Institution: University of Sussex

Unit of Assessment: 10 – Mathematical Sciences

Title of case study: Combating crop losses and improving global food supplies through mathematical modelling of "gene silencing"

Period when the underpinning research was undertaken: 2013 – 2019

Details of staff conducting the underpinning research from the submitting unit:

Name(s):	Role(s) (e.g. job title):	Period(s) employed by submitting HEI:
Dr Konstantin Blyuss	Reader in Mathematics	1 October 2010 – present
Dr Yuliya Kyrychko	Reader in Mathematics	1 October 2010 – present

Period when the claimed impact occurred: 2018 – 2020

Is this case study continued from a case study submitted in 2014? N

1. Summary of the impact

Insights from mathematical models developed at Sussex have been implemented in Ukraine (the world's largest producer of sunflower seeds and in the top six producers of potatoes, wheat, barley and corn globally) with the following impact:

- Improvement of farming practices in the Prydniprovsky region of Ukraine, resulting in increased crop viability by 7-12% and reduction in pest control costs by 15-20%. The region contains almost 20% of the total Ukrainian sown area of agricultural crops and occupies a third of the country's steppe.
- Development by Ukrainian biotech companies of natural biostimulants which have been used throughout Ukraine, increasing yields of wheat (by 11-21%), barley (9-18%) and sunflowers (19-63%), tomatoes (20-66%), potatoes (10-12%) and cucumbers (23-26%). These biostimulants reduce the need for environmentally-harmful pesticides, and are proving to be a successful viable alternative for organic farming.

2. Underpinning research

With the rapidly growing world population and major changes in the global climate, an important factor in sustaining food productivity and security is the ability to effectively and environmentally-safely protect agricultural crops. Their performance can be hindered by many factors, including significant risk from pathogens and parasites that annually cause losses estimated to be in the order of hundreds of billions of dollars. For many years, this problem was addressed using chemical pesticides, but the emergence of pathogen resistance and the potential harm to humans have necessitated the development of alternative approaches.

Having previously published several highly-cited papers on mathematical modelling of the effects of awareness on control of directly transmitted infections, in 2017, Blyuss collaborated with colleagues from Visva-Bharati University in India on modelling vector-borne infections of crop plants and their control [R1]. Their paper proposed a mathematical model for control of mosaic disease through application of nutrients (fertilisers) and insecticides. Analysis of the conditions for disease persistence produced quantitative results for the amounts of nutrients and insecticides that need to be used in order to achieve disease eradication, depending on various factors, such as the rate of disease transmission from vectors to plants, vector mortality, as well as the carrying capacity of crop plantations, and farmer awareness based on observing the proportion of infected plants.

Subsequently, in collaboration with colleagues from India and Ukraine, Blyuss conducted another study that mathematically modelled the effects of applying microbial biostimulants for the purpose of improving crop performance and protecting crops against parasites [R2]. That work identified quantitative relations between the amount of biostimulants required for disease eradication and various features of disease transmission, as well as parameters characterising the performance of

these biostimulants, such as their effect on inducing reduction in disease transmission from vectors to plants, and the increase in vector mortality. Although these models used an example of plant mosaic disease, the modelling framework and results are applicable to other plant pests and parasites.

One very promising approach for safe and effective control of crop parasites currently pursued by some of the world's largest agrochemical companies is that of RNA interference (RNAi), also known as “gene silencing”. RNAi is a fundamental biological process (discovery of this was recognized with a Nobel Prize in 2006), by which eukaryotic cells can control expression of specific genes after they are already transcribed. In the specific context of protecting plants from infection, RNAi is used in two different ways. Providing plants, by means of spraying or root soaking, with appropriate double-stranded RNA (dsRNA) can help them stop expressing genes that are necessary for a successful infection. On the other hand, RNAi can also be used to target parasites, where parasites feed on plants and ingest dsRNA that then triggers the process of RNAi against parasites’ own genes, thus reducing their fecundity and causing mortality.

The progress in developing RNAi-based tools for pest control, particularly microbial bioregulators, has been hampered by the lack of detailed quantitative description of the dynamics of RNAi in plants and parasites. To address this problem, Blyuss, Kyrychko and Neofytou at Sussex developed the first ever mathematical models of RNAi in plants [R3-R5] that provided quantitative insights into the mechanisms of how plants respond to infections by means of RNAi. These models showed how the optimal rate at which plant cells are able to express necessary dsRNA constructs, protecting them from infection and/or parasitism, depends on other fundamental parameters, such as plant cell maturation rate, the rate of spread of infection within the plant, as well as how RNAi in the cells of parasites can be amplified by using other types of small interfering RNAs (siRNAs). These results have provided quantitative predictions of the influence of different parameters on the effectiveness of RNAi in silencing plant genes involved in infestation, as well as the essential nematode genes.

Subsequently, Blyuss established contacts with colleagues at three research institutes of the National Academy of Sciences of Ukraine, and together they have used theoretical findings from these models to develop natural microbial biostimulants produced from soil bacteria that are capable of protecting crops against parasitic nematodes using the mechanism of RNAi. The results of this research have been published in Frontiers in Plant Science [R6], the world’s most cited peer-reviewed journal in the field of plant science, where it was experimentally shown how natural microbial biostimulants are able to reduce the levels of nematode infestation in wheat, while targeting essential nematode genes. By January 2021, this paper was viewed over 8100 times by readers from 38 countries and all continents. It has also attracted significant attention on Twitter and in the press.

3. References to the research

R1. F. Al Basir, **K.B. Blyuss**, S. Ray, Modelling the effects of awareness-based interventions to control the mosaic disease of *Jatropha curcas*, 2018, Ecological Complexity, vol. 36, pp. 92-100. doi: 10.1016/j.ecocom.2018.07.004

R2. **K.B. Blyuss**, F. Al Basir, V.A. Tsygankova, L.O. Biliavska, G.O. Iutynska, S.N. Kyrychko, S.V. Dziuba, O.I. Tsyliuryk, O.O. Izhboldin, Control of mosaic disease using microbial biostimulants: insights from mathematical modelling, 2020, Ricerche di Matematica, vol. 69, pp. 437-455. doi: 10.1007/s11587-020-00508-6

R3. G. Neofytou, **Y.N. Kyrychko**, **K.B. Blyuss**, Mathematical model of plant-virus interactions mediated by RNA interference, 2016, Journal of Theoretical Biology, vol. 403, pp. 129-142. doi: 10.1016/j.jtbi.2016.05.018

R4. G. Neofytou, **Y.N. Kyrychko**, **K.B. Blyuss**, Time-delayed model of immune response in plants, 2016, Journal of Theoretical Biology, vol. 389, pp. 28-39. doi: 10.1016/j.jtbi.2015.10.020

R5. G. Neofytou, **Y.N. Kyrychko**, **K.B. Blyuss**, Time-delayed model of RNA interference, 2017, Ecological Complexity, vol. 30, pp. 11-25. doi: 10.1016/j.ecocom.2016.12.003

R6. **K.B. Blyuss**, F. Fatehi, V.A. Tsygankova, L.O. Biliavska, G.O. Iutynska, A.I. Yemets, Y.B. Blume, RNAi-based biocontrol of wheat nematodes using natural poly-component biostimulants, 2019, Frontiers in Plant Science, vol. 10, 483. doi: 10.3389/fpls.2019.00483

4. Details of the impact

This Impact Case Study focuses on Ukraine, which, according to the latest data from the UN Food and Agriculture Organisation (Statistics Division), is the world's largest producer of sunflower seeds, and in the top six producers of potatoes, wheat, barley and corn globally. In spring 2019, Ukraine overtook China to become the third largest exporter of agricultural products to the EU after Brazil and the USA. Translation of research results [R1-6] has resulted in a better-informed practices around using fertilisers and pesticides to protect crops against pests, and in the development of natural, environmentally-safe crop biostimulants. This has led to the increase in crop yields by 7-12% with an associated reduction in expenses for crop protection of 15-20% in the Prydniprovsky region of Ukraine, which contains 20% of the total Ukrainian sown area and produces 8% of the world's sunflower. Natural microbial biostimulants developed through translation of theoretical results have already proved effective in increasing crop yields in major cereals, such as wheat, barley and sunflowers by 11-21%, 9-18% and 19-63%, respectively, while for vegetables, the yields for tomatoes, potatoes and cucumbers have increased across the country by 20-66%, 10-12%, and 23-26%.

Changing farming practices to improve crop viability and reduce pest control costs

In 2017, Blyuss visited Ukraine and signed a Memorandum of Understanding between the University of Sussex and the Prydniprovsky Scientific Centre of the National Academy of Sciences of Ukraine (PSC). In collaboration with colleagues from the Dnipro State Agrarian and Economic University and the PSC, during 2017-2018 Blyuss translated theoretical results from R1-3 into practical recommendations for farmers on optimal strategies of using nutrients, pesticides and microbial biostimulants for protecting crops against pests, based on monitoring the levels of infection at crop plantations. These practical recommendations, focused on crop pests native to the steppe region, were subsequently distributed to farming companies and agribusinesses in the Prydniprovsky region of Ukraine, with 40% of farmers in the region adopting these recommendations into their farming practices. In terms of reach, the Prydniprovsky region contains almost 20% of the total Ukrainian sown area of agricultural crops and occupies a third of the country's steppe. It has an abundance of fertile chernozem soil, and, according to latest data from the Ukrainian National Statistics Service, produces 20.2% of the total Ukrainian harvest of wheat, 21.8% of barley, 13.3% of vegetables, and 26.3% of sunflower [S1, link 3]. In fact, in 2019 that region alone produced 8% of the total amount of sunflower produced worldwide.

Professor Anatolii Bulat, President of the Prydniprovsky Scientific Centre, has noted:

> "Currently, monitoring and control of the processes of growth and development of agricultural crops on farming lands of various agribusinesses in the Prydniprovsky region are performed in accordance with the results of scientific research and technical recommendations developed by Dr K.B. Blyuss…. Through close collaboration with farmers and agribusinesses, … currently around 40% of farmers in Prydniprovsky region are already using above-mentioned technical recommendations. This has allowed farmers and agribusinesses to increase the survivability of crops by 7-12%, to improve quality and yields of cereal crops, and to reduce expenses associated with control of crop pests by 15-20%, compared to 2015-2017 when these techniques were not used. Reductions in expenses were associated with reduced fuel and personnel costs, as well as a substantial reduction in the amount of nutrients, fertiliser and pesticides being used. The latter factor is a particularly important achievement of the newly developed technical recommendations from the perspective of reduction and minimisation of the negative impact of pesticides on environment and biodiversity." [S1].

With 18% of the working population of Ukraine being involved in agriculture, new technical recommendations on monitoring of crop infestation and the use of pesticides have had a direct impact on performance of local agribusiness in the Prydniprovsky region.

Mr N. Nodzrin, Chief Agronomist of the company "Pisarevsky Oleksandr Stepanovych" based in Vodyane, has said: "As a result of applying practical recommendations regarding monitoring the levels of crop infestation with pests, as well as optimal use of fertilizers and biostimulants, that have been developed by Dr K.B. Blyuss together with colleagues, our expenses associated with control of crop pests have reduced by 12,5-18,0%, while the viability of wheat and barley plants

has increased by 7,5-10%." [S2].

Similarly, Mr Viktor Lisnyi, the Director of "Lisnyi Viktor Mykolayovych" company, specializing in producing cereal grains, sunflower and corn on its 11ha farm in Promin', has noted:

> "This year we have used a new approach to monitoring of plant growth and protection against pests using pesticides and fertilizers, developed by Dr K.B. Blyuss together with colleagues ... This has allowed us to not only increase the viability of wheat, barley, oat and rapeseed plants by 8-10% on average, but it has also reduced our costs for control of pests of these plants by 15-18%." [S3].

Enabling the development of successful organic biostimulant products for improved crop protection and yields

In R3-5, Blyuss and Kyrychko proposed and analysed mathematical models of RNA interference, explaining quantitatively how this process develops in plants, and how it can be used to target plant parasites. Results of these models helped their colleagues in the Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine select metabolites of soil bacteria that result in the highest levels of within-plant production of siRNAs protecting plants against parasites, and it was these metabolites that formed the basis of natural microbial biostimulants. After testing in laboratory, greenhouse and field conditions, in 2018 Ukrainian agro-investment and agrochemical companies "Bioinvest-agro" (http://www.bioinvest.com.ua) and "Imptorgservice" (imptorgservis.uaprom.net) produced these biostimulants and sold them to farmers and over 100 major agribusinesses around Ukraine and in Kazakhstan. The latter of these companies has annual sales of $500.000 - $1million. Besides direct economic impact in terms of profits to these agrochemical companies, the biostimulants have also resulted in a substantial improvement of crop performance for individual farmers and agribusinesses using them. More specifically, across different regions of Ukraine, these biostimulants have increased yields of winter wheat, barley and sunflowers by 11-21%, 9-18% and 19-63%, respectively, while for vegetables, the yields for tomatoes, potatoes and cucumbers have increased across the country by 20-66%, 10-12%, and 23-26% due to using natural biostimulants [S4, S5].

By virtue of being produced from natural products of soil bacteria, natural microbial biostimulants can be used for organic farming. This is extremely important, since organic farmers suffer substantial losses due to inability to protect their crops against pests using conventional pesticides. In this respect, these natural microbial biostimulants are providing a viable alternative, which allows to protect crops against parasites, while minimizing negative effects on the environment, and maintaining standards of organic farming. One of these biostimulants – "Phytovit", produced by "Bioinvest-agro" [see R6] – has been approved for organic farming and even received international "Organic standard" certification recognized by the EU and Switzerland [S4] and is currently used by some of the largest organic farming companies in Ukraine.

Results of theoretical and experimental work on natural biostimulants have been published in a joint research publication [R6] in Frontiers in Plant Science, the world's most cited journal in the field of plant science. Due to major significance of this work in terms of providing effective, and at the same time, environmentally safe, tools of pest control, this work has been covered by a large number of global media specializing in farming and agriculture. This includes articles in English, French, Italian, Spanish, Portuguese, Russian, and Turkish [S6].

The significance of this work has been recognised by an award to Blyuss and Kyrychko of Letters of Gratitude from the PSC, which is the highest honour that can be awarded (only once) to any scientist for their major scientific contribution to solving problems of significant practical importance. These awards were given to Blyuss and Kyrychko on the 17 May 2019 at the annual Day of Science ceremony, organised by the National Academy of Sciences of Ukraine [S7].

Natural microbial biostimulants are already widely used in Ukraine and Kazakhstan. The next phase of impact will concentrate on certification of other natural microbial biostimulants for the purpose of their use in organic farming. The largest Ukrainian producer of organic commodities, which is also the second largest organic grower in Europe and the biggest exporter of organic soybean and corn from Ukraine to the EU, is currently testing these biostimulants for the purpose of adopting them as main tools of pest control in its agricultural lands.

5. Sources to corroborate the impact

S1. A scanned copy of the signed and stamped letter from the Prydniprovsky Scientific Centre of the National Academy of Sciences of Ukraine, confirming the adoption of technical recommendations, as developed by Dr Blyuss, by farmers in Prydniprovsky region (clearly stating the level of uptake, and reach of impact), together with quantitative data on the improved crop performance. Also, National Statistics Service of Ukraine (summary of agricultural production)**:**

- http://ukrstat.gov.ua/operativ/menu/menu_e/cg.htm
- http://ukrstat.gov.ua/operativ/operativ2006/sg/sg_rik/sg_e/rosl_1991-2019_e.html
- http://www.ukrstat.gov.ua/druk/publicat/kat_u/2020/zb/04/zb_rosl_2019.pdf

S2. Signed and stamped letter from “Pisarevsky Oleksandr Stepanovych”, an agribusiness in Prydniprovsky region, confirming the use of technical recommendation developed by Dr Blyuss, as well as data on their benefits on this particular farm.

S3. Signed and stamped letter from “Lisnyi Viktor Mykolayovych”, an agribusiness in Prydniprovsky region, confirming the use of technical recommendation developed by Dr Blyuss, as well as data on their benefits on this particular farm, as well as data on their benefits on this particular farm.

S4. Signed and stamped letter from “Bioinvest-agro”, a biotech company in Ukraine producing and selling microbial biostimulants, confirming company’s own economic benefits in terms of profits, as well as an impact for farmers in terms of improved productivity and reduced crop losses due to pests. Also, see organicstandard.ua list of 2020 Organic Standard Certified Products for “Phytovit” organic certification (no. 2.3.2.48 on p. 97)

S5. Signed and stamped letter from “Imptorgservice”, a Ukrainian agrochemical company, producing and selling microbial biostimulants confirming company’s own economic benefits in terms of profits, as well as an impact for farmers in terms of improved productivity and reduced crop losses due to pests.

S6. A PDF with a selection of media stories covering this work.

S7. The Letters of Gratitude from the Prydniprovsky Scientific Centre of the National Academy of Sciences of Ukraine (NASU). Information about the Day of Science ceremony on the official governmental website of the NASU, which specifically mentions Dr Blyuss’ and Dr Kyrychko’s awards can be found here (PDF archive of webpage and translation are supplied, as webpage is currently being moved to a new server): http://www1.nas.gov.ua/rsc/psc/chronicle/Pages/nov_22_05_19.aspx