# pollination bag specialists





## investing in the future

Welcome to the third PBS International Research Update. In this edition we look particularly into the use of our pollination bags and tents in bioenergy crops, and follow-up on research highlighted last time.

You can also learn how, as part of our continued investment in research, we have received a prestigious Agri-Tech research grant, and we take on a picturesque project with tulips.

We bring the science of technical fabrics to the needs of plant breeders. The research featured shows how selecting the right pollination control, such as using our unique **dura**web<sup>®</sup> material, can offer huge economic benefit to plant breeding efforts:

- reduced contamination
- fewer repeats, hence faster development cycles
- substantial financial savings
- significantly enhanced seed production

## key findings

Aberystwyth University found that **dura**web<sup>®</sup> pollination tents for Miscanthus breeding produce >540% more seeds than isolation chamber crosses ✓

Recently published research from Oklahoma State University shows that **dura**web<sup>®</sup> pollination bags produce up to 100% pure self pollinated seed in switchgrass, higher than any other type of bag ✓

Early trials suggest that seed yields may be enhanced by choice of bag in the tulip breeding programme 
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### get in touch

To share your thoughts on any of the research in this update or suggest ideas for future research contact us at **support@pbsinternational.com** 

### pure-line production for switchgrass hybrid breeding

Research conducted by Oklahoma State University (OSU) into the development of inbred lines of switchgrass, *Panicum virgatum L.*, showed vast improvement in genetic integrity with the use of duraweb<sup>®</sup> pollination bags, which prevent contamination by foreign pollen (Adhikari et al, BioEnergy Research 2014).

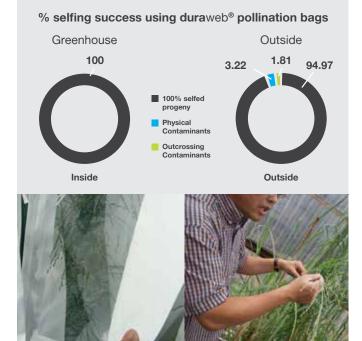
A warm-season perennial grass native to central and eastern USA, switchgrass is capable of growing on marginal agricultural land. Consequently, improved switchgrass lines have the potential to make a valuable bioenergy crop.

As a species which is naturally self-incompatible; blocking pollen from unwanted sources when attempting to self these plants is very important. Previous attempts made by OSU to develop inbred lines using 4 different bagging methods (Lawson paper bags, microfiber, muslin cloth, and pillowcases supported by chicken wire), resulted in a massive 39-80% contamination.

Researchers found that the 3D.75F **dura**web<sup>®</sup> pollination bags from PBS International raised selfing success in greenhouse conditions to an outstanding 100% success rate - no contamination or outcrossing of the progeny.

In the field, a more challenging environment on account of strong, persistent winds, they found **dura**web<sup>®</sup> bags gave 94.97% purity, with 3.22% physical contamination and 1.81% outcrossing. The authors conclude that small levels of contamination observed in field conditions were likely due to physical damage to pollination bags, handling, and due to high wind speeds.

The superiority of the **dura**web<sup>®</sup> pollination bags, acting as an effective barrier to external pollen, has allowed accelerated development of inbred lines and saved time on molecular verification of the progeny, making them an invaluable tool for switchgrass selfing.



Switchgrass inflorescences inside the duraweb $^{\circ}$  pollination bag (left) and Dr Yanqi Wu of OSU examining switchgrass plants in a glasshouse

### boost for Miscanthus seed yields

Research shared in the last research update, conducted by the Institute of Biological and Rural Sciences (IBERS) at Aberystwyth University, showed a substantial financial advantage associated with the use of duraweb<sup>®</sup> pollination bags in their *Miscanthus* breeding programme (publication pending). An economic analysis simulation across 1,000 crosses showed a predicted saving of £43,000 over glassine bags and £16,000 over plastic-and-paper bags as measured by cost-per-successful-cross.



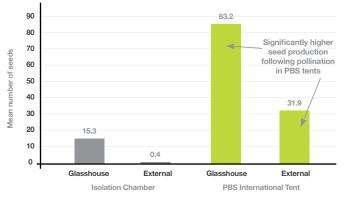
More recently, the team tested the performance of PBS International's **dura**web<sup>®</sup> pollination control tents for impact on *Miscanthus* seed production. Seed production rates were compared in PBS tents & plexi glass isolation chambers located outside & in glasshouses.

Once a potentially successful

*Miscanthus* variety has been identified, it is important to increase the amount of seed so that multi-plot trials and replicated plot trials can begin; failure to generate enough seed slows the progress of the breeding programme, or requires the use of extensive resources to carry out more replications of the same step.

The mean number of seeds produced per head was significantly higher for crosses in **dura**web<sup>®</sup> tents than in isolation chambers. For instance in **dura**web<sup>®</sup> tents inside glasshouse conditions an average of 83 seeds were produced per head compared to 15 in isolation chamber - a remarkable 543% increase.

It is thought that PBS pollination **dura**web<sup>®</sup> tents, like the bags, improve the crossing success rate and seed production by creating an optimal microenvironment. To test this, the researchers compared the temperature and humidity ranges, and found that **dura**web<sup>®</sup> tents showed smaller fluctuations for these parameters than the isolation chambers. Hence, the **dura**web<sup>®</sup> technology, which allows air and moisture to pass through whilst blocking unwanted pollen, may be the key to this.



#### Mean number of seeds per head

### does it stop foreign pollen?

For pollination control, the first requirement is to stop foreign pollen. At PBS International, we don't leave anything to chance and have several techniques to establish the suitability of materials for stopping particular pollen types.

### 1. calculate the pore size.

The typical method for this provides a maximum pore size and distribution of pore sizes. However a mean pore size of (e. g.) 40µm doesn't necessarily mean 20µm pollen grains will pass through. The "architecture" of the material affects the pollen's progress, and how likely it is to get stuck in the twists and turns a pore makes through the material. Even the shape of the pollen grains themselves can have an impact on its infiltration, e.g. spikey pollen grains would find it more difficult to pass through than smooth grains.

# 2. measure actual penetration of pollen grains.

This literally involves counting the pollen grains that pass through a given fabric at a given pressure. Wind speed, dampness of the material and mechanical stress can all affect pollen grain's success in passing through the fabric.

# 3. conduct a trial in experimental conditions.

This is easiest for out-crossing species where the female inflorescence is covered before anthesis, and any resulting seeds must arise from pollen gaining access through the cover, via damage or closure points. For hermaphrodite species the process may be more complex, requiring emasculation of female flowers or even

studying molecular markers in the offspring (see Adikhari et al 2014 example above). Sometimes simple phenotypic examination of resultant seeds can reveal contamination, e.g. colour of maize endosperm (Xenia effect).



We use all the above tests & more to assess the suitability of materials. We don't like to leave anything to chance when choosing products for a particular application, so contact us to discuss a material's suitability for your project.

### forestry revisited

In 2014 PBS International began to work with the North Carolina State University Tree Improvement Program (NCSU-TIP) to identify a product suitable as an alternative to kraft paper bags used by Loblolly pine seed producers in their Controlled Mass Pollination (CMP) process. Current methods cause concern that seed yields are not being maximized.

In spring 2014 & 2015 the work compared 4 materials and 3 designs alongside the current kraft paper bag, with and without a wire to keep the bag open. Results come in two stages, with flower survival after the "June drop" being the first indicator of performance. Cone and seed counts – the most important indicators of success - have to be calculated at harvest, 18 months after flowering. As a result all data are not available yet.

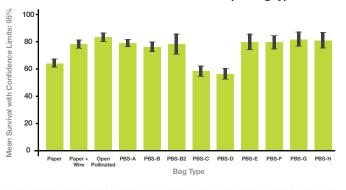
Results at this stage suggest that, unsurprisingly, the open pollinated control condition stands out as being optimal for flower retention. While the bulk of the remaining treatments are not significantly different from each other, one group of treatments were clearly unsuccessful – the kraft paper bag without a wire, and the "C-D" material. Nonetheless, usability research suggests that material "A-B" and the innovative cylindrical design used in the 2015 study are popular.

Ultimately no alternative to the traditional bag is a viable alternative unless it makes "sound economic sense". This is complex to calculate, involving the cost of labor to add wires and bags, the time taken to pollinate, cost of hiring equipment (eg lifts to reach as high as 80 feet) and the cost of the bags themselves. Some treatments may also provide a risk mitigation benefit against occasional weather events such as cold snaps.

The research will be concluded over the coming 15 months.



Mean flower survival in June per bag type



### product development for tulip breeding

Spring blooms were in focus as PBS International started a research programme to optimise seed yield in tulip breeding with Dr Arie Peterse of HobaHo, who conducts breeding operations for many Dutch bulb growers. Crossing often occurs in open fields for accelerated bulb development. Current crossing techniques can result in significant losses, up to 50% of some types of cross, from rain washing pollen off or contamination with foreign pollen. A method that prevents these problems is required.

Stopping foreign pollen by covering the flower with a pollination bag risks increasing the occurrence of disease due to lack of aeration. PBS International developed prototypes specifically for this setting, exploring material properties and bag design. Preliminary results suggest that current method of using aluminium foil over the pollinated stigma has similar seed yields to no coverage and that the best design / material combinations can enhance average seed yield, although further collaboration is required to be confident of optimal outcomes and an easy-to-use design.



### funding innovation

PBS International are delighted to announce the award of early-stage project funding worth over £300,000 through highly competitive Agri-Tech Catalyst programme, a joint venture from Biotechnology and Biological Sciences Research Council and Innovate UK, the UK's innovation agency.

The consortium in the project includes two academic institutes, and two UK based commercial organisations and a German seed producer. The project aims to explore the science and commercial feasibility to develop, from scratch, a new material which will exclude tiny pollen (15-20 µm) while allowing greater light penetration than existing options, and simultaneously encouraging climate control around the covered flowers. It is envisioned that new material will boost seed yields, reduce the need to repeat stages in greenhouse-based breeding programmes, and accelerate the pace of product development.

The work will continue for at least 18 months, and we are looking forward to pouring our decades of experience into this new product. We hope that it will transform the process for breeding crops such as sugar beet, grasses and wheat.

# We work with



#### what are others up to?

If you're curious about the range of breeding programmes going on, take a look at pbsinternational.com/news which frequently features bulletins about our customers' work.

For more information go to **www.pbsinternational.com** PBS International, Salter Road, Scarborough, YO11 3UP, UK +44 (0)1723 587 231

### introducing duraweb® mini bags

Following extensive feedback from our customers, we have extended our mini pollination bag range to include six new sizes.

Bag type	Code	Length (cm)*	Width (cm)*	Depth (cm)*
2D	M7.15	15	7.5	N/A
	M7.30	30	7.5	N/A
	M15.15	15	15	N/A
	M15.30	30	15	N/A
3D	3DM7.30.7	30	7.5	7.5
	3DM15.30.15	30	15	15

Expanding our mini bag range enables us to offer more standard options to breeders thereby saving money. These designs have already been used by a diverse range of programmes from Poplar & Willow breeding in New Zealand to Shaw's Agave conservation in the USA.

The mini bag range is made from the same **dura**web<sup>®</sup> material as our full sized bags to provide an equal level of breathability, durability, UV stability, moisture resistance and pollen protection for equally successful results whatever the size of your plant.

