



THE ANNUAL 2022

Journal for breeders and producers of plant material

Prophyta

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+31 (0)71 332 6262
info@naktuinbouw.com
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EDITORIAL ADVISORY BOARD

Theo Ruys, John van Ruiten, Sally van der Horst, Erik Toussaint and Marien Valstar

PUBLISHER/CHIEF EDITOR

Monique Krinkels

CONTRIBUTING EDITORS

Jeroen Balemans, Peter van Dijk, Daniël Ende, Fernando García-Bastidas, Sally van der Horst, Gert Kema, Tijs Kierkels, Smadar Kleiman-Shoval, Bénédicte Lebas, Hubert Lybeert, John van Ruiten, Anker Sørensen, Rose Souza Richards, Roland Willmann and Joyce Woudenberg

COVER PHOTO

Kitty Vijverberg

SUB EDITOR

Mireille McNutt

GRAPHIC DESIGN

Marcel Bakker, De Ontwerperij

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GENERAL INFORMATION

www.prophyta.org

ADDRESS

Blue Bird Publishers

VOF, Jan Kostelijklaan 16

1981 CG Velsen-Zuid

the Netherlands

phone +31 (0)255 521 852

prophyta@prophyta.org

www.prophyta.org

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On the cover: Embryo sac of a non-apomictic dandelion, showing an egg cell that awaits fertilisation. In apomictic dandelions, the egg cell omits this awaiting and starts cell division spontaneously

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In Short



Four new Gold Medal winners

LAST JANUARY, FLEUROSELECT presented its 2023 Gold Medal winners. Each one of these four breeding breakthroughs obtained an outstanding score for innovation, beauty and garden performance.

Helenium autumnale ‘HayDay Golden Bicolor’ is bred by Syngenta Flowers. It is the first *Helenium autumnale* from cuttings that is genetically suited for pot production. The dense branching results in loads of flowers that attract pollinators and wildlife. The brightly coloured golden-yellow blooms, with red undersides, flower from summer through autumn until the first frost. Being naturally compact, HayDay Golden Bicolor is not only suited for larger beds and landscaping, but also for patio containers and smaller borders.

Helianthus annuus ‘Lemon Cutie’ is bred by SeedSense. This *Helianthus annuus* is not only a dwarf variety with multiple branches, but it also has stunning and unusual semi-double, lemon-coloured flowers. Lemon Cutie is perfect for urban areas, such as small patches along footpaths, on balconies or in containers and also looks wonderful in larger beds, or even in landscaping. It has excellent germi-

nation and can be offered in seed packets, sold as plugs or grown in pots.

Rudbeckia fulgida var. *sullivantii* ‘Goldblitz’ by PanAmerican Seed is a newcomer in the Kieft Brand. It is a first-year-flowering solution for growers that are unable to reliably finish *Rudbeckia fulgida* Goldsturm under their natural growing conditions. Goldblitz plugs do not require special treatment during the production process, while delivering the same quality as the traditional Goldsturm. This brand-new *Rudbeckia fulgida* flowers approximately 14-21 days earlier, is more uniformly grown in its first year and has an excellent garden performance.

Verbena bonariensis ‘Vanity’ by Van Hemert & Co is clearly compact than existing varieties thanks to its shorter stems. The tight flower clusters have a more intense purple-blue colour and bloom from mid-summer until autumn frost. Vanity offers a long flowering season, attracts bees and butterflies and spreads a subtle fragrance in the early morning and evening.

WUR number one university

ACCORDING TO THE QS WORLD UNIVERSITY RANKINGS by Subject, Wageningen University & Research is the best agricultural university in the world. The influential ranking was released on 8 April. It is the seventh time in a row that the Dutch WUR ended as number one. For this ranking 1,543 universities were evaluated on 51 different subjects. About half of the QS ranking consists of a reputation survey (academic and employer), and the remainder consists of an analysis of the university's scientific impact (publications, H-index, citations).

PBR training course this autumn

NAKTUINBOUW IS ORGANISING ITS Plant Breeders' Rights training course in October, instead of June. It starts is on 3 October 2022 and will be held entirely online. The programme focuses on the implementation of the Plant Breeders' Rights system, based on the UPOV principles. The participants all work in the field of Plant Breeders' Rights. For nine weeks, they share ideas and experiences and networks with colleagues from all over the world. They start each week with a joint online kick-off, after which an online master-class follows, depending on the subject.

In 2021, 41 students followed the training course, a group larger than ever. This indicates the importance of a reliable Plant Breeders' Rights system is felt in many countries.

More information: www.naktuinbouw.nl

Research institute aims to improve photosynthesis

WAGENINGEN UNIVERSITY & RESEARCH (WUR) and the Photosynthesis 2.0 Research Fund (P2RF) are to establish a new independent institute for research on photosynthesis. Key objective is to learn how the efficiency of photosynthesis in food crops can be improved. That could contribute significantly to feeding the world. This decision was announced during the university's 104th Dies Natalis. The provisional name of the institute is the Institute for Advanced Studies for Photosynthetic Efficiency (IASPE). A total of 62 million euros will be invested in the new institute over the next decade. Photosynthesis is the most important biological process in the world. The efficiency of photosynthesis in field conditions leaves much to be desired. Plants use an average of only 1% of all the solar energy that reaches the earth, while this could easily be five or six times more. "This can, and should, change," says Louise O. Fresco, President of the WUR Executive Board. "If plants could absorb twice the amount of light, this would already have a huge impact on food production. That would be fantastic news for the Global South, but even better: this may teach us how to utilise such chemical-physical processes to generate sustainable energy."

Several plant varieties have already adapted to rapid growth through natural selection and have achieved a much higher photosynthesis efficiency than comparable varieties. One such plant is grey mustard, a plant native to the Mediterranean. From these plants, researchers could learn how to use their genetic developments to induce a similar transformation in food crops. The institute plans to produce 30 PhD students and 60 postdocs in the coming years.

Crisis or catastrophe

According to Oxfam, the world will be facing an unparalleled disaster. At the end of 2022, at least 860 million people will have to live on less than 1.75 euros a day. At the same time, food prices have skyrocketed. This Spring, the costs of a daily meal in Western Europe reached a peak higher than the crisis of 2011 and a further rise in food prices is inevitable, according to economists. For some people, this will mean a harsh choice between 'eat or heat'.

In the Sub-Saharan region, where people already have to spend half of their income on food, the situation will become even more dramatic. The causes: the raw material prices, packaging prices, transport costs and energy costs, which in turn are caused by the covid-19 pandemic and, of course, the invasion of Ukraine by Russia.

And there is very little we can do to prevent a crisis. Of course, we can punish Russia with sanctions for its cruel aggression. The US, UK and EU have put in place unprecedented financial penalties on Russia, and hundreds of international companies have pulled out of the country. In the short term, it might be doubtful whether a rich lady missing the opportunity to buy a new Chanel handbag or an oligarch losing his 'toy' yacht will have the desired effect but, in the end, the sanctions will be pummeling Russia's economy.

At the same time, Western Europe pays a price too, as the wheat, maize and sunflower oil which Ukraine produces and the natural gas which Russia exports are severely obstructed. In addition, the enormous influx of refugees will disrupt local economies.

Ukraine has found an alternative to export their produce, now that the ship routes across the Black Sea have been blocked. A train brings the produce to the West. And luckily, so far, seeds can still be sold to farmers, both in Russia and in the Ukraine, although the shortage of spare machine parts and in the Ukraine also of diesel, will hamper their activities. These are uncertain times and there is no way of telling what the future has in store for us. The one thing we can do to prevent a catastrophe is to provide farmers with the best seeds we have.

Monique Krinkels



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A blue sky over golden wheat fields

Monique Krinkels

It might not seem much, compared to the abhorrent pictures that the media shows of the Ukraine, but the war is also having devastating effects on world food security. The Russian invasion is impeding the Ukrainian farmers from sowing a large part of their over 40 million hectares of agricultural land. For more than 5000 years, the country's grain used to feed people from Europe to China and from Sweden to Eastern Africa.



The flag of Ukraine symbolizes a blue sky over golden wheat fields for a reason

According to a rough estimate of the Ukrainian agricultural research agency, APK-Inform, the war will allow farmers to sow 35% less grain and oilseeds this spring, compared to last year. Only in regions that are far from the frontlines are farmers able to work the land. Until recently, it was expected that one-eighth of all wheat on the world market would come from Ukraine this year. Also, 17% of the maize, 18% of the barley, 19% of the rapeseed, and no less than half of all sunflowers would normally be grown in Ukrainian fields.

According to farmers, it is difficult to get seeds, crop protection products and diesel. And there is also a long wait for parts for agricultural machines, if they are even available. "We urge the private seed sector to support measures ensuring that farmers and food producers in Ukraine continue to be able to access quality seeds and farming inputs, today and tomorrow. In the short term, this is critical to the survival of the country's farming sector; and in the long term, to its recovery and sustainability," says ISF Secretary General, Michael Keller. "ISF therefore calls for continued seed supply, enabling green lanes for critical agricultural inputs, such as seed, to be flown into the country."

EU-wide strategy

During a debate in April, European Agriculture Commissioner, Janusz Wojciechowski, acknowledged that action is needed for an EU-wide protein strategy. Due to the war in Ukraine, it is seen as a necessity to increase the European production of protein crops. Europe is largely dependent on the import of protein-containing raw materials.

The European Union expects grain exports to increase as a result of the war in Ukraine. That is what the European Commission says in its first quarterly report since the outbreak of the war between two

of the world's largest grain exporters. The commission predicts that EU exports of soft wheat will reach 40 million tons next season, against the 33 million tons previously estimated. Assuming normal spring and summer weather developments, total grain production could reach 297.7 million tons in 2022/23. That means an additional 1.5% year-on-year, with an expected soft wheat production of 131.3 million tons. Net exports of EU grains will increase by 11.5 million tons to 41.4 million tons, almost 40% more.

The commission also predicts a sharp drop in EU maize imports for next season, from the 14 million tons expected now to 9 million tons. The EU is a net importer of maize for animal feed and Ukraine is normally one of the largest suppliers.

In peacetime, if the grain price on the world market skyrockets, it is because maybe 1% less is harvested. But now it is possibly 10% worldwide, unprecedented in recent history. It is leading to rising food prices, as grain, maize and sunflower oil are ingredients of many edible products. 🌾

Finally we can meet and greet again

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Monique Krinkels

It has been long overdue. In 2020, the congress became the first ISF virtual event due to the covid-19 pandemic, the virus which was still circulating everywhere last year. 2022, will not become a 'twenty twenty too' and delegates from all over the world will assemble in the Catalanian capital Barcelona. Julián Arnedo, chair of the National Organizing Committee, welcomes all seedsmen.

For the first time in its history, the ISF World Seed Congress will take place in Spain. The country has a firm base in the seed sector worldwide. The Spanish seed market amounts to 585 million euros, which places the country as 5th in Europe, and 13th worldwide. Julián Arnedo, chair of the National Organizing Committee, is President of the Spanish national association of plant breeders ANOVE (Asociación Nacional de Obtentores Vegetales). This association brings together 56 plant breeding and seed producing companies and three research centres. "We are working with great enthusiasm to make it an interesting and attractive congress for all ISF members and other stakeholders," an enthusiastic Julián Arnedo states.

Favourable conditions

Spain is one of the largest countries in Western Europe, with a land mass of 50.5 million hectares, 40% of which is suitable for agriculture and/or horticulture. "We have exceptional climate conditions in Spain," says Julián Arnedo. "Thanks to that, a wide variety of crops can be grown. In the middle and north, for example, we have cereals (mainly wheat and barley) and maize (Bt maize), but in the south of Spain we have an important sunflower production, fruits in the east (stone fruit and citrus), berries (strawberry and blueberry) in the south-west and vegetables in the south. In Almería, the cultivation under plastic greenhouses is noteworthy and around Murcia you can find outdoor cultivation of salads." The country is the world's largest producer of olives, and of the olive crop, approximately 92% is dedicated to oil extraction, while the remaining 8% is used for

table olives. On average, a litre of olive oil requires four to five kilos of olives and Spain produces over half of the global demand for olive oil. It is the world's second largest producer of almond, tangerine and persimmon, the third largest producer of oats and artichoke and the fourth largest producer of grapes (but the second producer of wine after Italy and before France) to focus on the most remarkable facts. Julián Arnedo: "The fruit and vegetable sector is characterized, above all, by its orientation to foreign markets."

Impact of breeding

The independent Spanish Institut Cerdà published a report in November last year in which it quantified the impact of plant breeding in Spain. According to the

Seed trade (2019)

	Import		Export	
	Quantity (in mt)	Value (in million €)	Quantity (in mt)	Value (in million €)
Field crops	372,983	231	203,263	167
Flower seed	367	4	-	-
Potato seed	69,628	43	34,024	39
Tree seed	3	-	-	-
Vegetable seed	3,840	285	1,885	82
Total	446,821	564	239,172	287

Source: ISF compilation based on official statistics and international seed trade reports

Note: Only seed exports with a value greater than us\$ 1 million have been reported; flower seed includes seed of herbaceous and non-herbaceous plants cultivated mainly for flowers; field crops seed includes seed of pulses, cereals, industrial crops and forages; and vegetable seed includes seed of all vegetable crops.

Spain is the world's second largest producer of almonds

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'The seed market in Spain is growing. Not so much in quantity, but in quality and price,' says Julián Arnedo

Cerdá report, plant breeding has contributed more than 16,000 million euros to the Spanish economy. In 2019 alone, the breeding sector injected almost 1,000 million euros into the national economy. In the last thirty years, it has generated more than 400,000 jobs, 16,000 each year. In addition, plant breeders have created 62 Research and Development and Innovation (R&D&I) centres in Spain.

Most members of ANOVE are international operating companies, with a Spanish branch. "We have partners who work in citrus and almonds. The latter

is a crop that has been experiencing very significant growth in recent years. At the moment, there are no member companies working in grapes and olives," explains Mr. Arnedo. He underscores that the seed market in Spain is growing. "Not so much in quantity, but in quality and price. More and more resistant varieties are required."

Growing organics

The expansion of the organic food sector in Spain has been outstanding at all levels, according to a report published by the sectoral economic information company, Alimarket, from primary production to the processing industry, passing through the point of sale and ending at the consumer. In four years' time, the market has managed to double in size. Spain has established itself as the leading European country in terms of organic farming area, with 2.2 million hectares, ahead of, for instance, France.

In 2019, the growth of organic food in Spain would have been around 15% and its sales would already have been on the verge of 2,500 million euro. The annual double-digit growth thus maintained seemed to be continuing, at least in the medium term, as the large manufacturers had definitely launched themselves into competition with the specialists, which

Agricultural output (2019)

	Million €
Cereals	3,474
Wheat and spelt	1,043
Rye and meslin	48
Barley	1,203
Oats and summer cereal mixture	125
Grain maize	708
Rice	244
Other cereals	102
Industrial crops	795
Oil seeds and oleaginous fruits	309
Protein crops	46
Raw tobacco	59
Sugar beet	99
Other industrial crops	283
Forage plants	1,747
Vegetables and horticultural products	9,667
Potatoes	711
Fruits	8,416
Wine	1,053
Olive oil	2,579
Other crops products	158
Total crop output	28,600

Source: Eurostat



The International Seed Federation and the South African National Seed Organization warmly invite you to the ISF World Seed Congress 2023 in Cape Town, South Africa, from 5–7 June. Enjoy the generosity of our people, with the “I am, because we are” spirit of ubuntu against the unforgettable backdrop of the South African landscape.

www.worldseedcongress.com



Organizers



Nowadays, over 90% of the sales at Ramiro Arnedo S.A. is their own material



II

was undoubtedly giving the category a new boost. There is a special category where organic food is booming: Spanish consumers want to buy organics for their babies. Despite the decline in birth rates, organic baby food showed an increase of 24.2% in value and 28.2% in volume.

“Most of our members produce organic seeds and production is currently growing. I could not tell you if it is because of the increase in the number of vegetarians, or because we are increasingly aware that a healthy diet, based on fruits and vegetables, is much better for our health. The important thing is a healthy and balanced diet.”

Comunidades autónomas

Allowing for the cultural differences in this large country, Spain has seventeen autonomous communities, such as Andalusia, Catalonia and Basque Country and two autonomous cities, Ceuta and Melilla. To prevent increasing separatism and instability, the 1978 constitution provided for far-reaching decentralization. Each autonomous community has its own government and parliament with major legislative and executive powers. These ‘comunidad autónoma’, each have their own culture, sometimes their own languages and - important for foreign visitors - their own cuisines.

Barcelona is, for example, famous for its ‘pa amb tomàquet’ (toasted Catalan country bread scraped with garlic and lightly coated with the flesh of fresh tomatoes), ‘escalivada’ (smoky grilled vegetables - red peppers and eggplant - served with olive oil) and ‘espinacs amb panses i pinyons’ (fresh green spinach sautéed in olive oil together with raisins and pine nuts) to name just a few highlights for visitors of local restaurants.

The diverse cuisines have, however, a minor influence on regional food production. “The different crops in different parts of Spain are due to differences in climate and soil conditions. The trends and the production are being standardized more and more every day.”

Family owned

In daily life, Julián Arnedo is president of the seed company Ramiro Arnedo S.A. in Calahorra (La Rioja).

“It was founded in the 1940s by my father, Ramiro Arnedo Eguizabal. Initially it was a village store, which sold agricultural products, including seeds. My father was a real dealer, his business was trading.”

Until then, the farmers in the valley of the river Ebro lacked quality seeds.

Mr. Ramiro Arnedo Eguizabal became a promoter of horticulture in the region. In 1962, the company introduced the system of forced vegetable crop growing through the use of agricultural plastic, advocating what is today a reality in forced crops in Spain, and in particular in the Spanish south-east. In 1963, he introduced the first hybrid in Spain. The improvements that this first hybrid cucumber accounted for in horticulture were totally ground-breaking. “In 1965, my father got in touch with Royal Sluis and he started selling Dutch seeds in Spain until 1992, with great success.”

In that year, the company started its own breeding programmes in vegetables. “Due to the constant mergers in seed companies, we realized that we were going to run out of supply. So we started our own research and development of varieties. By this time, I was already working in the company. Nowadays, over the 90% of our sales is our own material, and the capital still belongs to the family.” The company has several facilities, such as Tecnosem, for pelleting seeds, research stations in Almería and Murcia and a pathology laboratory in Calahorra.

Besides vegetable seeds for professional growers, the company also has a seed catalogue for hobby gardeners with vegetables, flowers, grasses and aromatics. “The hobby market is growing, but the hobby gardeners buy plants, so they are customers of the nurseries. The hobby market is elaborated in the company. Everything is sold under our brand name ‘Jardiver’. In case of the aromatics, we only sell them for the hobby market. Nowadays it is trendy to have your own ‘garden’ in the kitchen or on the window sill. In any case, it is a marginal business.” 🌱

Olive oil called **liquid gold** for good reason

John van Ruiten

12 Say 'olives' and you say 'Spain'. Olives and olive oil are regarded as being very good for human health. The fruits contain unsaturated fatty acids at a high content. Olives are also used for the production of squalene, used in cosmetic manufacturing and in the production of vaccines.

• **Olives are one of the most important** products of Spanish agriculture. With a total of 400,000 producers, 2.7 million hectare and an annual average production of 1.2 million tons of olives, the country takes care of more than 50% of the EU production and 40% of the global amount of olive oil. Over 90% of the production is used for oil. Around 8% are table olives to be eaten in salads, dishes or as snacks. Spain is by far the world market leader in olives and olive oil. And the importance of the crop in Spain is still growing.

For the fresh market there are green and black olives. Green olives are the immature olives. Black/purple is the colour of ripe olives. But be aware: green olives can be coloured to black by processing them. Normally after harvesting it takes six to ten months to process the olives. It is necessary to keep them a long time in brine, dry salt, salinized water to remove the bitter taste caused by oleuropein.

For olive oil, the EU recognizes 59 regions of origin that are allowed to bear their specific 'protected designation of origin' (PDO). Olive oil can be identified to be produced and processed in this region. Spain has 20 of those regions of origin. The PDO

system in olives is similar to the system used in wine production.

Roman times

The history of growing olives in Spain is dating back to Roman times. The Romans spread the cultivation of olive trees and groves (orchards) to the west. Olives and olive trees were not only important because of the fruits, but also the olive branch was an old symbol of fertility, peace and luck and wisdom. Well-known is also the use of olive branches to make olive wreaths as a crown; it was the prize for winners already at the ancient Olympic Games in Greek Olympia. The branches of wild olive trees (*Olea oleaster*) were cut by boys with a pair of golden scissors.

The use of olive oil ('liquid gold') however was already widely spread 3000 years ago by the Phoenicians (a people from the area of Syria/Lebanon), sailing to all parts of the Mediterranean and selling it there. Olive oil was not only eaten, but also used for ritual reasons, as perfume, as massage oil and as lamp oil.

Records of first cultivation of *Olea europaea* dates back to 6000 BC in the Near East region Palestine and

Squalene and squalane

A very special use of olives is for the production of squalane, the hydrogenated form of squalene. It is a very stable fatty compound, not subject to oxidation. Squalane is a common ingredient in skin care products such as skin creams, sunscreens, food applications and vaccines. Possibly squalene is also one of the most promising functional ingredients in a healthy Mediterranean diet.

Traditionally these compounds are produced from the livers of sharks. Every year millions of sharks are being caught and trade legally and illegally. For 1,000 kilo of squalene 2,500-3,000 sharks are needed. Since nearly two in five shark species are on the IUCN Red List this is regarded as very undesirable. Therefore, the last 20 years the production of squalene from plants is becoming important, taking now care of almost half of the world demand. Olive oil is one of the most important plant-based sources. Rice is also being used for squalene production. Studies in Korea in rice revealed the mechanism for the biosynthesis of squalene and



The covid-19 pandemic has cost the lives of thousands of sharks as their liver oil is an ingredient of the vaccines

the fundament is created for breeding rice with higher squalene content. Data for squalene content in olive varieties are limited and the reason for differences measured must be further analysed.

Ir. J.E.M. van Ruiten is director of Naktuinbouw, Roelofarendsveen, the Netherlands, j.v.ruiten@naktuinbouw.nl



The ancient Greeks believed that the goddess Athena had created the olive tree. Hippocrates called it 'the great healer', Homer named it 'liquid gold', while Greek/Roman physician Galen praised it for its positive effects on health

Jordan. From there cultivation spread to Crete, Turkey and Greece and later on to Italy and Spain (around 100 BC). The Spanish word for olive, *aceituna*, has an Arab origin ('*zaytunah*').

Evergreen trees

Botanically the genus *Olea* consists of around forty evergreen tree species that naturally occur in Mediterranean Europe, South Asia and Africa. Introduction in the Americas by the Spanish took place since 1560 in Peru and a little later in California. Olive varieties all are from the species *Olea europaea*, but some of the other species are used for rootstocks (mainly *Olea oleaster*, *Olea hupidata* and *Olea sylvestris*). Many of the varieties can grow on their own roots, but the use of rootstocks can help if varieties are not rooting easily or in specific conditions of related to soil disease resistance. The rootstocks are also used to obtain some dwarfing of the trees. Rootstock varieties used are among others *Arbosana*, *Corbella* and *Limoncillo* for the creation of high-density orchards. Olive trees are either self-pollinating or (if the cultivar is self-incompatible) planted together with another variety/selection to fertilize each other. They are then wind-pollinated. After planting it takes five to ten years before the trees start flowering and producing olives. But once they are mature an orchard continuous to grow olives for more than a century before the production of the trees decreases. On average the production of one tree is around 25-30 kilogram per year. And it takes 5-8 kilogram to press one litre of olive oil out of it.

Varieties

Worldwide over 1,500 varieties (and a huge number of selections) are recognized and identified, nowadays also with their DNA profile. Most cultivars are old clonal selections and were locally selected over more than 5500 years in the various growing regions.

Typically, these clones were grown in the region of selection did not spread widely. In Spain more than 250 varieties are known of which about 25 are still grown at an economic scale. Most important varieties are 'Arbequina' (in Catalonia), 'Cornicabra' (in central Spain), 'Hojiblanca' (in Andalucia), 'Picual' (province Jaen) and 'Picudo' (in Granada).

Breeding olive varieties with cross breeding programmes was initiated in the 1960's. Most 'early' breeding programmes were done in Israel and Italy. In the 1980's in many other countries active breeding has started. Important breeding goals are the shortening of the non-productive juvenile period of trees (which used to be over 15 years), disease resistance and quality aspects. Adaptability of new cultivars to new high density planting systems designed for mechanical harvesting is also an important breeding goal.

The number of varieties protected by plant breeders' rights is still low. In the EU/CPVO system six varieties have active PBR. Over twelve applications are being investigated.

Propagation of olives can be done in various ways. The most widely used technique is mist propagation of leafy stem cuttings, obtained from one year old vigorous shoots. Also grafting is still being used. Scions (4-5 cm long) of young shoots are grafted on seedling rootstocks. Micropropagation using axillary buds occurs, but this method is costly and the results for many cultivars are not yet very successful.

Diseases

Relevant diseases in the production of olives are *Verticillium dahliae*, *Pseudomonas* and the olive fly *Bactrocera*. Especially *Verticillium* is causing a lot of concern and lower susceptibility of cultivars (full resistance is not (yet) available) is important when creating new plantations. Olive fly can cause very great damage in fruit production and the quality of olives. Strategies to



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 **mercado** 

Olive oil called **liquid gold** for good reason

- battle the pest are the use of insecticides in combination with IPM techniques and also the use of less susceptible cultivars. Biological control is in development but this gives not yet full protection.

The most dangerous disease however is *Xylella fastidiosa*, for olive a relatively new pathogen that occurs in Europe (South Italy/Puglia) since 2013. There are no methods to cure infected plants. Measures from the European authorities to try to stop/slow down the spread of infections to other regions in Europe are in place. Main vectors are humans (transporting infected plants to other regions) and an insect vector of the disease (meadow spittlebug, *Philaenus*). In the last ten years no other olive production sites in Europe have been infected, although *Xylella* (some

other variants) have spread to fruit production of *Prunus* in Spain.

Another important strategy to combat diseases in vegetatively propagated plants like *Olea* is propagation hygiene and propagation in a disease control and prevention programme.

The EU (EPPO) developed in the 1990's a recommendation for a certification system for olive trees/varieties and rootstocks. It is operated on an (voluntary, non-official) basis by producers of young olive trees. Crucial elements in this certification programme are pathogen testing of viruses/viroids/phytoplasmas, maintaining pre basic material in protected circumstances and retesting mother plants. 🌿

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“Making vegetables available for everyone.”

This is the ambition of Hilal Kanik and Canan Acarbulut, tomato breeder and selection co-ordinator tomato respectively, both working for Rijk Zwaan in Antalya. Read their story on rijkszwaan.com.

Sharing
a healthy
future



Freedom of choice or licence to produce

Daniël Ende

16 Is sustainability something you can and should measure in variety trials and should there be threshold criteria to be able to register a variety in the EU? What would that mean for farmers' choice and genetic diversity?

• **The European Commission is reviewing** the current EU seed marketing directives. These directives regulate, among others, the requirements that Plant Reproductive Material (PRM) of (new) varieties have to meet in order to gain access to the European market. The focus is on identity, quality and plant health. For example: new varieties of agricultural field crops, such as wheat and potato, have to meet official requirements in variety registration based on Distinctness, Uniformity and Stability (DUS), as well as a good enough or better score on value for cultivation and use (VCU). Moreover, all the seeds and tubers sold on the market have to be certified for identity, quality and plant health by national competent authorities.

For vegetables, currently only DUS is required for market access, where no official VCU is carried out and the quality control is a private responsibility under official supervision. DUS tests are also used in the process of granting plant breeders rights (one key – several doors).

In ornamental crops, even DUS requirements are not mandatory, plant breeders for ornamental crops can make do with an officially recognised description (ORD) of the variety. Quality control is also a private responsibility.

With all these differences in the ways market access is regulated, it seems to be a complex system.

Simplified regulation

One of the core motivations of the European Commission to start the policy review is the idea that the legislation has to be simplified, that it has to be harmonised and terminology aligned more. There are

eleven directives now. In addition, goals regarding sustainability, biodiversity and climate adaptation, as defined in the Green Deal, should be integrated more explicitly in the new legislation. The amateur market and seed conservation networks and the organic sector receive special attention in the efforts of the European Commission in the ongoing review. Some of the current seed marketing directives first came into effect in the 1960s; you could call them quite old. Political and policy attention for sustainability, however, is not exactly new either. The 1987 Brundtland report, 'Our Common Future', put the issue of sustainability on the policy tables, defined as: "meeting the needs of the present without compromising the ability of future generations to meet their own needs". That was 35 years ago. Let us look at some of the challenges the European Commission faces in the review and how they interact with sustainability and diversity.

Level playing field

One argument for the review is a more harmonised approach in the EU. The level playing field seeks to achieve fair and equal opportunities for competitors in the same market in the European Union. This poses the first challenge; what is the market for plant reproductive material? Is it fair to label all the trade in PRM as one single market? Then why have different directives for types of PRM, or even the same species for different value chains?

Is it fair that sunflower for agricultural production has to go through all the hassle of official DUS and VCU testing and certification, whereas sunflower for the ornamental market can register based on an offi-

The issue with heterogeneous material

The value of diversity in relation to sustainability is clear. A small number of mainly organic plant breeders ask to enter the market with heterogeneous material, populations mostly. This is plant reproductive material that does not fit in the regular variety registration DUS; by definition they lack uniformity, within the population and through the years as the population evolves, making field performance unpredictable at best. Even though there may be distinct benefits in cultivation of such material, it also introduces great

risk to fair marketing and food security. As it is impossible to define the variety, there is a risk of fraud. Another issue arises in registration of such material in distinguishing material from one another. Moreover, there are crops for which variety definitions in DUS protocols are already heterogeneous in nature, such as clover and grasses. Within the organic sector, some control is possible through organic certification. Outside the organic sector, this control is not in place.

Dr. D. Ende is policy officer at Plantum, Gouda, the Netherlands, d.ende@plantum.nl

Sunflowers are an excellent example of the differences between the regulations that an agricultural crop and an ornamental have to meet. The first one has to undergo official DUS and VCU testing and certification, whereas the second one only has to be officially described and registered



cially recognized description and that is it? And would sunflowers for ornamental purposes have to comply with the same sustainability criteria as those grown as field crops in a very different production system?

Performance indicators

How does one measure or prove sustainability performance? If your focus is on agricultural field crops, where the current system is originally based on food security policies, it might make sense to add some traits in the VCU-trials on top of current disease resistance, abiotic stress and other agronomical traits that benefit yield and harvest of quality products in a certain region. You could add traits such as water or nitrogen fertilizer use efficiency per yield output or protein produced, and possibly additional disease resistance. The logic is plainly visible, unless complex additional tests will significantly increase costs, which may reduce the number of varieties offered for testing, and result in less varieties contributing to diversity on the market.

Vegetables

Should the European Commission impose criteria for all crops, including vegetable and ornamentals, this would create more problems than it solves. At first glance it seems easy enough, but if you think about it or try to define what those criteria should be for vegetables and ornamentals, you will quickly see why this is much easier said than done. Variety testing, like VCU, is a performance comparison between new varieties and varieties already in the market. The VCU crops are tested under open field conditions and testing for disease resistance, abiotic stress, etc. culminating into yield. This yield for agricultural

crops in most cases is a raw material for further industrial processing: cereals are processed into flour for bread, pasta, etc., sugar beets for sugar, sunflower for oil, etc. When you look at vegetables, it is much less straightforward, which makes fair comparison testing impossible.

Just think of the number of trials you would have to design. There are easily more than thirty important vegetable species that we cultivate in the EU. Cultivation systems are varied as well; there are varieties developed for outdoor or indoor cultivation, for spring, summer or autumn harvest, for cultivation in a greenhouse or screen house, foil tunnel, as a rootstock or graft, and each variety has its optimum temperature for the optimal yield. Then there are different uses for the harvested products. Tomato is a good example: some varieties are suitable for direct consumption in a salad, fresh or sun dried, others are better suited for pasta sauces or ketchup, purée, etc. Then there is consumer preference in colour, size, flavour, etc.

Just this selection of aspects relates to various hundreds of viable market segments - good luck creating standardized trials for market access based on these differences, with or without sustainability criteria. Even in the agricultural field crop potato there are so many market segments (starch, chips, fries, table, specialties, etc.) that there is a push towards regulating that crop, more resembling vegetable market access without VCU trials.

Diversity is sustainability

In 'Our Common Future' (<https://bit.ly/3u9oR5B>), part of the problem described is that "...the planet is also being impoverished by the loss of races and



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From a regulatory viewpoint, there is a huge difference between an ornamental sunflower and an agricultural one



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varieties within species...” (p. 125) and that we need to preserve all biodiversity. It is good to notice here that, under the European Green deal, there is a Biodiversity Strategy that has the intention to combat biodiversity loss. Are we losing diversity even before it is made available on the market?

Ornamentals is the market with the most liberal (almost) no threshold market access regulation. The consumer benefits from this in a near endless diversity of flowers in many species, shapes, colours and sizes to meet every trend and make people happy. The only thing regulated is directed towards a fair market; varieties are identified in a cheap and lightweight way and are registered in order to assure that the creator of that variety benefits from it in a fair way and to prevent fraud.

In vegetables the stakes are higher. The market, cultivated areas and investments in new varieties of these crops are much bigger than for ornamentals. This warrants stricter variety registration demands, as well as more importance to distinguish strictly between varieties. Therefore DUS testing is mandatory for market access. An added benefit is that some disease resistances are tested as a trait in DUS testing. Quality assurance is a private affair between the seed company and the farmer.

In agricultural crops, we deal with mostly staple foods that provide food security. Just look at the problems arising worldwide when the Ukrainian wheat production is uncertain for 2022. In agriculture, we are dealing with tens of thousands of hectares of cultivation and high stakes for food security and affordability. This warrants the strictest market access rules. DUS testing shows identity of varieties. In official VCU tests, we find objective independent information that makes the next yield potential predictable for farmers

and through quality certification the farmer can trust the quality of the actual seeds to sow, tubers to plant, etc.

Freedom of choice

For an implementation of new seed marketing directives that contribute to actual sustainability, the European Commission has to strike a balance between unlimited diversity on the market and discriminating among new varieties that would contribute to defined sustainability traits. Farmers benefit from the freedom of choice; they know soon enough which variety best suits their cultivation method and local circumstances. A problem that arises when every new variety must contribute to defined sustainability traits is that often new varieties with a novel beneficial unique trait are not among the best scoring varieties overall.

If the variety is denied market access, the novel trait will not be available for cultivation nor for further plant breeding by competing plant breeders. This is in direct conflict with the Brundtland definition of sustainability, as such an approach limits variety within species and thus jeopardizes the chances of future generations to provide for their needs. It is not just about that single variety, but also its offspring that may benefit future generations.

Even though it is useful for farmers to receive information on sustainability traits of new varieties, it should not result in thresholds to the market beforehand. The European Commission must allow for maximum diversity in market access for new varieties. Every compromise towards diversity should be kept to an absolute minimum and only for good reasons, like direct threats to food security and a fair market. Sustainability will benefit. 🌱

An important step towards apomictic crops

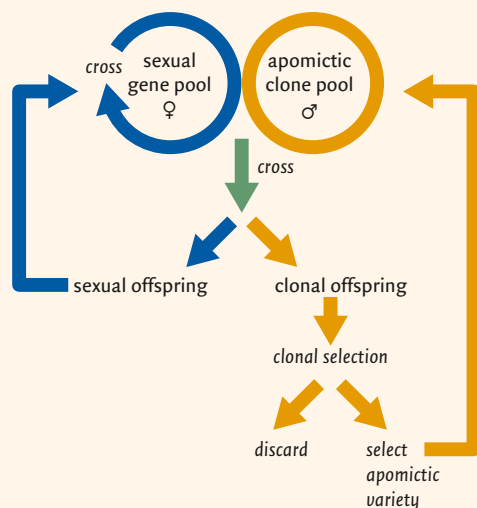
Peter van Dijk

20 If we look at the faces of two monozygotic human twins, they are nearly indistinguishable, while among a large population of non-twins, no two faces are exactly alike. Undoubtedly, face shape is a very complex genetic trait. Monozygotic twins make us aware of the exceptional high fidelity replication capacity of clonal reproduction.

Apomixis breeding with a sexual gene pool and an apomictic clone pool. In the dandelion system, apomixis genes can be backcrossed to the sexual gene pool to generate many different apomictic clones, which can be selected based on early trials. High performing new apomictic clones can be multiplied and brought to the market as new varieties

In plants, apomixis is clonal reproduction via seed. Apomixis results in offspring genetically identical to the mother plant. It is a natural reproduction system that occurs in low frequency in many plant families. It is estimated that 1 in every 1,000 plant species is apomictic. Some apomicts are well-known, such as dandelions and blackberries. Apomixis does not occur naturally in important agricultural crops and is a long-cherished wish for plant breeding. As early as 1935, the great Russian plant geneticist, Georgi Karpechenko, wrote: "Maintenance of desirable gene combinations would be possible if we introduce apomixis into valuable selected plant material." Apomictic hybrids would preserve heterosis of hybrid vigour in perpetuity. Of course, many crops can also be asexually propagated through vegetative reproduction, but apomixis has a much higher multiplication rate, cheaper storage and transport, and avoids spreading viruses.

sexual selection



In the absence of apomixis in crops, researchers turn to wild plants to identify the genes underlying apomixis. Developmentally, there are two crucial differences between apomixis and sexual reproduction. First, meiosis must be skipped (apomeiosis); otherwise, the chromosome number is halved, and the parental chromosomes recombined. Second is the development of the egg cell into an embryo without fertilization, called parthenogenesis. It has

been found that two major genes control these two components in natural apomicts.

Recently, scientists from the plant research company KeyGene (Wageningen, the Netherlands), together with scientists from Wageningen University & Research, Plant and Food and Lincoln University, Lincoln, New Zealand and the Japanese plant breeding company Takii, identified the gene for parthenogenesis (PAR) in dandelion. Moreover, they succeeded in having this PAR gene in lettuce make parthenogenetic embryos (Figure 1), a breakthrough that they published in January in the leading scientific journal *Nature Genetics*.

PAR breakthrough

By means of radiation-induced deletions, KeyGene researchers were able to narrow down the location of the PAR gene to a chromosome region of 40 predicted genes. By knocking out the genes in this region with CRISPR-cas9, one gene was found that led to the loss of apomixis. Putting this gene back in the plant with the deletion resulted in the restoration of apomixis, thus proving that this was the correct gene. The PAR gene is a variant of a gene that also occurs in sexual plant species. In other words, parthenogenesis is not caused by completely different genes but by an altered expression of normal sexual genes. Therefore, introducing parthenogenesis in sexual crops is a real possibility, even without GM techniques.

The PAR gene is the first parthenogenesis gene discovered in a dicot species. Another parthenogenesis gene, BBML (BabyBooM-Like), had already been found in the monocot grass *Pennisetum*. Like PAR, this is also a variant of a gene present in the genome of sexual plant species. This was further supported by the finding in rice that the atypical expression of the sexual BBML gene in the egg cell caused parthenogenesis. In the egg cell of sexual rice, the BBML gene is not expressed, but the active paternal allele is introduced from the pollen tube upon fertilization. To put it simply: the apomictic egg cell thinks it is already fertilized.

There is a revival of apomixis research. This trend is fuelled by technological developments, such as long-read genome sequencing and CRISPR-Cas gene-editing technologies, that now make it possible to identify genes in non-model species. The results

Dr. P.J. van Dijk is Distinguished Scientist at KeyGene N.V., Wageningen, the Netherlands, peter.van-dijk@keygene.com



A demonstration of apomixis breeding. There are eight neo-apomictic hybrid lines between the sexual rubber dandelion and the apomictic common dandelion generated by crossing. The six progeny plants within each neo-apomictic line are morphologically identical, contrasting with the large morphological differences between the eight neo-apomictic lines

recently achieved also have a stimulating effect on further research. However, much work remains to be done. So far, no apomeiosis genes in natural apomicts have been identified, although several research groups are working hard on this. The apomeiosis gene in dandelion, called *DIP*, from diplospory, is dominantly inherited and only affects female meiosis (pollen meiosis is normal). KeyGene has identified the *DIP* locus and candidate genes in this locus are now under investigation.

Endosperm factor

So far, this article has focussed on clonal embryos without fertilization to maintain desirable gene combinations. However, the endosperm, a tissue that nourishes the developing embryo, also arises from fertilization (so-called 'double fertilization'). In nature, most apomictic species form sexual endosperm through fertilization. However, dandelions produce endosperm autonomously, without fertilization. In such a system, pollination is unnecessary, making seed production much cheaper and more reliable once introduced in crops. Autonomous apomixis is also attractive because populations of insect pollinators are on the decline worldwide.

What could apomixis crop breeding look like in the future? Like finding the apomixis genes, KeyGene researchers used a 'copy-dandelion' strategy. The crux is that dandelion apomicts produce normal pollen. They are clonal in the seeds but sexual in their pollen. That makes it possible to cross apomicts with sexual plants. Some of the offspring will inherit the genes for apomixis from the pollen donor and thus will be apomictic. Their offspring are genetically identical. To demonstrate that this is not only a theoretical idea but also possible in real life, KeyGene has developed

an apomixis-breeding showcase. Inter-specific hybrids were created between two dandelion species: the apomictic common dandelion and the sexual rubber dandelion. Both species are highly heterozygous. After a screen with DNA markers for the dominant *DIP* and *PAR* apomixis genes, plants with the markers were grown and seeds were harvested. The morphological variation in the progenies is shown in Figure

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1. Between neo-apomictic lineages, there was great morphological variation. However, within these, there was great uniformity. Variation was fixed within neo-apomicts, although the progenies were highly heterozygous. This example is a clear demonstration of apomixis breeding.

Figure 2 shows schematically apomictic breeding according to the dandelion model. Selected new apomictic lines with desired properties can be multiplied quickly in great numbers by seed and tested at an early stage in trials. Apomictic plants selected after the trials can be backcrossed as pollen donors with plants from the sexual gene pool. Well-performing apomictic plants can be propagated and brought to the market as a new variety.

Contrary to what is sometimes heard, apomictic crops therefore do not mean the end of plant breeding. Another concern sometimes raised is that apomixis breeding will lead to monocultures and decrease genetic variation. However, apomicts are highly heterozygous, and continuous development of new varieties will be necessary to battle co-evolving pests and pathogens. Just as the development of the printing press resulted in many different books at the end of the medieval period, we think that apomixis breeding will give rise to the production of many different varieties.

A reality

A well-functioning dominant apomixis allele has to be developed only once because it can be introgressed via pollen in different sexual backgrounds. We may see the first apomictic crops in the fields by the end of this decade. Although that may seem a long way off, it is not much longer than the time it takes to bring a completely new variety to the market. 🌱

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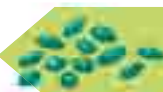
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Refugees learn to grow their own food

‘It was difficult to believe that we could cultivate the rocky soil and produce something to eat,’ says Edna Asante

Monique Krinkels

Some 1,750 refugees’ households in the Rhino refugee camp in Uganda have dramatically improved their farming skills thanks to the Knowledge Transfer team at East-West Seed. Despite the covid-19 pandemic, the Nutrition and Income Generation Intervention project, which started in 2019, has been an overwhelming success.

In early March 2018, 22-year-old Edna Asante, together with her husband and three young children, had to flee from their hometown in South Sudan. As for many South Sudanese citizens, war, violence and hunger drove the destitute family to this decision to save their lives. With over a million people on the run, South Sudan is one of the largest refugee crises in Africa.

Hospitable country

Neighbouring Uganda is one of the most hospitable countries in the world, in a region that is faced with economic crises, political instability, war and ethnic violence. It is the third largest refugee-hosting nation in the world. The country’s government has made welcoming and caring for refugees an important part of its national policy. Uganda has therefore set up relatively friendly regulations that ensure refugees have rights to education, work, private property, healthcare and other basic social services. Not only people from South Sudan, but also from the Democratic Republic of Congo have found safety in the many refugee camps.

Until recently, every refugee was entitled to a small plot of land on which they could build their own cabin and grow vegetables in a kitchen garden. “In the beginning, life was very hard,” says Edna Asante. “We relied only on the food rations given to us by the World Food Programme, which consisted mainly of maize and beans.” Her family had been allocated a small plot of land of 50 x 50 metres in Rhino Camp in the North West of Uganda. It houses over 185,000 refugees, mainly from South Sudan.

“The plot of land we were settled on was very rocky. We lost hope that we could farm the land, as it was difficult for us to believe that we could grow vegetables in this soil,” explains Edna Asante. That changed in 2019. “We were mobilised in our respective communities and briefed about kitchen gardening by East-West Seed Knowledge Transfer. The trainers explained how to proceed. We couldn’t wait to set off to achieve what they promised us. Practical training in vegetable production began, which led us to the promised land. Now we are able to produce more than enough vegetables to avail of a balanced diet for our household and even sell some to buy our household necessities like soap, sugar and salt.”



The project focused on reaching refugees as well as farmers from the host community with East-West Seed Knowledge Transfer activities. The aim: household nutrition as well as vegetable market and business development

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It is not only refugees who profit from the training. The Knowledge Transfer team also reached out to the farmers in the community around the Rhino refugee camp. East-West Seed established 237 demo fields on which they trained 7,287 farmers.

Using the learning plot as a hub for vegetable training, they trained community-based volunteer trainers on vegetable production, use of digital media to reach out to non-operational areas, trained like-minded partners, implemented vegetable production projects and trained agro-input dealers to enable them to easily provide accurate advice to farmers. Quality agro-inputs proved to be vital in improving extension services to the farmers.

Asante’s dream

The results were staggering. The participants had realised an average of 611% return on investment for tomatoes, 512% for watermelon, 621% for cabbages, 470% for onions, 611% for eggplants, 481% for pumpkin and 340% for kitchen gardens.

“Together with my husband, we are glad that life is slowly improving,” says Edna Asante. “We continue to practise what we have learned in our daily farming activities. We are glad we now have knowledge of vegetable production. Hopefully, when the situation in our country stabilises, we can return home. Our plan: to start commercial vegetable production in South Sudan and train our neighbours to do the same.” 🌱

Fleuroselect from pencil to pad

Sally van der Horst

24 Until recently, experts at the Fleuroselect trial grounds judged entries during the season with a notebook and pencil in hand. The final conclusions were entered manually into an Excel file and sent to an Evaluation Committee for final review, and the all-important evaluation of the potential winners. Today, the judges can enter their findings directly into a software environment that captures, calculates and stores the data online.

• Each year around 25-30 new ornamental varieties compete for the coveted Fleuroselect Gold Medals or are entered for Fleuroselect Novelty Protection. • Before an Evaluation Committee, supporting the Fleuroselect Board of Directors, can come to a final conclusion, expert judges spend a full season assessing the entries on a regular basis, evaluating where they meet required criteria and documenting their findings. The administrative part of this comprehensive task has now been streamlined using Mercado software.

The trials

In the spirit of Members for Members, Fleuroselect trials take place at its own member locations, both at breeding and distribution companies. Currently 15 locations all over Europe offer both their expertise and space in their greenhouses and trial fields to run the trials. Categories include outdoor field trials and indoor pack and pot plant trials. In keeping with market developments, new trial categories include container production field trials, patio trials for varieties more suited to performance in pots, and trials for the upcoming trend in patio vegetables. Characteristics for evaluation include innovation, flowering season, tolerance and floriferousness and importantly, the judges also assess the variety's potential use and value for the Grower, Retailer, and Consumer. Each trial location nominates a judge or group of judges who monitor the trials throughout the season. Up until 2021, not much had changed in the scoring method since the first judges walked the trials 50 years ago. Trial book in hand, the judges reviewed the trial throughout the season making notes on how each of the varieties performed, until at the end they sent in their final evaluation to Fleuroselect on paper giving numeric scores and adding comments. Fleuroselect then entered these scores and comments in a standard excel file for final evaluation by its Evaluation Committee panel of experts and by the Fleuroselect Board of Directors who approved the final results.

Change to Mercado

The Fleuroselect organisation first looked to fully digitalise its trials over five years ago. Various software applications were available including E-brida, but these were all dedicated to breeding trials, rather

than pure evaluation trials. With the development of Mercado, Fleuroselect found a software package which could be adapted to suit its specific requirements. Information on entries, including their cultivation information and an initial image supplied by the breeder, is added onto the platform where it is available for all the judges to view. Trial holders can then call up this information when sowing and planting the trial. When the time comes to evaluate the performance of the trial, judges enter their findings via an online app. At the end of the trial season the judge can review his findings and make one final upload into the system. This information is then processed, evaluated by a panel of internal experts and finally the results can be published.

The information in the system can be shared with the breeder company entering the variety so that they are able to see the performance of the variety over the full set of European trials and use this for internal evaluation. The organisation uses the results to determine the Novelty Status and/or the Gold Medal qualification. One of the major benefits Fleuroselect has seen in the Mercado application is the photography module. Judges can take and upload images throughout the trial season. These are available for own use but are also indispensable for evaluation by the committee and finally for the entrant, who can physically see how the variety performs across all the trial locations. Any breeding company will tell you that archiving photographs during the breeding process requires considerable organisation and Mercado is able to simplify this.

Across the water

When Fleuroselect was set up in 1970, there was already a successful trialling organisation in the U.S.A., All America Selections, which had been testing both flowers and vegetables since the 1930s. All America Selections already started trialling with Mercado in 2020 and once again Fleuroselect looked to follow suit. While trials in the U.S. do not judge for internal variety protection and include a full range of vegetables for home gardening as well as flowers, the trialling systems are basically similar. Fleuroselect thanks All America Selections for sharing their expertise in the development of its own dedicated Mercado software.

S. van der Horst, BSC.
is Secretary General of
Fleuroselect, Voorhout,
the Netherlands, sally@
fleuroselect.com.



Each year on average 25-30 new varieties are evaluated during the season by experts at 15 trial fields throughout Europe. Judging criteria include innovation, flowering season, tolerance and floriferousness

The history of Fleuroselect spans over half a century. Back in 1970, a group of the top seedsmen of the day founded the Fleuroselect organisation. Their key objective was to test or trial new varieties of ornamental pot and bedding plants bred by members for two main purposes: to grant Novelty Protection, and to award the very best in breeding. Fleuroselect Novelty Protection is a unique form of variety protection based on a Gentleman's agreement whereby if the variety is proven in the trials to be sufficiently uniform, new, and different from existing varieties, all members of the organisation agree to respect the ownership and refrain from reproduction. They also agree to refrain from selling the variety from any source other than that approved by the breeder. This protection can be purchased for up to 25 years and currently over 300 varieties are on the protection list. Should a variety in the trials be of exceptional value and show a breakthrough in breeding, then the variety can be awarded a Fleuroselect Gold Medal. The Gold Medal is recognised worldwide as a mark of breeding excellence and gives a huge promotional push to a variety as it enters the sales chain. It is supported by a full promotional campaign from the Fleuroselect organisation, and many Gold Medal winners have become standards in the ornamentals industry.

Product & Trial management

Several years ago, Agri Information Partners (also supplier of breeding software E-Brida) indicated a gap in tooling between the R&D/Breeding processes and the effective introduction, positioning and sales of a variety. Mercado was developed to bridge that gap.

Clients were looking for a solution to manage all their varieties and information in one central location. Varieties were placed on the product website, used in the catalogue, and trialed in different regions and circumstances. Photographs were taken and shared, and information was scattered around the company in different systems, excels files and folders. Mercado



has been developed as tool to provide that sole source of truth that customers are looking for.

With Mercado the user oversees their portfolio in one location.

In managing product life cycles, traits, or trials a change need take place only once. From automatic website connection to catalogue and from trial set-up to online dashboards, information can be shared with colleagues.

All processes are unique which is why in Mercado the system is flexible. Trials can be set up and information collected with an easy-to-use app. Specific product life cycle phases can be defined, and segments managed. Once data have been collected, specific sets can be selected and shared both internally and externally.

Brand new

Entry into the trials is open to varieties which are new and not yet offered commercially. They can be either seed-raised or vegetatively propagated. Entrants choose whether they wish to apply for Novelty Protection and/or a Gold Medal. They provide details on the cultivation of the variety and recommend comparisons i.e., control varieties which are currently available in the global marketplace. The entries are evaluated by a panel of internal experts for their suitability and the panel can recommend alternative comparison varieties. A knowledge of the vast assortment of varieties and series in the pot and bedding plants arena is essential in securing a valid trial.

Support

As owners and developers of the software, Agri Information Partners has supported Fleuroselect with excellent backing throughout the process, helping to optimise and secure the future of the trials for the full benefit of the members and for the whole industry. Fleuroselect looks forward to rolling the software out again in 2022 and beyond, supporting members and judges with a software tool that meets modern standards while maintaining that very special individual expertise of the judges, without whom the trials could not succeed. 🌱

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Growing green

Monique Krinkels

26 For the seventh time, the Dutch International Horticultural Exhibition, Floriade, has been organized. Until 9 October, the world can discover the solutions innovators have found to make a densely built-up city greener, more fun, more liveable and more sustainable. 'Growing Green Cities' is the common denominator of sixty hectares of land, forty inspiring country presentations and a spectacular greenhouse complex.

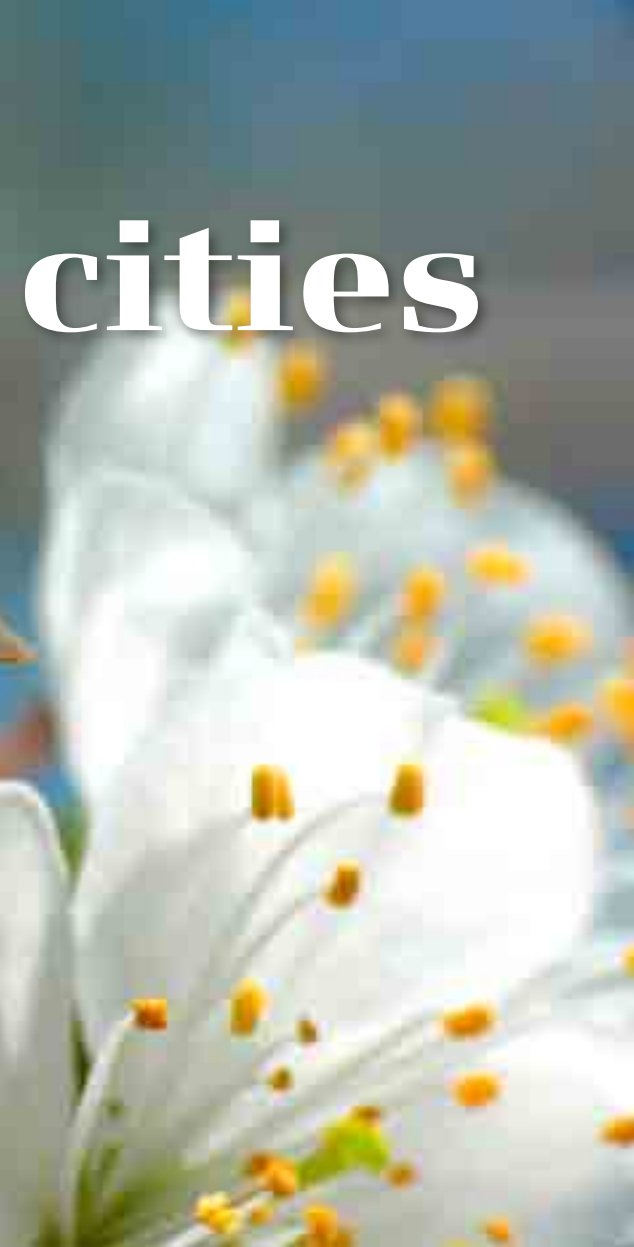


Growing Green Cities goes much further than planting a lot of green together. Think of new technologies, future products and a keen eye for solutions for global urbanization, food supply, health and energy production. The four sub-themes of Floriade 2022 are: 'Greening the City', 'Feeding the City', 'Healthying the City' and 'Energising the City'. Subjects that have been made visible in numer-

Each lot of the Classics & Future Moments project has a small square where you can stand to take a selfie with a floral artwork as the background



ous pavilions, the gigantic greenhouse complex, the arboretum and the rich art and culture programme. Over two million visitors are expected. Floriade started in the 1950s, when the Dutch Horticultural Council, in which ten horticultural organisations participate, sought a novel way to promote the Dutch horticultural sector to the world. In 1960, the first Floriade was held in Rotterdam. Since that event, every ten years a park has been designed. The Bureau International des Expositions (BIE) has recognized Floriade as a world horticultural exhibition that meets the A1 criteria of the International Association of Horticultural Producers (AIPH). This year, Floriade is being held in Almere, a relatively new, fast-growing city that was founded in 1975 on an impoldered part of IJsselmeer lake. It is situated four metres below sea level. The city is focused on sustainable green area development. 'The living lab', as Floriade is demonstratively called, shows on the one hand the aim of Almere to be a green city, on the other hand the ways in which the horticultural sector is constantly innovating in the field of sustainability



cities

With many flowering plants, such as the common white jasmine (*jasminum officinale*), Biotopia is a home to bees

and food production. Innovative participants from all over the world present their best practices to consumers and colleagues.

Much to see

The arboretum is the green engine of the Floriade. This gigantic living tree, flower and plant encyclopaedia is a feast for nature enthusiasts. All trees and plants are in order of their botanical name, after a creation by architect and urban planner, Winny Maas. It will constitute the Expo's green structure which you can walk through.

Among the many trees are black pines (*Pinus nigra*), but also *Pinus wallichiana*, that originates from the Himalaya, White, Grey, Canadian and Dutch poplars, and chestnut and dwarf horse chestnut trees. A lane of Japanese cedar trees serves as a shelter for birds and other small animals. In its country of origin, the Japanese cedar is considered as one of the most important forestry trees.

The Australian Eucalyptus trees might seem to be the odd one out, considering the warm and dry climate

in its country of origin. However, thanks to the mild winters we have had in recent years, they have been surviving better and better. They grow up to two metres a year, which makes them fast-growing forest trees. Ecologically, these trees from 'Down Under' have less to offer. Their essential oil, on the other hand is highly valued.

Flower art project

Close to the central square of Floriade, eight colourful gardens are laid out for the flower art project, Classics & Future Moments. Here, people walk along beautiful works of art, just like in a gallery, but these works of art consist of plants and flowers. The gardens are slanted, so you can admire them from afar.

More art that can be admired are trees to be found in the Weerwater, an artificial lake created by sand winning. The name, literally translated as 'again water', alludes to it being part of an impoldered area where later a water feature was added. The so-called 'Bobbing Forest' is an artwork created by the company, Mothership. It consists of twenty recycled sea buoys



The horticultural exposition covers sixty hectares



The arboretum is a living encyclopaedia with an abundance of flowers and trees

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Food Forum presents the Menu for the Future. This menu is based on sufficient, safe, healthy and contemporary food for everyone, without exhausting the earth. It is based on local and seasonal products and vegetables and legumes play a leading role

In the art pavilion M., five (inter)national artists take you on a journey into their future of nature



From the cable cars, visitors have a splendid view of the Bobbing Forest

from the North Sea filled with 20 Dutch elm trees.

Spectacular glasshouse

In the ultramodern glasshouse, 'The Green House', the Dutch horticultural sector will let visitors see, smell and taste the impact that horticulture has on everyday life. The striking design is inspired by Crystal Palace, which was built in London in 1851. Different types of glass and mirrors have been used to create surprising effects and remarkable lighting effects. 'The Green House' consist of three parts. In the 'Experiential Greenhouse', visitors can marvel at the attractiveness and variety of flowers, plants, vegetables and fruits. In the cultivation part of the greenhouse, the 'High-tech Greenhouse', visitors can see what innovative, circular and sustainable cultivation looks like on the basis of the four demo crops: strawberry, pepper, gerbera and potted anthurium. In the 'Technology Innovation Centre', the visitor gains insight into future cultivation methods such as a robotic vertical farm, drones and other types of robots. A total of thirteen competition days will take place

at The Green House, divided into forty crop groups. Growers of flowers and plants exhibit their best products in one of the crop groups and have them reviewed by a professional jury, specially appointed for this purpose. The jury consists of members from the production field, as well as from marketing and trade. There will also be a public jury.

Foreign participants

Not only the Dutch horticultural sector promotes itself at the Floriade; forty foreign countries add their own view on how their cities could become greener. Belgium, its Regions and Communities have chosen a triangular piece of land of about 500 m2 on the waterfront to promote the country's horticultural characteristics and green innovations. The pavilion's architecture is designed to minimise its environmental impact. Each material chosen is fully reusable or degradable, low-carbon, regenerable and natural: earth, wood, hemp, naturally compostable bioplastic, all these materials will be reused or recycled at the end of the Floriade in October.



In the German Garden, more than 3,000 perennials and grasses, 200 woody plants and almost 300 climbing plants can be found



In the Japanese pavilion, beautiful flower compositions named ikebana are demonstrated. It is also called 'kad' - the way of the flowers

With countless references to the triangle, the design created a playful space, a place of relaxation where families can take the time to try out wooden games. The Belgian Garden also offers an inspiring trail, for example the edible plant path. Germany presents sustainable solutions, ideas and concepts for modern urban horticulture under the name 'BIOTOPIA – Growing Community'. The country is making a valuable contribution to the exchange of ideas about one of the most important themes for the future. The exhibition presents new technolo-

gies, innovative products and examples of optimum realisation with a special emphasis on sustainability. More than 3,000 perennials and grasses, 200 woody plants and almost 300 climbing plants can be found in the German Garden. The garden is also filled with plants that attract insects, making a home for a bee colony.

Another remarkable participant is Qatar, with a design named 'Desert nest'. The pavilion is inspired by the well-known 'pigeon towers' that provided a resting place for pigeons in the desert. The traditional shapes of the four towers are combined with new 3D printing techniques and sustainable materials in the pavilion. Therewith, Qatar displays its vision, developments and innovations to make the desert greener in order to eventually realise green cities.

Japan shows typical horticultural and agricultural techniques that the country is proud of. Japanese floral splendour and culture will also be exhibited by, among other things, bringing 'ikebana' to Floriade. 'Ikebana' is the Japanese art of making flower arrangements with beautiful compositions. 🌿

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Official audits to check proper implementation

John van Ruiten

This year the EU has started a programme with official audits in Member States to examine if the new Plant Health Regulations and its requirements are being properly implemented in the daily practice of inspections and checks at operators' premises. In addition, the correct form/layout and use of plant passports is being judged.

Two years ago, the new Plant Health

- Regulation 2016/2031 in the EU came into force. New requirements for plant passporting, imports of seeds and plants, and high-risk plants were introduced. The long list of quarantine pathogens was evaluated and a huge number of diseases lost the Q-status and were

considering the widespread distribution nowadays. Other countries in the EU want to maintain the heavily regulated status and emergency measures related to it. There is especially a lot of concern about the recent importation of infected seedlots of tomato and sweet pepper from countries like

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China, Israel and Peru, in spite of the requirements with intensive inspection and PCR testing of all seedlots.

The EU is also very worried about discovering that plant passports were already on the packages of seeds originating from outside the EU. That is forbidden.



EU renewed Regulations for four important potato diseases are coming up

transferred to the new RNQP list. Specific tolerances were formulated.

New pathogens

This year, from 11 April 2022 onwards, a considerable number of new pathogens, mainly viruses in fruits (amongst others strawberry, blueberry and cherry) and grapevine species, is being added to the Q-list. Some of them are new, whereas others were regarded as RNQP up until that date. The tropical root-knot nematode, *Meloidogyne enterolobii* - naturally occurring in many countries such as China (where it was originally detected), the Americas and Africa - now also has a quarantine status. This nematode is quite harmful to eggplant, tomato, pepper and cucumber.

TOBRFV

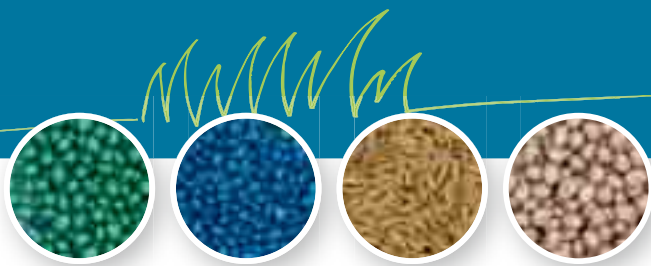
The TOBRFV dossier is still causing a lot of concern to the EU authorities, as well as to growers and seed and plant raising companies. A debate on the formal status of the virus has started. It is now Q-status, but some argue that it should be transferred to RNQP,

can be imported into the EU. This was an action last year that caused a lot of concern with companies that have regular production of cuttings in many third countries for export to the EU. The EU authorities announced that from 1 January 2023 onwards, the countries not only have to declare that they are free of *Xylella* (based on a monitoring programme) but they also have to supply statistical information on which this conclusion was based.

Potatoes

For the potato sector, a set of renewed, evaluated EU Regulations for four important diseases is coming up. It is expected that voting on these proposals will take place this spring and that they will come into force next year. The regulations contain requirements for the control and conditions to keep plant material (seed potatoes) free from *Globodera* spp (potato cyst nematodes), *Synchytrium* (potato wart disease), *Clavibacter sepedonicus* (ring rot) and *Ralstonia solanacearum* (brown rot). 🍅

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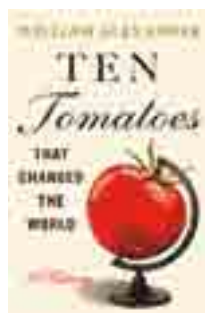
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SEED & SERVICES

Ten Tomatoes that Changed the World

An author on a mission

Monique Krinkels

- From a poisonous, stinky fruit, used as ornamental or even abused as a projectile, tomatoes have become number one in popularity as a fresh vegetable. In 'Ten tomatoes that changed the world', William Alexander is on a mission to get tomatoes the respect they deserve. He wrote a story that is an indispensable addition to the bookcase of people interested in the history of food and especially tomatoes.



'Ten tomatoes that changed the world' by William Alexander will be published on 7 June 2022 and costs US\$ 13.99

This could have become the shortest book review ever written: "Fascinating stuff". In 'Ten tomatoes that changed the world', William Alexander portrays the history of tomatoes from 1520, when Hernando Cortes conquered the Aztec capital city Tenochtitlan (today's Mexico City), until the present day. The book is not a simple summary of dates and facts, but is overflowing with anecdotes, interviews, travel stories, local knowledge and details well worth knowing which Alexander has unearthed.

Xitomatl

The history starts with the Franciscan monk Bernardino de Sahagún. In 1529, he described an abundance of varieties of xitomatl, which the local people used to flavour soups and stews, to fry and as a sauce ingredient. The Aztec people had been breeding tomatoes for at least a thousand years before the arrival of the Spanish conquistadores. Seeds of these fruits entered Europe in Seville, Andalusia, the Spanish port where the galley ships unloaded their booty. From there, the tomato would slowly, very slowly gain ground in Europe and the Americas. First as a (stinky) ornamental and – after 300 years – as a healthy food. In his book, Alexander relates how, in 1548, the Grand Duke of Tuscany, Cosimo de' Medici, received a strange fruit from the new world in the kitchen of his Palazzo Vecchio, Pisa, Italy. The Spanish family of his wife, Leonora Álvarez de Toledo, had sent him the seeds from Spain. The occasion was dutifully described by his steward in a polite thank you note, naming the fruit pomodoro or pomo d'oro (golden tree fruit). Half a century later, the ribbed, spherical pomodori were immortalized on the doors of Pisa's famous cathedral. It took a long time before the ornamental became a popular vegetable. In 1628, Giovanni Domenico Sala, physician in Padua, compared tomatoes with locusts, spiders and crickets, and names them 'strange and horrible things that a few unwise people eat'. In the Gardener's Dictionary, published in 1731, it is noted that 'the plants emit so strong an effluvium as renders them unfit to stand near a habitation'. It took until 1694 when a tomato recipe showed

Tomatoes are depicted on the 17th century carved doors of the cathedral of the Archdiocese of Pisa, dedicated to Santa Maria Assunta



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up in an Italian cookery book, 'Lo scola alla moderna', by Antonio Latini.

Valuable remedy

The popularity of tomatoes took off when in 1832 a cholera pandemic reached America. In New England, John Cook Bennett promoted the health benefits of tomatoes as, thanks to this vegetable, people are 'much less liable to an attack of Cholera and ... would in the majority of cases prevent it.' A few years later, tomato pills were developed. Competitors in this market were merchant Archibald Miles and physician Guy Phelps, calling it 'The most safe and valuable remedy ever discovered'. Besides protecting against cholera and syphilis, tomato pills would be beneficial against dyspepsia, jaundice, bilious diseases, gravel, rheumatism, coughs, colds, influenza, catarrh, nervous diseases, acid stomachs, glandular swellings of all kinds, costiveness, colic, headaches, etc. In Europe, Spain and Italy, people slowly got used to eating tomatoes. Pizzas were invented in Naples, Italy, around 1750 as a poor man's meal, but it took until 1880 before tomatoes became a common ingredient. At the same time, tomatoes were added to the Spanish traditional dishes, gazpacho and paella. The book also contains the history of Campbell's condensed tomato soup, introduced in 1897. As canning whole tomatoes was rather inefficient due to the round shape of the tomatoes, tomato soup saved transport costs and storage space. In the same period, the 'waste' product ketchup was introduced (made from overripe, unripe, wormy tomatoes unsuitable for canning). By the end of the 19th century, no less than 800 unique brands of ketchup were for sale in

the USA. But as consumption increased, the leftovers of the canning industry were no longer sufficient. In 1876, Henry J. Heinz, a son of German immigrants, moved into ripe tomatoes and increased the percentage of tomato solids to make his famous 'catsup', using his mother's recipe. The company became the world leader in this market, with more than 650 million bottles sold each year in the USA alone.

Consumption peaked

In the 20th century, tomato has reached a peak in consumption and production. The Dutch developed new ways to grow tomatoes in heated, artificially-lit greenhouses to become independent of climate and season. Alexander narrates, for instance, the hunt for a tasty tomato to be grown in Canada by Paul Mastronardi from Lemmington, Ontario. He found a remarkable tomato in Europe, a hybrid variety that combined juiciness, high sugar level, low acidity and lack of mealiness. Mastronardi Produce registered the name 'Campari' in the USA as a trademark, using the original variety and its offspring. Sales of this tomato-on-the-vine got a boost when Tony Soprano in the HBO-series, The Sopranos, preferred 'Campari' to make his Gabagool sandwich (Neapolitan slang for Capicola).

The last chapter of the book continues with recent and future developments that amaze the author. New varieties that produce ever better tasting tomatoes, which are grown indoors in mega-greenhouses or in vertical farms, in precision microclimates, managed by computers and harvested by robots, it is all new to him. 🍅

ISHI-Veg develops grow-out based **detection** method

Joyce Woudenberg, Hubert Lybeert, Roland Willmann, Smadar Kleiman-Shoval and Rose Souza-Richards

34 *Pseudomonas syringae* is a seed-transmitted bacterium which can cause Zucchini vein clearing disease of squash. Depending on the climatic conditions, necrosis of cotyledons and leaves, vein clearing of leaves, and stop of plant growth can occur. The seed industry joint efforts in developing a validated seed health test to detect *P. syringae* in squash seed.

Pseudomonas syringae is a bacterial pathogen that affects an abundance of plant hosts and occurs in a wide variety of environments. Over fifty different pathovars have been described and phylogenomic analysis showed that *P. syringae* does not form a monophyletic species in the strict sense, but a wider evolutionary group that also includes other species. In 2004, a *P. syringae* strain was identified as the causal agent of diseased plantlets of *Cucurbita pepo* subsp. *pepo* in plant growers' facilities in Europe. The bacterium was isolated from very young, stunted plants of *C. pepo* subsp. *pepo*, hereafter referred to as squash. This pathogenic bacterium was shown to be seed-transmitted, and depending on the climatic conditions, the symptoms could be strong (necrosis of cotyledons, necrosis of leaves, vein clearing of leaves, plants stopping to grow) or absent from the same source of seeds. Cool temperatures and humidity were the most favourable conditions for the expression of the disease, also referred to as the 'Zucchini vein clearing disease'.

Facing an increase of the Zucchini vein clearing disease incidence on the field, the seed industry felt the need to develop a validated seed health test to detect *P. syringae* in squash. Considering the known molecular diversity in *P. syringae* strains, the International Seed Health Initiative for Vegetable Crops (ISHI-Veg, an industry led platform for the development of seed health test methods) developed a grow-out based

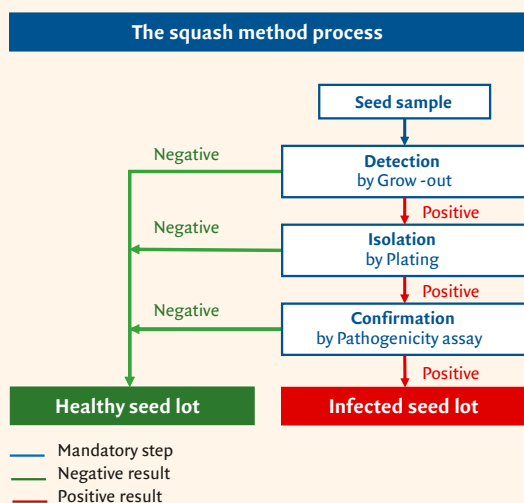
seed health method to test squash seed lots for viable, infectious *P. syringae*. A grow-out is an assay in which seeds in a seed sample are sown under disease-conducive conditions, and plants are subsequently examined for the presence of infection or disease symptoms caused by the pathogen.

Seed health test

The seed health method described by ISHI to detect infectious *P. syringae* in squash consists of growing out seeds under controlled environmental conditions in a greenhouse or climate chamber followed by isolation of the pathogen in pure culture from symptomatic seedlings. Its pathogenicity is confirmed on squash plants by a pathogenicity assay.

One thousand seeds are planted in a sterile potting mix in a greenhouse or growth chamber free from other sources of *P. syringae*. Relative humidity is maintained above 70% and temperature is maintained between 25 and 30°C during the day and 15 and 18°C during the night. After 14–21 days, when seedlings have two developed true leaves, each seedling is carefully inspected for symptoms. Isolations are made from seedlings showing any symptoms of disease on the selective levan-borate-cephalexin medium amended with actidione and lincomycin (LBC+AL). To confirm the pathogenicity of the suspected *P. syringae* colonies, identified by morphology, colonies are transferred to a fresh plate of LBC+AL medium and a bacterial suspension is inoculated on four squash seedlings of approximately one week old by wetting the upper side of both cotyledons with a cotton swab soaked in the bacterial suspension. Inoculated seedlings are incubated for seven days at 18–24°C and 50–85 % relative humidity after which they are scored for pathogenic reactions which are characterized by necrotic spots on the cotyledons.

A colony is considered as positive if at least one inoculated seedling (out of the four) shows symptoms. A seed lot is considered positive if at least one colony obtained from the suspected plants give a positive result in the pathogenicity assay. In order to avoid potential cross contamination when performing the grow-out assay, it is recommended to follow ISHI-Veg Best Practices (<https://worldseed.org/our-work/phytosanitary-matters/seed-health/ishi-veg-method-development/>).



Dr. J.H.C. Woudenberg, Dr. H. Lybeert, Dr. R. Willmann, Dr. S. Kleiman-Shoval and Dr. R. Souza-Richards are members of the International Seed Health Initiative for Vegetable Crops (ISHI-Veg), International Seed Federation, Nyon, Switzerland, R.SouzaRichards@worldseed.org



The symptoms of Zucchini vein clearing disease vary from necrosis of cotyledons and/or leaves to none at all

The validity of the grow-out based method for the detection of *P. syringae* in squash seed was assessed according to the ISHI-Veg guidelines for the Validation of Seed Health Tests (https://www.worldseed.org/wp-content/uploads/2020/12/MVGuidelines_v3_November-2020.pdf). Validation is a process that determines the fitness of a method for its intended purpose and is a requirement of method development. ISHI-Veg has identified six performance criteria to make the assessment, which are in line with those used by other accreditation bodies for developing seed health tests, analytical specificity, analytical sensitivity, selectivity, repeatability, reproducibility and diagnostic performance.

The performance criteria measured during method validation confirmed that the grow-out method for

the detection of *P. syringae* from squash seeds is suitable to detect contaminated seed lots with viable and infectious *P. syringae* bacteria in squash. All 36 pathogenic *P. syringae* isolates derived from squash grow-outs tested in the specificity experiments yielded typical symptoms and 14 non-*P. syringae* isolates derived from squash material yielded no symptoms in the grow-out and pathogenicity assay, demonstrating the analytical specificity of the method. It was shown that the grow-out assay can detect one pathogenic *P. syringae* infected seed out of 1,000 seeds, the required analytical sensitivity.

Additionally, it was demonstrated that the bacterial seed load that causes symptoms differs significantly per strain. Experiments on 14 seed lots representing 10 different squash varieties, coming from five different production years and six different countries of origin, showed that there is no significant variety-related or production conditions effects that influence the method, showing the selectivity of the method fits for purpose. The repeatability of the method was demonstrated by comparison of the homogeneity, comparative test and stability results performed by the organising laboratory, which were in alignment. The three participating laboratories in the comparative test found the healthy lot to be healthy and the two contaminated lots (highly and medium infection) to be positive for *P. syringae*. The ten repetitions of 100 seeds in the comparative test resulted in the testing of 1,000 seeds, the recommended minimum sample size to detect *P. syringae* on squash seeds. Therefore, the reproducibility of the method was also considered to be fit for purpose. 🌱

ISHI-Veg

ISHI-Veg aims to secure the delivery of sufficiently healthy seed to customers by developing methods for seed health testing that are internationally recognized as reference methods and accepted as industry standards (<http://www.worldseed.org/our-work/phytosanitary-matters/seed-health/ishi-veg-method-development/>). The validated grow-out based seed health method to test squash seed lots for viable, infectious *P. syringae* was first published online in March 2022 by the International Seed Federation (ISF). (<https://www.worldseed.org/our-work/phytosanitary-matters/seed-health/ishi-veg-protocols/>) together with its validation report (<https://worldseed.org/our-work/phytosanitary-matters/seed-health/ishi-veg-validation-reports/>).

Warming climate is a windfall for cabbages

Tijs Kierkels and Jeroen Balemans

36 **Brussels sprouts and cauliflowers look nothing alike. Despite this, botanically, they are the same species: *Brassica oleracea*. And their ancestor grows on the rocky coasts of England and France: the wild cabbage. However, the types of cabbage we eat today did not originate from England or France at all, but from more than 4,000 kilometres away.**

That the ancestry of cabbages is not European is not just interesting for historians, but for plant breeders too, who, due to climate change, are working on creating sustainable and resilient crops that are better able to withstand drought and heat. Guusje Bonnema, Plant Breeding researcher at Wageningen University & Research, can be rightly called a 'Brassica specialist'. She has been working with cabbages for over twenty years. "Their diversity is fascinating. With tomatoes, there are big ones and small ones, but you always eat the fruit. With

cabbages, you can eat the leaves (kale, head cabbage), the stalks (kohlraabi), the axillary buds (Brussels sprouts) or the flowers (cauliflower, broccoli). Previously, we researched how this huge variation could have come about. But that still did not answer the question of where it happened." In an article in the scientific journal *Horticulture Research*, PhD student Chengcheng Cai and Bonnema, together with colleagues from Wageningen University & Research, describe their search. "It was only possible because we managed to collect an unprecedented number

of accessions from all different *Brassica oleracea* vegetables; modern hybrid varieties and old land races from gene banks from all over the world," says Bonnema.

It took a lot of effort to gather all of the varieties, but it turned out to be a gold mine. The researchers made DNA fingerprints and were able to use markers to estimate the relationships between the different varieties.

Tin trading

There were two steps to domestication. "Greek and Roman writers mentioned these Brassica vegetables as far back as 400 BC. They described very diverse kale-like varieties, probably like curly kale and palm cabbage, but also described already very large cabbages. These were varieties selected from the wild cabbage," she says. After the first step in domestication in Western Europe, the second took place in the Middle East. The genetic research shows that these old, kale-like plants from Western Europe played a role. But how did they end up in the Middle East?

Bonnema: "It is highly likely that the tin trade played a large part. Tin was mined in Cornwall and Galicia and brought to the Middle East by ship around 2,500 BC. The boatmen took vegetables and seeds with them for the journey." This is how early kale varieties ended up in the Middle East. It is likely that both head cabbages and cauliflowers were selected from those first leafy kale crops. Spontaneous crossbreed-

The Brassica family has produced a wide range of vegetables. From white cabbage, broccoli, Brussels sprouts and cauliflower to Chinese cabbage, kohlrabi and kale



Convergent domestication

Cabbage, kohlrabi and cauliflower are related, domesticated by farmers centuries ago from the wild *Brassica oleracea* progenitors. In Asia, an entirely different species, *Brassica rapa*, developed into crops with similar appearances, such as Chinese cabbage, pak choi and turnips. Together with scientists from the Chinese Academy of Agricultural Sciences, Wageningen UR scientists have explained how it is possible that these two Brassica varieties at two very different global locations developed into so many diverse, but often very similar crops.

Brassica species were probably so suitable for domestication and breeding due to the triplication in their genomes. This means that large segments of DNA and many genes are present in the genome in triplicate. Sixteen million years ago, this triplication developed in an ancestor of the Brassica species. These so-called paralogous genes ensure that variations of genes can be selected in the offspring. This occurs without any loss of essential functions, as these functions are still performed by the paralogous genes.

More information: <https://www.wur.nl/en/newsarticle/How-Chinese-cabbage-and-white-cabbage-became-alike.htm>

Ir. M.J.M. Kierkels is independent journalist and co-editor in chief of the Dutch horticultural magazine *Onder Glas*; drs. J.J.P.F. Balemans is communications consultant at the Plant Sciences Group of Wageningen University & Research, Wageningen, the Netherlands, jeroen.balemans@wur.nl

The *Horticulture Research* article can be found here: <https://academic.oup.com/hr/advance-article-abstract/doi/10.1093/hr/uhaco33/6532230>



Wild cabbage on the French cliffs

Analysis method

First, the researchers set up large field trials using the collected accessions. Guusje Bonnema: “We needed a phenotyping of characteristics, and we also had to see whether material from gene banks was actually what it should be.”

They then took DNA fingerprints, using DNA material that they extracted from the young leaves of all 1,000 different accessions. “This is easy with modern hybrid varieties; they are all the same. With gene bank material from cross-pollinated plants, the accessions are heterogeneous. We then chose a single plant per accession, in the hope that it would be representative,” she says.

It is of course unfeasible to determine all 600 million base pairs in the DNA of all 1,000 accessions and compare them with one another. That is why the DNA was first cut into small pieces, which were fitted with adapters, and that subset of DNA was processed by the sequencer. This resulted in 14,152 locations where the DNA bases differed, and these so-called SNPs were used to signify the relationships between all 1,000 accessions. “These SNPs were fairly well distributed over the chromosomes, so that you got a representative image. Using techniques from biosystematics and population philology, we compared each accession with all the other accessions,” she says.

The results are notably visualized on a heat map and a relationship tree diagram. On the heatmap, colours indicate the relationships between the accessions. Blue is identical, dark red very different. What stands out immediately is that the cauliflowers are all very similar genetically.

“A phylogenetic tree – relationship tree diagram – is not so easy to create. You think with a tree that it keeps branching out. But because the varieties are also crossed with each other or with wild species, branches sometimes come together again. That is difficult to visualize; such a tree therefore only provides a general picture. You can clearly see that there are two separate domestication lines,” says Bonnema.

ing with other wild local Brassica varieties that grow on Mediterranean coasts may have played a role too. These old varieties developed into modern cabbages. “So our current cabbages all come from there,” she says. The new varieties travelled back to our region with the increasing trade.

Climate change

Why is it important to know all this? “We see that genetic variation in modern hybrid varieties is not that substantial, while the variation in the gene banks is much greater. If a plant breeder is looking for new crop characteristics, it is best to start here. In Turkey, Syria and Lebanon - where our cabbages originated from - many land varieties with a great deal of diversity still exist. These are countries with warmer and drier climates. To respond to climate change, we need varieties that can better withstand hot summers in the field. Now you can look more specifically at where to get these characteristics from,” says Bonnema. There have been numerous interesting insights during the research. For example, the cauliflower turned out to be a strange case: it is an inflorescence that no longer blossoms but continues to grow. “A whole series of mutations were necessary for this particular form. It was kind of a genetic bottleneck the species passed through. As a result, all varieties of cauliflowers are very similar; there is very little genetic variation. But the genetic distance to other *Brassica oleracea* vegetables is still very large. Every other vegetable (cabbage, kohlrabi, etc) has more in common with the wild cabbage than with the cauliflower,” she says. 🌱

Regulation of seed too often excessive

Bénédicte Lebas and Rose Souza Richards

38 Several regulated pests are not scientifically justified, resulting in unnecessary phytosanitary measures. For a pest to be regulated on a crop, there needs to be scientific evidence to demonstrate that firstly the crop in question is a natural host for the pest and that secondly seed is a pathway for the host in question under natural conditions.

• **Eggplant** (*Solanum melongena*) is an important vegetable crop with a worldwide production that has steadily increased over the years to reach more than 566 million of metric tons for the year 2020 according to Statista, ranking it as the fifth largest vegetable crop. Eggplant was first domesticated in Asia and later cultivated in Europe.

Maize (*Zea mays*) is the most important cereal crop with a worldwide production of 1,207 million of metric tons for the year 2021/2022 according to Statista. Maize is a staple food for many people. Furthermore, there is an increase in the use of maize in industrial products such as biofuels.

International Movement

Vegetable and grain seed are produced in many countries, shipped to central facilities for processing, sanitation and upgrading, treating, testing, and repacking before being re-exported. Seed moving internationally are subject to phytosanitary regulations to minimize the risk of introducing or spreading pests worldwide. However, in some instances the phytosanitary measures imposed are unnecessary as seed is not a pathway for the entry, establishment or spread of the pest in question.

Currently, the number of regulated pests on eggplant and maize seed are 76 and 166, respectively (<https://pestlist.worldseed.org/public/pestlist.jsp>). However, the crop is not a host for 19% of these pests. Furthermore, out of the regulated pests for which eggplant and maize is a known natural host, seed is not a pathway for 36% and 63% of the regulated pests in maize and eggplant respectively in accordance with the ISF database.

For a pest to be considered an issue on a crop, there need to be scientific evidence showing that the crop in question can spread the pest, meaning it is a natural host for the pest before considering its regulatory status as per the International Standard for Phytosanitary Measure (ISPM) 38 – International Movement of Seed. For all the pests listed in the table, no such information was found. Some pests are known to have a specific host range. For example, the bacterium *Acidovorax citrulli* is a pathogen of cucurbits while the oomycete *Albugo candida* is a pathogen of cruciferous crops. Both pathogens are regulated on maize although maize is not a host. Another example is the

formae speciales of fungi which is based on the pathogenicity to a plant species. *Fusarium oxysporum* f. sp. *melonis* (in eggplant regulated pest list) and *Fusarium oxysporum* f. sp. *cucumerinum* (in maize regulated pest list) are pathogens specific to melon and cucumber, respectively.

Natural hosts

Furthermore, some pests can have a wide experimental host range, but their natural host range is rather limited. For example, *Columnea latent viroid* and *Pepino mosaic virus* are both experimental host of eggplant but there are no reports of natural infection. Several factors can favour mechanical transmission under experimental conditions, such as, type of isolates and its concentration used for inoculation, the cultivar species, and the environmental conditions. Therefore, it is of utmost importance to consider the host range under natural field conditions as recommended by ISPM 38. Pests for which eggplant or maize is a natural host but seed is not a pathway can be classified in different groups based on their pest biology, their life cycle, and their epidemiological characteristics. Plants can harbour a range of pests that affect different parts of the plants such as leaves, stem, fruits but not seed. This is exemplified with foliar pathogens such as *Cercospora zeae-maydis* (fungus) in maize, *Peronospora hyoscyami* (oomycete) in eggplant that can affect leaves, stem but not seeds.

This is similar for pest that mainly inhabit in soil and plant debris and can affect roots, leaves and stem

More to read

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Dr. B. Lebas and Dr. R. Souza Richards, RPLI, International Seed Federation, Nyon, Switzerland, R.SouzaRichards@worldseed.org



List of regulated pests that are not known to be a natural host in eggplant and maize. Pest that can experimentally infect the host have their name followed by (E). Further details can be found at <https://pestlist.worldseed.org/public/pestlist.jsp>.

Type of pest	Eggplant	Maize
Bacterium	<i>Pseudomonas corrugata</i> <i>Pseudomonas syringae</i> pv. <i>aptata</i> <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>	<i>Acidovorax citrulli</i> <i>Burkholderia gladioli</i> pv. <i>alliiicola</i> <i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i> <i>Herbaspirillum rubrisubalbicans</i> <i>Pectobacterium atrosepticum</i> <i>Pseudomonas syringae</i> pv. <i>lachrymans</i> <i>Xanthomonas axonopodis</i> pv. <i>vasculorum</i> <i>Xanthomonas axonopodis</i> pv. <i>vitians</i> <i>Xanthomonas vasicola</i> pv. <i>holcicola</i> (E)
Chromista	<i>Spongospora subterranea</i>	-
Fungus	<i>Fusarium oxysporum</i> f. sp. <i>melonis</i> <i>Synchytrium endobioticum</i> <i>Thecaphora solani</i> <i>Verticillium albo-atrum</i>	<i>Calonectria ilicicola</i> <i>Cercospora sorghi</i> <i>Choanephora cucurbitarum</i> <i>Diaporthe sclerotioides</i> <i>Didymella holci</i> <i>Didymella pinodes</i> <i>Fusarium oxysporum</i> f. sp. <i>cucumerinum</i> <i>Fusarium roseum</i> <i>Microdochium nivale</i> <i>Pseudocercospora ulei</i> <i>Sarocladium oryzae</i> <i>Septoria cucurbitacearum</i> <i>Sporisorium cruentum</i> <i>Syncephalastrum racemosum</i> <i>Verticillium albo-atrum</i>
Insect		<i>Helix aspersa</i>
Mollicute		<i>Candidatus Phytoplasma asteri</i>
Nematode		<i>Aphelenchoides besseyi</i>
Oomycete	<i>Pythium spinosum</i> (E)	<i>Albugo candida</i>
Viroid	<i>Columnea latent viroid</i> (E)	<i>Pepper chat fruit viroid</i>
Virus	<i>Arabidopsis mosaic virus</i> <i>Pepino mosaic virus</i> (E) <i>Strawberry latent ringspot virus</i> <i>Tobacco rattle virus</i> <i>Tomato black ring virus</i>	<i>Squash mosaic virus</i> <i>Sugarcane streak virus</i> <i>Tomato ringspot virus</i>

Maize seed is not a pathway for 36% of the regulated pests in accordance with the ISF database

of plants but not seed. This is the case of most of nematodes (e.g., *Longidorus* spp., *Meloidogyne* spp., *Pratylenchus* spp.) which mainly parasite underground parts of plants. Other type of pests can also be associated with soil and plant debris because of their life cycle (or part of) taking place in this area. For example, the beetle *Diabrotica* spp. has its life cycle occurring in the soil where maize plants are grown and pupae stages of *Leptinotarsa decemlineata* and *Liriomyza trifolii* occur in soil of eggplant field.

Seed transmission

Sometimes pests are known to be seed-transmitted on other crops but there is no evidence of such event in the crop in question. This is the case of the virus Tomato mosaic virus which is transmitted through seed of tomato. Although eggplant is known to be a host, albeit rarely, there are no references found indicating seed as a pathway for this virus in eggplant. Similarly, for the fungus *Pyrenophora teres* which is known to be seed transmitted in barley (family Poaceae) but for which there is no scientific evidence in maize seed.

Other times pests have been demonstrated to be seed transmitted under experimental conditions in the crop in question, but no evidence was found that it can occur in the field. This is the case of the fungus *Phaeocytostroma ambiguus* in maize seed. In 'Maize diseases: A reference source for seed technologists', McGee cites two research articles that conducted experiments under laboratory conditions with artificially inoculated seeds, but no work was cited in the field.

Seeds can carry harmful pests and pest risk assessments are required to scientifically assess the risk associated with seeds. However, as demonstrated with the eggplant and maize regulated pest lists, several regulated pests are not scientifically justified, affecting the global movement of seeds. 🌱

39

Genetic **diversity** secures sustainable future

Anker Sørensen, Fernando García-Bastidas and Gert Kema

40 Banana production for export markets worldwide relies heavily on a single clone, the Cavendish banana. Cavendish produces very tasty fruits but is susceptible to a number of threatening fungal diseases. Resistance breeding and utilization of the genetic diversity available in 'wild' banana material will secure the sustainable future of banana production. The Yellowway R&D and breeding company is focused on achieving this goal.



Gross Michel plantation in the mountain area of Colombia, which is still free of *Fusarium Race 1* infection (A). Eventually, the soil-borne fungus will also reach these plantations, as can be seen in (B)

It is time for change. It is time for improvement. The worldwide dessert banana industry is huge at about 120 million tons or 660 billion bananas! Exported production accounts for about 17% of total production or 112 billion bananas according to the Food and Agriculture Organization (FAO). These exportable dessert bananas rely almost completely on the performance of no more than four genetic groups, including the globally dominant Cavendish cultivars. Yellowway is a new R&D company for banana research and breeding initiated by KeyGene, Chiquita and MusaRadix and supported by Wageningen University. The goal of Yellowway is to breed more sustainable and tasty varieties suitable for both domestic and export markets.

Biotic challenges

The main export banana variety, Cavendish, faces major biotic challenges caused by fungal diseases, that threaten global production. These are Fusarium wilt of banana (FWB), also known as Panama disease, and black leaf streak disease (BLS), commonly known as Black Sigatoka.

The most obvious challenge is the rapid spread of the infamous Tropical Race 4 (TR4) - a genetic lineage of soil-borne *Fusarium* fungi causing FWB. This lineage originates from South-East Asia and has now, since 2019, also reached Colombia and Peru, which are among the main producing countries in Latin America, triggering the alarm for the whole region as the dispersal of TR4 is hard to control. The other major threat is the air-borne fungus *Pseudocercospora fijiensis* that causes BLS, devastates plantations around the world and is the major reason for the unsustainable cultivation of bananas, as it can only be controlled by an extensive fungicide input, usually at weekly intervals throughout the year.

To date, there is no commercial effective control method for FWB. Only circa 17% of the total global banana production is dedicated to export, the remaining 83% is destined for local markets, where bananas are a staple food, covering much of the energy intake of the local inhabitants. These local banana varieties are also threatened by many diseases, including FWB and BLS. This underscores the urgency for developing new varieties for a variety of regions and climate conditions. Yellowway,

Dr. A. Sørensen is Vice President New Business at KeyGene N.V., Dr. F.A. García-Bastidas is Head Banana Breeding Programme/Panama disease at KeyGene N.V. and Prof. Dr. Ir. G.H.J. Kema is Professor at Wageningen University & Research, Wageningen, the Netherlands, contact: anker.sorensen@keygene.com



Indonesian market displaying local banana cultivars

therefore, also contributes to a project to genetically improve bananas for East Africa, which is funded by the Bill & Melinda Gates Foundation.

Indeed, banana breeding, supported with high-resolution genomic knowledge and molecular research and selection techniques, is ready to effectively address the various threats by delivering diverse, new and resistant varieties. We do not need a proof-of-concept: Cavendish bananas did the same trick, as it saved the industry due to their durable resistance to FWB caused by Race 1 strains around

the world since the 1950's. However, its success developed into a Cavendish monoculture that is now threatened by TR4 and – contrary to the past – there is no replacement available. As a consequence, growing areas are being moved to novel areas free of the TR4 fungus, which in time will also be infected. Hence, Yelloway was founded to modernize and accelerate banana breeding using science-based tools that are already common practice in many other crops.

Cavendish cultivars are triploid. They are sterile,

Yelloway

A unique alliance has been created in the Netherlands to meet the challenge of banana breeding; Yelloway. Chiquita, the major international banana company, whose mission is to grow and sell bananas around the world in a sustainable and socially acceptable way; KeyGene, the world leading R&D company in agricultural biotechnology, for crop innovation through molecular breeding; and MusaRadix, a Wageningen-based company engaged in enhancing social and technological innovations in the banana industry, have joined forces and established Yelloway. Together these companies have allied themselves with the Tropical Phytopathology group at Wageningen University, led by Prof. Gert Kema, the world leading expert in banana research and banana phytopathology. This alliance is uniquely positioned to understand and utilize genetic diversity in order to address the challenges of developing novel banana varieties.





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Banana diversity depicted as phylogenetic analysis, indicating the relative relatedness of the various wild-type bananas based on the different patterns of 7.5 million SNPs. The known ancestral groups can be clearly identified. The * indicates accessions carrying resistance to TR4.

A. Banana seed development after crossing



B. Germinating embryos dissected from banana F1 seeds in-vitro



C. Plantlets regenerating from banana seeds



heterozygous and vegetatively propagated. Hence, Cavendish is not the ideal starting point for breeding. Therefore, a novel route has been designed by the partners who founded Yelloway. The so-called wild, diploid banana germplasm is an invaluable source of genetic diversity for different traits (www.crop-diversity.org/mgis/), but it has hardly been explored, let alone deployed. It comprises groups of truly wild seed-bearing (non-edible) genotypes as well as parthenocarpic edible clones, which are often cultivated as landraces.

Over the last 10 years, a sub-set of accessions has been screened for diverse traits, and especially for resistance to FWB. Currently, we have compiled a com-

prehensive database of sources for resistance to the two major fungal diseases. Recently, this germplasm has been genotyped at high resolution (7.5 million SNPs) to categorize the accessions into phylogenetic clusters and to associate them with ancestral clones. Interestingly, the results indicate that germplasm with resistance to FWB can be found in many phylogenetic clusters across the germplasm collection.

This resource of genotypes and phenotypes is our basis for a genomics-based breeding approach. Current efforts focus on identification and a comparative analysis of resistance genes to build a database that allows us to accurately select and combine genetic resources in a tailor-made cross-breeding scheme. In parallel, our ambition is to understand the molecular mechanisms underlying resistance to FWB and BLS, using our genetic and genomic databases. This will help us to select and design the most appropriate schemes towards novel domestication of edible banana types that meet market demands but are - contrary to Cavendish - resistant to FWB and BLS.

Dutch greenhouses

A major challenge in banana breeding is the success of crosses – due to fertility issues – and seed set. We have developed protocols to grow bananas under controlled conditions in greenhouses in the Netherlands and to induce flowering. The timing of flowering is critical - per pseudo-stem, one truss develops and every flower in the truss is amendable to cross-fertilisation, but only for a very short time (few hours). Nevertheless, we established effective protocols for cross-breeding in the greenhouse and the first offspring populations have already been generated.

However, once the seeds are harvested, germination is a serious bottle-neck. Therefore, we have developed high throughput and reliable embryo rescue protocols to guarantee the survival of segregating populations. Every seed is a new and unique plant, representing a new recombination palette of characteristics, which potentially represents a new variety. With these protocols in place, we can now upscale the breeding process.

In addition to the genomics-assisted cross-breeding approach, we apply various other techniques to 'correct' Cavendish. These include genome editing and mutations breeding to develop TR4-resistant Cavendish. Preliminary results of these efforts by other groups around the world are promising and will lead to novel opportunities for banana growers. 🌱

Flower mixtures as food-to-go for insects

Monique Krinkels

44 Biodiversity, climate change and the survival of pollinators are trendy topics nowadays. In many countries, initiatives are being proposed to save the wild flora and to support bees and butterflies. A 'honey highway' on roadsides, dikes, industrial estates and fallow ground could be part of the solution. EconSeeds is specialized in creating seed mixtures that serve as food-to-go for these indispensable critters.

Worldwide, insect numbers are plummeting according to a global scientific review published in Biological Conservation. More than 40% of insect species are declining and a third are endangered, the analysis found. The rate of extinction is eight times faster than that of mammals, birds and reptiles. The total mass of insects is falling by a precipitous 2.5% a year, according to the best data available, suggesting they could vanish within a century.

Food

Apart from the influence of climate change and the detrimental effects of the use of insecticides, insects starve due to a lack of sufficient flowering plants. Bees visit flowers for pollen and nectar, butterflies for the nectar and as a food source for their caterpillars. They eat next to nothing else, so blooming flowers are crucial for the survival of these beneficial insects. And humankind does need them. Between 75% and 95%

of all flowering plants on earth - among which 1,200 food crops - need pollinators to propagate. Especially bees make excellent pollinators, as pollen is the only source of protein that they feed to their developing offspring.

In the Netherlands, half of the 360 species of wild bees are in danger of disappearing completely and the same trend can be observed in other European countries. Governments all over the world are well aware of the essential role pollinators play for food production, saving biodiversity and mankind's wellbeing. Countless initiatives from all sides are taken to save bees, butterflies and hover flies. Providing them with nutritious food is a basic requirement and that is what EconSeeds is specialized in.

EconSeeds

The history of EconSeeds is short. It was only in 2008 that Hans Veenstra took over the economy seed range,

Research

The EIS Insect Knowledge Centre, together with the Butterfly Foundation, has conducted research on behalf of the Dutch Ministry of Infrastructure and Water Management into the occurrence of butterflies, bees and hoverflies in the Honey Highway. The Honey Highway initiative provides food for bees, butterflies and other pollinators. So far, 500 kilometres of roadsides have been transformed to 'flower ribbons'.

The verges along one of the busiest motorways were sown in 2015 and have since developed into herbaceous vegetation with 38 types of flowers. In the study, the Honey Highway is compared with verges a little further along the highway that have been constructed in the same way, but have not been seeded. The spontaneously emerged vegetation there consisted of 28 types of flowers in 2019.

Last year, almost three times as many butterflies were found on the Honey Highway as on the control verges. The verges seem to be a success, especially for the group of farmland butterflies. Such high numbers of the brown sand eye (*Maniola jurtina*) and European common blue (*Polyommatus icarus*) were found that it is likely that these species reproduce in the Honey Highway. The hay bug (*Coenonympha pamphilus*), which is quite rare in the area, also turned out

to have found its way to the verge. Hoverflies show a similar result: considerably more were found in the sown flower strips than in the spontaneously developed verges.

The Honey Highway was less successful for bees. They preferred to stay on the control verges. Since 2017, the number of bees in the Honey Highway has actually decreased, while it increased in the control verges. Because wild bees have a limited radius of action (several hundred meters), it is important that the nest and food are close to each other. However, the Honey Highway now consists of high and dense vegetation and therefore offers food but little nesting space. There is more open sand in the control verges, where bees can dig their nests.

In addition to sowing with a suitable flower mixture, it is therefore also important that the verges are well managed. In the Honey Highway, areas with open sand could be created locally to make the flower ribbons more accessible to bees again. The study also found that the flowering roadsides (both sown and unsown) were richer in pollinators than the surrounding grasslands. Therefore, both types of roadsides have added value in a flower-poor environment.



Flowers pollinated by butterflies are often red or orange, because those colours attract butterflies



A pollinator hotel attracts beneficial insects

when the renowned Dutch flower seed company Kieft Seeds was purchased by Ball Horticulture. Both the OP flower varieties as well as the contract flower and vegetables seed production came into his hands. Eight years later, the French company

Bertrand, famous for its flower mixtures, was added and last year HM Clause's flower mixtures completed the colourful palette.

"In total, we have over 1,200 varieties of annual and perennial flowering plants," says Hubert Keller, commercial director of EconSeeds in Hoogkarspel, the Netherlands. Where Hans Veenstra has earned his spurs as seed production manager, Hubert Keller is focused on company development. The third person to complete the picture is Pierre Byache, who is responsible for composing the 170 different mixtures in the seed production facilities in the Loire Valley, France.

EconSeeds has thirty employees, half of which work in the facilities in France and the other half in the Netherlands. The company is growing steadily and they just moved to new, larger and more suitable accommodation. In France, the company owns several production locations in the surrounding area of Angers. Furthermore, there are about 150 growers in the Loire valley region who produce seeds, and many more in Chili, India and China.

Part of the Bertrand collection is also available as organic seeds. Six of the mixtures and 25 varieties of flowers meet the international organics standards. Among the seed mixtures, EconSeeds has introduced

a selection of edible flowers and aromatic herbs, used to decorate plates and to spice up summer salads. In addition, there are three organic varieties for the pharmaceutical industries: cornflower (*Centaurea cyanus*) against rheumatic conditions, Californian poppy (*Eschscholzia cal.*) against sleeplessness (hence the Dutch common name 'nightcap') and mari-gold (*Calendula officinalis*) with anti-inflammatory characteristics.

"Seed cleaning is done mainly in France. Batches are then sent to the Netherlands where the seeds are tested for germination in our laboratory, with state-of-the-art equipment. The next stage is to assemble packages of seed mixtures for professional, as well as consumer use according to the 'recipes' Pierre has provided us with," explains Hubert Keller. At present a brand-new seed mixing machine is operating in the Netherlands.

Artwork

"The art of creating mixtures is to assemble varieties that fit well together, create a nice picture, with plants of diverse sizes and produce flowers during different seasons. A single mixture may contain over 40 species, combining high diversity and a good balance of species, which develop day after day to create a natural and colourful environment," explains Pierre Byache. Not only to ensure visual pleasure to people, but also as a continuous source of food, as insects are picky creatures.

"For instance, bees are attracted by borage (*Borago officinalis*), also known as starflower, for its rich amount of nectar, but sunflowers and single corn flowers are also popular. Some wild bees are, however, highly specialized and are focussed on just a few species.

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Butterflies on the other hand do not only need their food security, they also seek out plants to safely lay their eggs on and feed the caterpillars. Corncockle (*Agrostemma githago*) is among their favourites.” Flowers pollinated by butterflies are often red or orange, because those colours attract butterflies. “Creating new varieties is not really necessary, with the vast number of varieties already in our possession. Only if a colour becomes less sparkling, do we actively select the brightest flowers to propagate.”

Ville fleurie

It is not only the insects’ preferences Pierre Byache has to take into account. As EconSeeds sells flower mixtures around the globe, the natural conditions such as soil, climate, shade or direct sunlight have to be taken into account. And the consumers’ preferences of course. “The French market for ornamentals is well developed,” says Hubert Keller. There are many ‘villes et villages fleuris’ (flowery cities and villages) in France. Hanging flower baskets in the streets, colourful roundabouts planted with annuals and perennials and brightly coloured flower beds in the parks are a common scene. It all started in 1959, when the French state launched a contest to enhance the attractiveness of urban areas. Successful com-

EconSeeds has over 1,200 varieties of annual and perennial flowering plants to choose from to create their 170 different flower seed mixtures for professional as well as amateur use

munes are awarded with a badge showing one to four flowers. Today nearly 5,000 cities have attained the ‘ville fleurie’ status. They display their flower badge on road signs as you enter.

“The demand changed from ornamental plants to seed mixtures, as budget cuts forced local authorities to look for a new approach to the contest. The Bertrand collection was superbly suited to be used in an urban environment,” according to Hubert Keller. “Today, many roundabouts and parks are floral paradises for insects, thanks to flower mixtures.”

The French consideration for the protection of bees is not restricted to urban areas. ANSES, the French Agency for Food, Environmental and Occupational Health & Safety, has made recommendations aimed at strengthening the regulatory framework for the protection of bees and other pollinating insects. EconSeeds supports this strategy. Last summer, EconSeeds took over the complete catalogue of seed mixtures of HM Clause, which possessed seed mixtures especially designed to be used on roadsides and fallow fields in the countryside.

Honey highway

In the Netherlands, the interest in flowering plants is as yet lagging far behind. Most local governments of cities and villages use easy to maintain grass and shrubs to provide their citizens with a green environment, only coloured by stray daisies and dandelions. However, this attitude is changing fast. “Flower mixtures are a growing market in the Netherlands,” Hubert Keller observes. One of the reasons is the initiatives to help the bees survive. For instance, during the last five years, over 500 kilometres of roadsides, dikes and railway tracks have been planted with flower mixtures, creating a honey highway, as this initiative is called.

The seed mixtures the Dutch government opts for must consist of plants that are attractive to insects, but should also be native to the area, thereby promoting biodiversity. It fits within the EU’s biodiversity strategy for 2030, which aims to reverse the degradation of ecosystems and manage them sustainably, addressing the key drivers of biodiversity loss. Using the carefully assembled flower seed mixtures of EconSeeds seems to fit in perfectly with this plan. 🌱

Quinoa defies climate change and salinization

Tijs Kierkels

48 The wild ancestors of quinoa thrive on poor, salty, dry soils near Lake Titicaca in the Andes. An excellent starting point for a crop that must be resistant to climate change and salinization and is suitable for a low-meat diet. The Plant Breeding department of Wageningen University has been working on ever better varieties since the 1990s. Nowadays with advanced techniques such as automatic phenotyping.

It is a familiar story: food production must increase for a growing world population while climate change and salinization are making growing conditions more difficult. You really need a crop that can withstand drought and salt well, and makes few demands on the soil apart from that. And then you end up with quinoa. For decades, the Plant Breeding department of Wageningen University has been the linchpin in the breeding of this crop, where numerous applications were reviewed, such as gluten-free 'grain', animal feed and cut flower. Nowadays, quinoa has the wind in its sails as a gluten-free super-food, rich in proteins, minerals and vitamins. But it started out much more down to earth in 1990. "Under the leadership of my predecessor Dick Mastebroek, we were looking for contenders for the fourth arable crop. The idea was that Dutch arable farming was too dependent on potato, wheat and sugar beet and that an alternative was needed. In collaboration with the Centre for Genetic Resources in the Netherlands (CGN), we came up with potential contenders: oil crops, fibre hemp and quinoa, then best known as 'gierstmelde'," says Robert van Loo, research leader of the Breeding for Abiotic Stress Tolerance programme. Quinoa has a cultivation history of thousands of years, but traditional varieties from South America had two unpleasant characteristics. "The ripening

is day-length sensitive – that is problematic in our region. And in addition, the seed skin is very bitter. It contains saponins, soapy substances, which give you a dry mouth and sore throat. That is why the seed skin must always be removed and the seeds must be rinsed," he says.

Ultimately, these were not the biggest hurdles. Both the day-length sensitivity and the bitterness each appeared to be based on only one main gene. Although the desired trait is recessive, by crossing and backcrossing patiently, the first non-bitter varieties emerged: Atlas and Riobamba. They found their way as an ingredient for gluten-free bread. "A solution for people with gluten intolerance, but that turned out to be a very small market. At a certain point, it collapsed," he recalls.

But there were other applications within sight. *Chenopodium quinoa*, as the Latin name goes, is a tall crop from the amaranth family with striking, colourful flower plumes. "Very suitable as a cut flower. It was supplied as a summer flower at the auction and the prices were quite good. But that application also turned out to be too fashion sensitive," says Van Loo. However, there are still a few growers who breed it to a limited extent.

Silage

The next step: application as silage. For example, on ploughed grassland, which has the advantage that the nitrogen present does not leach out. Quinoa, as mentioned, is not demanding. It did not work out and Van Loo is still a bit displeased about that. "My belief is that it certainly has prospects. The milk production of cows that received a silage quinoa mix was good. The protein content of the silage is lower than, for example, grass/clover, but it was possible to solve that by further breeding. We have become the victims of bad publicity after a somewhat unfortunate organic cultivation. So the development stopped immediately."

French élan

The breakthrough came from renewed



Field experiment with different genotypes of quinoa

Ir. M.J.M. Kierkels is independent journalist and co-editor in chief of the Dutch horticultural magazine Onder Glas, tki@xs4.nl.



The drought tolerance of 180 genotypes is currently being measured by NPEC

focus on human consumption. Furthermore, the impetus for this came from France, and indeed from the agricultural economist Jason Abbott, then working at Limagrain. “He was looking for solutions for people with gluten intolerance (coeliac disease) and came across quinoa as a good grain substitute at a conference for dietitians,” says Van Loo. Abbott promptly set to work and set up the cultivation in France together with a cooperative of farmers who had their own cleaning plant. They first meticulously made that gluten-free and then they were in business. However, they did really need better performing breeds. That is why Abbott knocked at the door in Wageningen, instead of, for example, the much more logical French INRA, because he knew about the breeding programme. A high-yielding silage type was at the origin of modern consumer varieties

You really need a crop that can withstand drought and salt well, and makes few demands on the soil

such as Dutchess and Bastille. They are high-yielding and also have large white seeds, which look good. “Under favourable conditions, they yield 2 to 3 tonnes per hectare. We are now working on a new series of varieties that can yield 3.5 to 4 and sometimes 5 tonnes. Incidentally, that is also desirable in order to be able to compete with grains.”

The cultivation in France now covers 4,000 hectares, worldwide it is about

6,000 hectares with Wageningen varieties, including in Chile, Argentina, USA, Canada and Japan. “You can run a breeding programme for that,” he says. The total global quinoa acreage is actually 200,000 hectares.

Farmers in the Netherlands also grow the crop, and then supply it to GreenFood50, located on the Wageningen Campus, which turns it into food ingredients. This company has the licence for cultivation in Benelux. Jason Abbott was in possession of the licences for the whole world, excluding Benelux, but sold them to Radicle Crops, also located on Campus in Wageningen. Wageningen University & Research receives royalty income from its varieties.

Low-meat diet

Over time, the focus in breeding has shifted from productivity to sustainability aspects, such as drought and salt tolerance. The special characteristics of quinoa mean that the crop is well suited in many aspects to a more sustainable agriculture. “With the right cultivation, ie. with a relatively low nitrogen supply, the seeds contain up to 15% protein. This makes the product very suitable for a vegetarian or low-meat diet. In comparison: chicken meat contains 20% protein. One hundred grams of quinoa is therefore equivalent to 75 grams of chicken in terms of protein consumption. Even though you need much less land for production. Cultivation clearly contributes to the transition to vegetable protein consumption,” says the research leader.

Another sustainability aspect is that the cultivation is compatible for more marginal soils and difficult climate conditions. “For that reason, we are in contact with research centres in Dubai, Saudi Arabia, China and Vietnam. These are all regions where drought

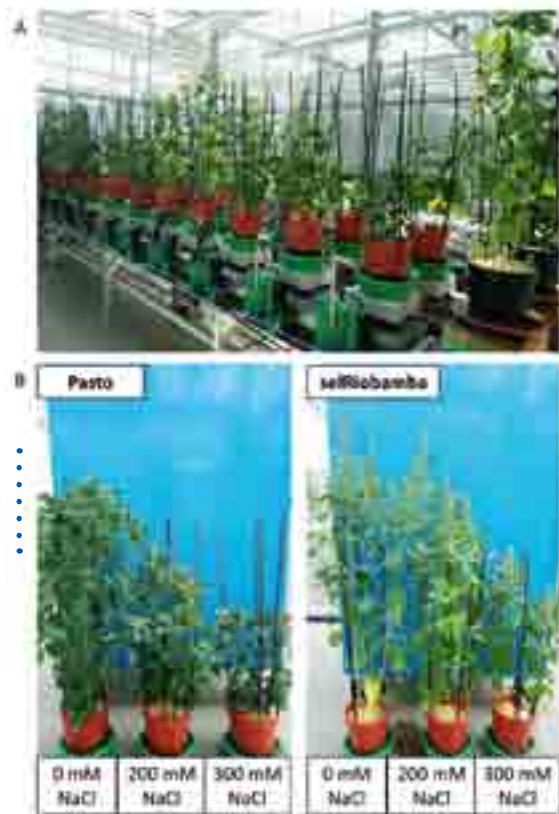
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Netherlands Plant Eco-phenotyping Centre

The Netherlands Plant Eco-phenotyping Centre (NPEC) plays a crucial role in the search for crops that are resistant to drought, salinization, diseases and other consequences of climate change. It concerns complex traits that are based on genetic traits, some of which are known, some of which are not. An extensive phenotyping makes it possible to link the desired traits to DNA markers. Phenotyping is the analysis of the external/discernible characteristics of a breed/genotype under certain conditions.

In NPEC, large quantities of plants of different genotypes are being exposed to different treatments and their observable reactions are automatically recorded. It concerns external characteristics such as size, leaf surface, leaf angle, colour and the like, but also leaf temperature, water absorption, transpiration and photosynthesis. The measurements are taken automatically; the data can then be analyzed with artificial intelligence. That is very useful information for breeders.

Wageningen University & Research is perfecting the method of automatic phenotyping together with NPEC. The facility is also open to third parties, such as breeding companies.



50 The varieties Pasto and selRiobamba each deal in a different way with increasing salinity. The first is a 'saver', the last a 'spender'

and salinization are a serious problem. Then you need crops that can deal with that.”

Quinoa has its origins on its side. A crop that slowly acclimatizes to increasing salinity, can even withstand seawater, although growth is then very marginal. That is of course extreme, but real salinity, for example an EC of 10 dS/m, is not a problem for good growth.

Variety comparison

“In collaboration with the University of Quito, Ecuador, we tested the salt tolerance in different varieties. Especially the potassium supply can become a problem with increasing sodium content. Quinoa is relatively capable of keeping the potassium/sodium ratio in order, but it differs per variety.”

Moreover, salt and drought provide the same challenge for a plant, so that salt tolerance in a variety automatically also means drought tolerance. The first research in this field was carried out by Viviana Jaramillo Roman as a PhD student, in collaboration with WUR colleagues. She now works as research and development manager at Radicle Crops. The research was published in *Frontiers in Plant Science* in August 2021.

She compared two existing varieties for the ways in which they deal with increasing salinity. Van Loo: “One breed closed its stomata fairly quickly to limit evaporation (and therefore water loss) as it became more difficult to absorb water. The other continued to grow. These are two different types of strategies: you have a ‘saver’ and a ‘spender’. In any case, it indicates that there is genetic variation for the reaction to salt. This gives the breeder tools for the appropriate ideo-

type for certain circumstances,” Van Loo explains. The ‘spender’ has the advantage that it continues to grow, but it does make its environment increasingly salty and therefore more unsuitable for itself and subsequent crops. The ‘saver’ grows more slowly but causes less exhaustion.

Growth and water uptake were monitored using the Plantarray 3.0 phenotyping platform, a precursor to the NPEC facility (see box). “The plants were automatically weighed every three minutes. Their water dosage, soil moisture, EC, irradiation, greenhouse climate and plant temperature were constantly measured automatically. This allows you to establish links between the stomata opening and the (cumulative) evaporation. You see daily patterns and the differences between the varieties.”

Accelerated breeding

The advantage of the continuous automatic measuring is that you only have to measure for a limited number of days. If everything had to be measured by hand – for example, the stomata opening – the research would take much longer, and even then there would be much less data. Moreover, the machine can do it more accurately.

The drought tolerance of 180 genotypes is currently being measured, this time actually in NPEC. “It’s the F2 plants that we have had since 2011. There is a lot of variation: big vs. small, bitter vs. non-bitter, and so on. We already have a clear picture of the genome; that was already published in *Nature* in 2017. Using the automatic phenotyping, we can visualize the reactions to environmental conditions and the build-up of biomass in a non-destructive way within a few weeks,” says Van Loo.

This is part of a project run by Topsector Horticulture & Propagation Materials. The intention is to achieve a faster method of breeding. The automatic phenotyping is a tool to be able to link traits to DNA markers. When those markers have been identified, it is easier to achieve F1 hybrids. Quinoa functions as a model crop; the method can then also do a useful job with other new crops. 🌱

Learn more: www.frontiersin.org/articles/10.3389/fpls.2021.634311/full

Seed drying & equipment

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Drying of seed
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(B) per box individual.
Per box a fan (C)
and heating source (D).
Extracting outside air (E),
dehumidified air (F)
or inside air (G).

Individual closed box dryer



Individual closed drying units for conditioned drying of seed in boxes.

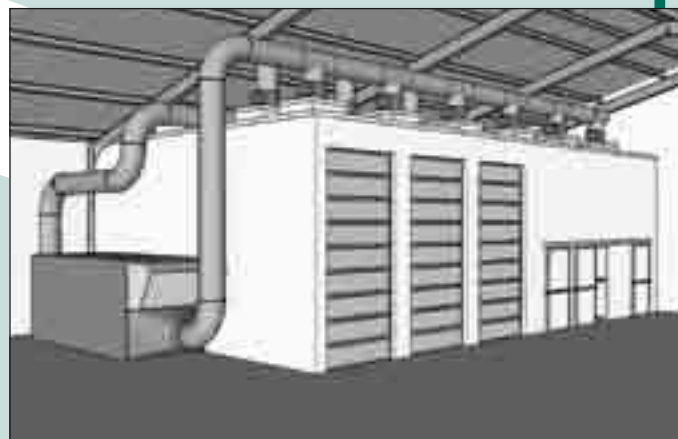


Optimal use of dried air

Central hybrid air dryer (left) to be connected to different drying installations, drying units or drying rooms; optimal and economic use of dried air.

Humidification of too dry seed

Humidification unit (A) to increase moisture content of too dry seed without making the seed wet. Damp air will be distributed through the seed by any kind of aeration system. The safest way for automatically humidification of your seed



Drying seeds in closed rooms and Seed vault for storage

Central hybrid air dryer for drying rooms (left) and seed vault (right). Storage of previous seed at 15°C and 20% RH or 10°C and 25% RH.

 **Agratechniek b.v.**
van Nieuwenhuizen

Anna Paulowna, The Netherlands

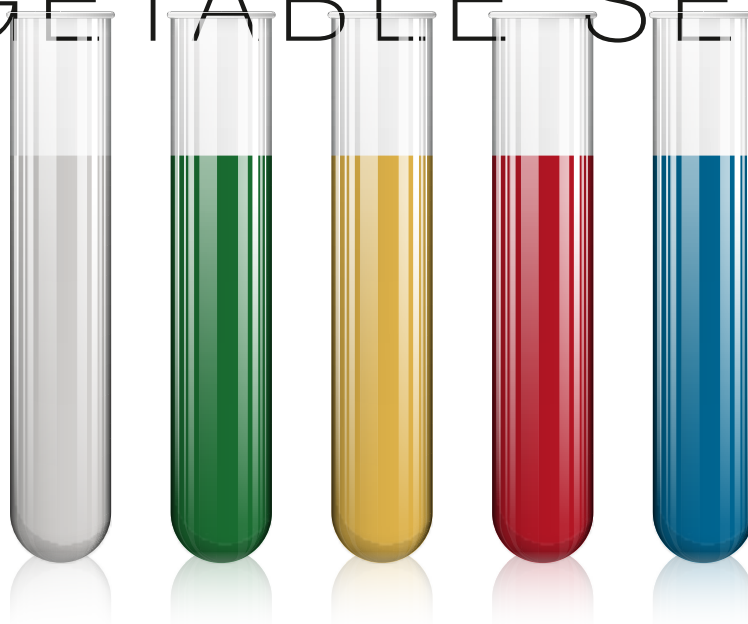
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