[3], P. Weidong [4]

1 Facultad de Ciencias Biológicas, UANL. AP 46-F. San Nicolás de los Garza N. L., México.

2 Department of Entomology, Texas A&M University, College Station, TX, USA 77843-2475.

3 Facultad de Medicina Veterinaria y Zootecnia, UANL, Escobedo, N.L., México. 4Beijig Key Laboratory of Bioelectromagnetics, Institute of Electrical Engineering. The Chinese Academy of Sciences, 6 Zhougguanchun Beiertiao, Beijing 100190, China.

Fungal endophytes are microorganisms that can live within plant tissues without causing apparent damage. Among the different kinds of endophytes isolated to date, there are several that have potential beneficial uses as biocontrol agents, including endophytic entomopathogenic fungi. A recently survey isolated a range of putative beneficial fungal endophytes in different tissues of cotton cultivated in Texas. We examined the endophytic effects of one of these isolates (*Paecilomyces sp.*) along with a commercially available fungal entomopathogen (*Beauveria bassiana*), identified as strain GHA, on host plant use by fall armyworm (*Spodoptera frugiperda*). First, we measured preference and performance using no-choice tests in laboratory feeding assays. *S. frugiperda* larvae were assayed for feeding on foliage obtained from control and inoculated cotton plants. Results indicated that 4th instar larvae fed less on leaves obtained from *B. bassiana* inoculated plants and inoculation with Paecilomyces sp. does not seem to have any effect on fall armyworm feeding preference. As *Spodoptera frugiperda* is an economically important pest in both México and US, we tested the effect of both endophytes on cotton field trials during the years 2012 and 2013 in Texas. To monitor for the presence of fall armyworm, we did regular scouting along the cultivation period. We did not find evidences of fall armyworm on control, *Paecilomyce sp.*-inoculated nor *Beauveria bassiana*-inoculated plants. However early in the season we observed some moths in the periphery of our cotton field trials. In addition, in 2014 we conducted two field trials using corn and sorghum to test for the effect of *Beauveria bassiana* strain GHA and another *Beauveria bassiana* strain, isolated in México and identified as BB42. Results early in the season indicated that both *Beauveria bassiana* strains have positive effect on the germination and plant-stand up in both corn and sorghum field trials.

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Construction process of a non target organisms research network in Latin America

M. Andrea Orjuela-R., Francisca Acevedo-Gasman

Coordinación de Análisis de Riesgo y Bioseguridad. Comisión Nacional para el Conocimiento y Uso de la Biodiversidad. Liga Periférico-Insurgentes Sur 4903, Parques del Pedregal. Tlalpan 14010, México D.F. Email: morjuela@conabio.gob.mx, facevedo@conabio.gob.mx

Biotech crops have expanded their commercialized crop area to 175.2 million hectares distributed among 27 countries since 1996. Applications for the environmental release of 21 genetically modified (GM) plant species have been registered in Mexico since 1988. Some transformation events of GM cotton and soybean have reached commercial status, while GM maize has gone through experimental and pilot releases. Risk assessments consider the occurrence of possible adverse effects on the conservation and sustainable use of biodiversity, one of these being the damage that could be eventually generated on non target organisms (NTO). These kind of effects are important not only because they could translate into biodiversity loses but also because losing certain species might affect ecological functions and services provided, i.e. pollination, natural plague control or soil fertility. In order to address this subject, CONABIO promoted a workshop entitled “Advances in development of methodologies for assessment and monitoring of potential effects of genetic modified crops on non target organisms” which took place in July 2013; this work reports on this meeting. The event had an assistance of 51 participants (2 from Colombia, 2 from Brazil, 1 from USA and 46 from Mexico). The main goal was to bring together a group of researchers as well as people involved in risk assessment to learn about the subject, discuss and generate strategies to collaborate in identifying a common way forward for working together. During four days we discussed some strategies and tried to put in practice a methodology already developed in Brazil using information obtained from different Mexican sources and taking advantage of the expertise of the different researchers participating in the activity. Information gaps were identified and a directory of experts and research groups was obtained. One of the principal conclusions of the Workshop was the need to build an interactive network which could be a powerful tool for knowledge exchange between people working and living in different countries. We are now working in building this network, we have a Facebook profile named “Red Temática de Organismos No Blanco” and a twitter account @redtemonb that are the first steps in this process.

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Environmental interaction studies in support of environmental risk assessment of biotech crops

Aqeel Ahmad

Environmental Interactions Team Lead, Monsanto Company, Agronomic and Environmental, Assessment Sciences, 800 North Lindbergh Blvd. St. Louis, MO 63167. Tel: (314) 694-7150. Fax: (314) 694-8774. E-mail: aqeel.ahmad@monsanto.com

Environmental interaction evaluations are conducted as part of the plant characterization of a biotech crop and are considered in an environmental risk assessment (ERA). The purpose of environmental interaction evaluations is to assess interactions of the biotech plant with the receiving environment, including non-target arthropods (NTA), abiotic stressors, and diseases relative to a conventional control. Data generated by these evaluations are used outside of the tiered system where they are useful during problem formulation and aid in the environmental risk assessment to reduce uncertainty of unintended effects through collection of en planta data. The approach taken can be utilized across crops, product concepts, and world areas, and can be developed on a case by case basis for different regulatory requirements. An overview of the study design and key recommendations are discussed here. The design of field evaluations for the biotech plant includes replicated test, control, and reference plots in randomized blocks. The references are commercially available varieties or hybrids. The inclusion of references and the conduct of experiments at different locations provide a context for interpreting measured differences due to natural variability and local differences in agro-climatic conditions such as soil, weather, nutrients, and other abiotic and biotic stressors. This study design collects meaningful arthropod abundance data while maintaining a reasonably sized study to minimize land requirements and to allow for multiple sites in the interest of more robust study. This study focuses on arthropods closely associated with the plant, exhibiting low mobility, and a clear path of exposure (e.g., non-target herbivores) for detecting potential effects. Statistical analyses are performed on taxa present in sufficient numbers. This approach is to have a minimum level of abundance for each taxa sampled as a criterion to allow for a more robust analysis of potential treatment effects. An inclusion criterion is established where a given arthropod must have an average count per plot per collection time (across all materials) of ≥ 1. The environmental interaction data is compared between the biotech crop and the conventional control. Potential significant differences are first assessed in the context of the reference range generated from the conventional references that are also included in the design to aid in the interpretability of the data. Data from multiple sites and years allow for an evaluation of the consistency of potential differences. Local NTA field evaluations are commonly required for cultivation approvals of Bt crops often without consideration for the existing tiered approach data or other lab and field data from other geographies. The uniqueness of the agricultural setting or the diverse nature of a country are sometimes referenced as justification for local NTA field trials. As such, an assessment of the similarity of arthropod taxa across regions can help clarify how commercial maize fields compare across regions. The occurrence of similar taxa in commercial maize growing areas across regions can be used to justify the transportability of NTA data. Furthermore, any differences noted in the assessment would aid in identifying taxa that require additional consideration.

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Implementation of normalized procedures (biosafety measures, standards & technical guides) for the risk assessment of NTOs in Mexico

Jaime E. Padilla-Acero, David Lugo[1], Giovani Medina[2], Lydia González[3], Arturo de Lucas[4], Montserrat Benítez[3], Marianela Palomera[4], Josefina Perea[5], Nicolás Díaz[5], Juan Manuel De-la-Fuente[2]*

**AgroBIO México A.C. and associated member companies. E-mail:* jpadilla@agrobiomexico.org.mx

1 Dow AgroSciences México;

2 Monsanto México;

3 Syngenta México;

4 DuPont-Pioneer México;

5 Bayer CropScience Mexico.

Environmental Risk Assessment (ERA) is a previous requirement to the environmental release of GM crops in many countries such a as Mexico. Here, there is an extensive legislation covering agbiotech applications including the Biosafety Law on GMO (124 articles), its By-law (73 arts.) with most of regulatory procedures, which contains the 'Special Regime for Maize Protection' and additionally, national standards related to biosafety. Procedures for the ERA of GM agricultural or forest varieties are being elaborated through one of these NOM (Norma Oficial Mexicana), which should, among other requirements, document most biosafety measures planned/ imposed in release applications/ permits. Furthermore, a multi-sectorial group also gathered to discuss and elaborate a technical guide for addressing potential risks to Non-Target Organisms (NTOs) from Bt (Resistant to Insect Pests) GM varieties in field conditions. The guidelines include criteria for: a) the inventory of entomofauna present in different eco-regions (level IV according to the North American Commission for Environmental Cooperation)1; b) the selection of representative or surrogate NTO species within functional groups; c) a tiered evaluation of potential adverse effects and d) some monitoring during advanced phases of evaluation. In Mexico, GM cotton is already at the commercial phase in extensive northern areas, but maize is still stuck at the pre-commercial phase in reduced areas, so different schemes have been adopted for each crop under the principles of Annex III of the Cartagena Protocol, the 'problem formulation' approach, and the experience on NTO work of selected authors and institutions around the globe. Advances on this implementation process will be briefly presented.

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Bio-business: Identifying and Creating Market Opportunities.

Alfredo Godinez Yerena

CIETT, UANL, Monterrey, Nuevo León, México

For the generation of knowledge-based economy, there are different tools for assessing entrepreneurial projects which also define the feasibility of bringing them to market. Michael Porter, supports this assessment by core generation business, which is known as a business model; the development of a business model makes it a practical way to evaluate who are our customers, competition, technological advantage, and suppliers, thus defining the value proposition (the project) with a viability of going to market or the disadvantage of making adjustments to our enterprise project.

To make an assessment of the viability of our business, the behavior of the final consumer has to be considered; for projects in science and technology, the same factors are taken into account. Maslow, through his study of the pyramid of needs, contributes to achieving a focus on what kind of market opportunity is presented to our business, are we in the right market? This question is easy to answer by finding the part of the pyramid where we stand on, and where we want to go; it is easy to identify which errors are committed when heading to a market that is not made for our business. Other tools to identify the feasibility of my project is through the business models "tech push and market pull", which provide us guidance of what proposal we want to turn into business, first our proposal and the scope it has, and on the other hand, to gives us a higher success rate, what the market wants and suggested answers to that need.

It is not only the idea generation, but also it is important to consider the environment or encourage it, to have a higher margin of success, identify strengths and diminish our weaknesses, to find the best strategic schemes for the output of products and marketing, ensuring the providers' portfolio and focus on technology watch. All this will help us our business to have a sense of permanence in a market that today is constantly evolving, and demands more practical developments, when one enters that market.

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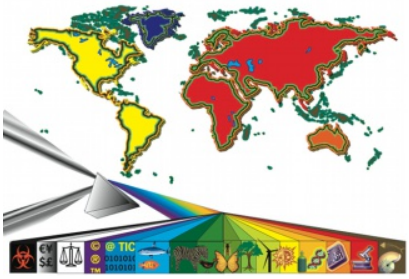

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Rogelio Rodríguez-Sotres

Faculty of Chemistry

Universidad Nacional Autónoma de México

Tel (+52)5556225285

Fax (+52)5556225329

e-mail: sotres@unam.mx

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