Europe's project to control sharka

SharCo project leader **Dr Véronique Decroocq** explains the work and successes of the programme seeking to establish pioneering methods to battle sharka disease in stone fruit trees

Photo courtesy of S. Decroocq

The SharCo project is founded on three pillars; can you outline what each of them are, and how they relate to the overall aims and objectives of the consortium?

SharCo is constructed on three pillars addressing respectively the epidemiology of the disease, breeding and other efforts to develop genetic resistance to the disease and practical application and dissemination efforts to provide information and tools to stakeholders. The first pillar targets the virus itself, aiming to understand its spreading behaviour, its diversity and the physical control measures that could be used to limit its spread in nurseries and in orchards. The second pillar focuses on the host plants and how we can modify and reduce the interaction of the virus with its perennial hosts in order to provide durable resistance in the field and in nurseries. The third pillar is application, and deals specifically with the behaviour of stakeholders and producers and aims to disseminate best practice elements in stone fruit tree production, both in nurseries and orchards.

Part of the remit of the project involves the development of novel and high throughput detection systems which will warn of sharka outbreaks. Can you describe how such systems might operate or function in practice?

Different technologies are currently being developed and tested in SharCo in order to detect rapidly and accurately any PPV isolate and new viral outbreak. All strategies are based on an exhaustive evaluation of the viral diversity worldwide, which will provide a reference against which to compare any newly identified isolates in Europe and elsewhere, and will provide the extensive sequence information needed to develop new strain/isolate characterisation

DR VERONIQUE DECROOCQ, PROJECT LEADER

techniques. The first such typing tool being developed is based on microarrays produced with oligonucleotides covering the major parts of the viral genome. The second one is based on the selective amplification of viral DNA followed by multiplexed analysis of single nucleotide polymorphisms (SNPs). It is important that they are applicable everywhere in Europe, in any laboratory and at low cost, the goal being to provide the EU plant protection and quarantine services with tools that are both reliable and cost-effective.

How important do you consider the development of PPV outbreak prediction mechanisms through the mapping of PPV diversity and characterisation of epidemic properties? What do you hope to achieve through this?

For practical reasons, understanding the epidemiological behaviour of the sharka virus in orchards is a formidable task. However, such information is needed if we are to develop reliable models that will allow us to predict accurately the development of epidemies. We believe that bringing together information about the virus diversity and about its spatial distribution and spread will allow us to make major advances in our understanding of these epidemies and will provide us for the first time with reliable estimation of crucial parameters that are so badly needed in modelling efforts. In turn, we are confident that the availability of reliable models will provide us and, ultimately all stakeholders, with better predictive tools that should greatly improve the efficiency of eradication and management measures.

By what means does the programme evaluate and measure its success?

SharCo is a problem orientated project. Ultimately, the success of SharCo will therefore be evaluated by the adoption of the new or improved tools and strategies by nurserymen, growers, by all parties concerned by eradication or containment efforts and, of course by an improvement of our overall ability to control this disease. Participation of end-users in SharCo training workshops and the SharCo research workshop that will be held in Sofia in September 2010 are the means for us to evaluate the success of the knowledge transfer process that is a key element in the adoption of SharCo results, and of their gradual introduction in practice. Of course, there will be many more practical indicators of progress, and the development of the first integrated database on PPV diversity or the release in France and Spain of the first resistant apricot varieties adapted to the European market, will represent particularly visible milestones.

Plum Pox Battle

The Sharka disease ravages fruit production the world over, causing widespread socioeconomic and environmental damage. Now, the **SharCo** project, headed by **Dr Véronique Decroocq**, is finding innovative ways to detect and combat the disease

Photo courtesy of S. Decroocq

SHARKA IS THE most devastating disease that affects stone fruit trees worldwide; indeed, the extent of damage caused in Europe alone over the last 30 years is estimated to be 10 billion euros. The disease is caused by the Plum pox virus (PPV) in stone fruit tree species of the sub-family Prunoideae, such as peach, apricot, plum, cherry and almond trees. The infection alters fruit development and induces fruits to drop before ripening. In most cases, fruits affected by PPV cannot be sold and up to now, the only available solution to reduce the spread of the virus has been to eradicate infected trees.

The SharCo project has been established to confront this blight from European nurseries and orchards by seeking innovative solutions to the problem. Dr Véronique Decroocq is the coordinator of the programme, and a specialist in plant molecular biology and genetics. She stresses the dangers of the virus for European agriculture, especially in more deprived regions: "The disease is devastating for stone fruit producers and nursery workers in Eastern and Southern European countries, jeopardising severely the sustainability of rural regions already impoverished".

THE PROJECT AND SHARKA DETECTION

SharCo is a transnational project funded by the EU's Seventh Framework Programme. Not limited to a knowledge-based project, SharCo intends to be an application driven initiative, linking researchers and end users. As Decroocq is keen to emphasise, the project relies on a range of producers and scientists to meet the aims of producing an industry and an environment better equipped to deal with the disease: "Sustainable production and consumption of healthy, fresh fruits depend on our capability to reduce those impacts and to provide sustainable and realistic measures that will be, by the end, validated and applied by stakeholders, nurserymen and fruit growers," she explains. The project attempts to encompass all stakeholders and collaborates with 16 SharCo partners across 11 countries, while covering the entire chain from planting material to orchard management. The scope of outcomes from the project is hoped to be equally wide, including resistant varieties, management and cultivation guidelines and optimised survey and detection methods.

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The challenge for the SharCo team is to find mid-term and long-term control strategies. Current containment methods failed to prevent the disease spreading in Europe and its introduction to new territories such as the U.S., Canada and more recently, Japan. To date, one of the problems has been the inefficacy of routine detection methods, as Decroocq explains: "Up to now, the disease is mostly detected visually, meaning that it can only be noticed several months to several years after the true infection event". By that time, the disease could have been spread further by aphids, or multiplication practices such as grafting. Decroocq knows that early detection is an important requirement to reduce work further down the line: "The earlier the infection is detected, with an accurate and sensitive tool, the better the efficiency of eradication or containment practices, with a lower impact of containment measures on the producers, since fewer trees will need to be removed". Once established, to be able to slow down the spread of the virus, its driving factors must be identified, and so studying the epidemiology of the disease is essential.

NEW RESISTANCE

Currently, once a plant is infected, there is no possible cure, and nor does there seem to be much chance of one being developed in the near future. Instead, SharCo is concentrating on combating the virus by manipulating host plant factors. New genetic tools are being developed for speeding up the selection of PPV resistant varieties. Recent studies by the team demonstrated that

the weed, Arabidopsis thaliana, is an alternative and promising model of study of the plant/PPV interactions. Indeed, Arabidopsis plants in which the host proteins, eIF(iso)4E or eIF(iso)4G, are deleted, proved particularly resistant to PPV. Decroocq sees this as very important, as it points to which host proteins the virus needs: "This data is very important since it shows that the virus relies critically on these host proteins to complete its infection cycle," she explains. "If the virus is no longer able to 'find' one or the other of those indispensable factors, it cannot mount a productive infection, and the plants are in effect resistant to infection." The implication is that SharCo will be able to manipulate host plants and turn down the expression of such genes using this strategy to provide PPV resistant trees for growers.

Currently, SharCo is engaged in screening the natural diversity of Prunus host species. By analysing trees from the centres of origin of the stone fruit trees (China and Caucase) the equivalent genes are surveyed for mutation, providing resistance to the virus. The knowledge acquired from Arabidopsis has been a great asset for SharCo's methods, as Decroocq details: "Having the information on the identity of the eIF(iso)4E or eIF(iso)4G factors is critical for these efforts, as it allows us to use high throughput targeted approaches rather than go through lengthy classical phenotyping assays". SharCo is also testing various RNAi strategies to combat PPV, although this is only to evaluate the feasibility and efficiency of these approaches, and to use them to gain a better understanding of the molecular interactions between virus and host.

BRANCHING OUT

Dissemination of the project's work is a vital last step and the SharCo consortium is dedicated to make an impact in this regard. Through the network of partners, workshops offer endusers training, as Decroocq describes: "SharCo partners have started to develop a tight collaboration with National Plant Protection Organizations (NPPOs), extension services and nursery / fruit producers, through the organisation of training workshops in the national languages of the local stakeholders". Training workshops have been held in Poland and Turkey, and a discussion and feedback forum has been established and is to be continued through the development of cultivation guidelines. Despite such encouraging signs, the project has not been without its challenges, not least in dealing with the international community, as Decroocq illustrates: "Some countries, outside of the EU, do not acknowledge the presence of the virus in their area and thus, even if PPV was detected in material originating from those regions, we cannot bypass national laws and publish the data". Furthermore, planting GMO crops in fields is prohibited in many EU countries, restricting the opportunities to check the validity of RNA interference techniques.

The virus itself offers up one major obstacle – very high evolutionary potential, and a diversity that leaves SharCo with potential complications. Decroocq believes SharCo can have the right measures in place to overcome this: "Diagnostic techniques need to be robust enough to allow detection of all isolates, typing techniques have to be developed, and efforts are needed to evaluate to what extent epidemiological properties are general," she asserts. Large and cost-effective experimental trials in several European countries presenting a constrasting environment, from neighbouring industrial orchards in western nations to home gardens in the mid-eastern countries, will also help tackle PPV's diversity in the future. Meanwhile, training workshops are planned throughout 2010 in Czech Republic, Bulgaria and Romania to continue the dissemination of the expertise of SharCo and to bring about a sustainable solution to the sharka problem. If, as SharCo hope, early warning measures are in place, PPV susceptible plants are replaced by resistant cultivars and new cultivation techniques are implemented, then the incidence of sharka can be significantly reduced, if not eliminated, in many areas.

SYMPTOMS OF SHARKA DISEASE ON APRICOT FRUITS (PICTURE COURTESY OF D. BOSCIA, SHARCO CONSORTIUM)

SharCo

SHARKA CONTAINMENT IN VIEW OF EU-EXPANSION

OBJECTIVES

SharCo seeks to develop knowledge of PPV epidemiology and stone fruit genetics with the aim to respond to the requirements of end-users, such as fruit producers and authorities.

The project will propose new PPV resistant varieties, a code of best practices to reduce contamination and spread of the disease in nurseries and orchards, and will develop a decision support system, which will enable producers to confront outbreaks in the most appropriate and efficient manner. Furthermore, the project will submit guidelines for stone fruit cultivation and trade with policy makers and plant protection services as particular stakeholders.

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