



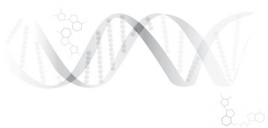
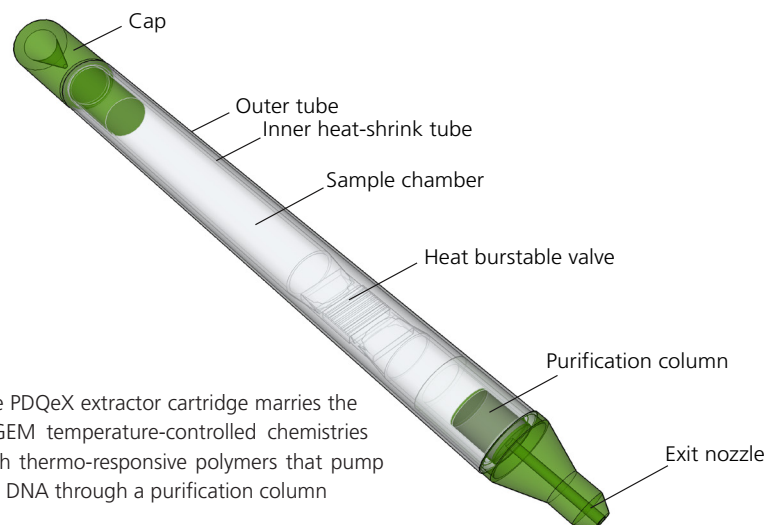
The PDQeX *phytoGEM* System
Rapid, automated DNA extraction from plants

PDQeX Portable and the PDQeX₂₄₀₀



The PDQeX Cartridge

Extracting DNA from biological samples can be laborious and frustrating. The PDQeX system from ZyGEM takes the tedium away by combining powerful enzyme-driven extraction chemistries with an innovative extraction cartridge.



The PDQeX *phytoGEM* System



Sampling, storage and DNA extraction in a few easy steps

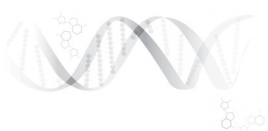
Sampling takes just seconds. Leaves are crushed in the field onto a storage card using a special tool. With long-term storage of samples built into the procedure, there is no extra work or freezers packed with bags and tubes.

The DNA extraction process using the PDQeX technology, is rapid and automated, going from leaf on a plant to PCR-ready DNA in approximately 15 minutes. The extraction chemistry uses ZyGEM's potent thermophilic proteinase combined with a cocktail of mesophilic cell wall degrading glycosyl hydrolases. These enzymes systematically lyse cells, destroy nucleases, digest the protein and release the DNA. Extractions are performed in disposable cartridges that are designed to remove inhibitory polyphenols and polysaccharides.

The *phytoGEM* system can be easily adapted to work with different types of plant tissue and different samples.

What sets *phytoGEM* apart?

- Easy sampling and long term storage solution
- No centrifugation
- Single-step extraction and purification
- Superior inhibitor removal
- Sample to PCR-ready DNA in 15 mins
- Closed-tube system prevents contamination.



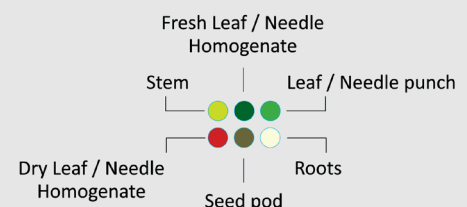
Validated plant species and tissues using the PDQeX phytoGEM system

Each plant species has its own unique difficulties for nucleic acid extraction. This can be due to a number of causes: sample toughness, different inhibitors, polyphenols or different polysaccharides. The *phytoGEM* system has been tested and optimised on a wide variety of species and tissue types. The table below shows where we are so far. This list is by no means comprehensive but is growing week by week.

If you have your own favourite plant and it is not on this list, contact us from our website. We should be able to help you.

		Rice (<i>Oryza sativa</i>)- 3 varieties			Peach (<i>Prunus persica</i>)
		Maize (<i>Zea mays</i>)- 2 varieties			Pear (<i>Pyrus sp.</i>)
		Wheat (<i>Triticum aestivum</i>)			Rhubarb (<i>Rheum rhabarbarum</i>)
		Potato (<i>Solanum tuberosum</i>)			Dock (<i>Rumex sp.</i>)
		Tomato (<i>Solanum lycopersicum</i>)			Monterey Pine (<i>Pinus radiata</i> spp.)
		Thale cress (<i>Arabidopsis thaliana</i>)			Japanese cedar (<i>Cryptomeria japonica</i>)
		Broccoli (<i>Brassica oleracea</i> var. <i>italica</i>)			Mediterranean cypress (<i>Cupressus sempervirens</i>)
		Spinach (<i>Spinacia oleracea</i>)			Redwood (<i>Sequoia sempervirens</i>)
		Lemon (<i>Citrus limon</i>)- 4 varieties			American sycamore (<i>Platanus occidentalis</i>)
		Orange (<i>Citrus reticulata</i>)- 2 varieties			Nikau palm (<i>Rhopalostylis sapida</i>)
		Grapevine (<i>Vitis</i> spp.)- 4 varieties			Bamboo palm (<i>Chrysalidocarpus lutescens</i>)
		Kiwifruit (<i>Actinidia</i> spp.)- 2 varieties			Chinese windmill palm (<i>Trachycarpus fortunei</i>)
		Blueberry (<i>Vaccinium</i> sp.)			Alexandra palm (<i>Archontophoenix alexandrae</i>)
		Apple (<i>Malus pumila</i>)			Fan palm (<i>Pritchardia gaudichaudii</i>)
		Plum (<i>Prunus domestica</i>)			
		Apricot (<i>Prunus armeniaca</i>)			

KEY



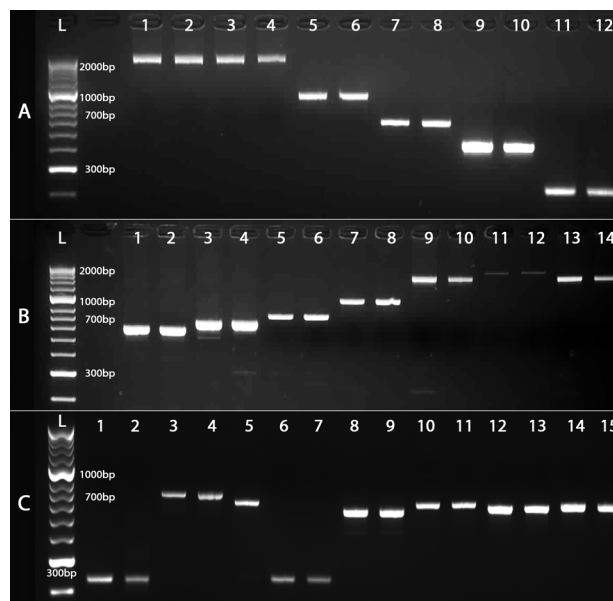
Plants and plant tissues in the phytoGEM repertoire (so far).

Typical results using *phytoGEM*

DNA yields will vary from species to species. Some example numbers are in the table below. These were obtained from three x 3 mm punches from the storage card.

Plant	Yield (ng in 100 µl)
<i>Arabidopsis thaliana</i>	150 – 218
<i>Oryza sativa</i>	103 – 299
<i>Citrus limon</i>	96 – 166
<i>Triticum aestivum</i>	110 – 132
<i>Solanum lycopersicum</i>	282 – 360
<i>Zea mays</i>	90 – 152
<i>Solanum tuberosum</i>	96 – 154
<i>Vitis</i> spp	144 – 256

The DNA is suitable for standard end point PCR...



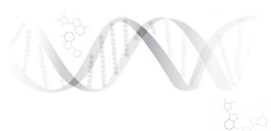
Agarose gel images showing amplicons generated from plant extractions. Fragments were generated using primer combinations targeting sequences in 5 genes: *adh* (alcohol dehydrogenase), *act2* (actin 2), *matK* (maturase K), *rbcL* (RuBisCO large subunit) and *psbA* (photosystem II) ranging from ~200–2300 bp.

(A) *Arabidopsis*: 1-4: *act2*, 5-6: *matK*, 7-8: *rbcL*, 9-10 *psbA*, 11-12: *adh*

(B) Rice: 1-2: *rbcL*, 3-4: *psb*, 5-14: various fragments of *adh*.

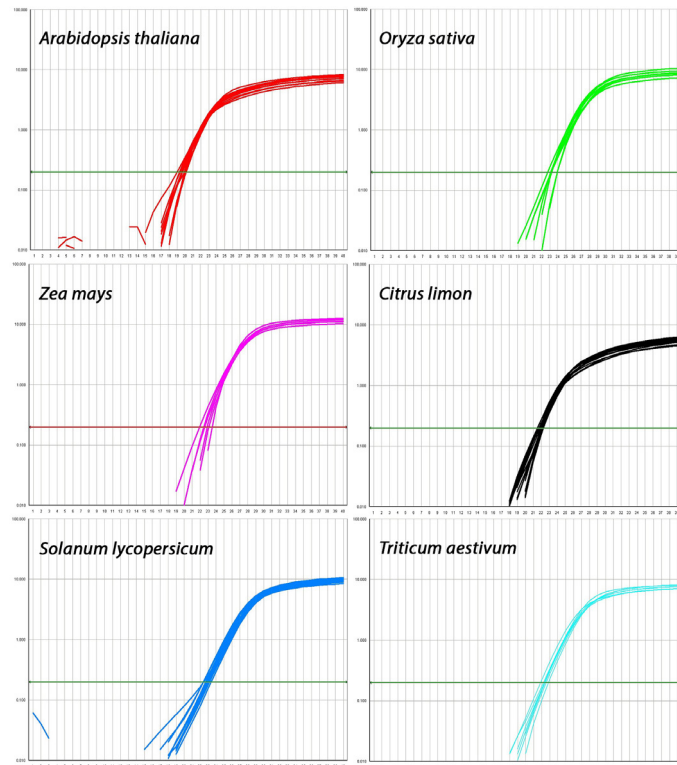
(C) 1-2: lemon *adh*, 3-4: lemon *rbcL*, 5: lemon *psb*, 6-7: maize *adh*,

8-9: maize *psb*, 10-11: maize *rbcL*, 12-13: tomato *psb*, 14-15: tomato *rbcL*.



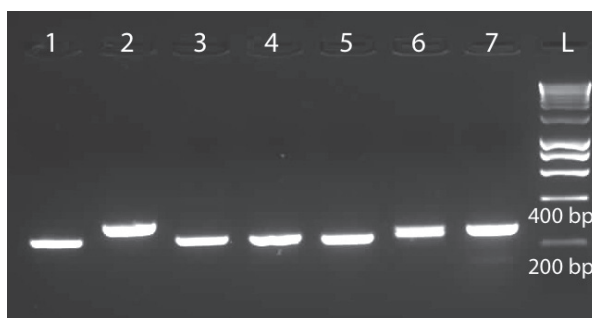
Typical results using phytoGEM

... and is also of excellent quality for qPCR

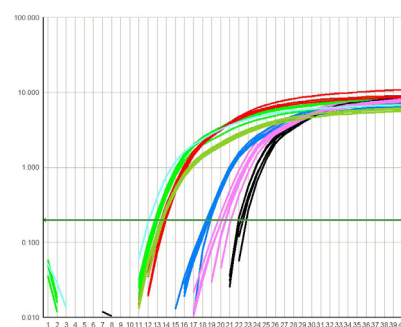


qPCR plots of alcohol dehydrogenase (*adh*) gene fragments amplified from phytoGEM extractions from 6 plants.

The phytoGEM system can also be used for extracting fungal DNA

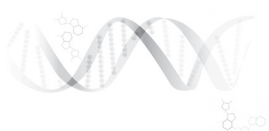


Agarose gel separation ITS amplicons from **1.** *Cladosporium* sp.; **2.** *Cadophora malorum*; **3.** *Geomyces* sp.; **4.** *Aspergillus* sp.; **5.** *Penicillium* sp.1; **6.** *Penicillium* sp. 2; **7.** *Penicillium* sp. 2.



qPCR plots of amplified ITS regions.

- Cladosporium* sp.;
- Cadophora malorum*;
- Geomyces* sp.;
- Aspergillus* sp. 2;
- Penicillium* sp. 1 3;
- Penicillium* sp. 2;
- Penicillium* sp. 2.



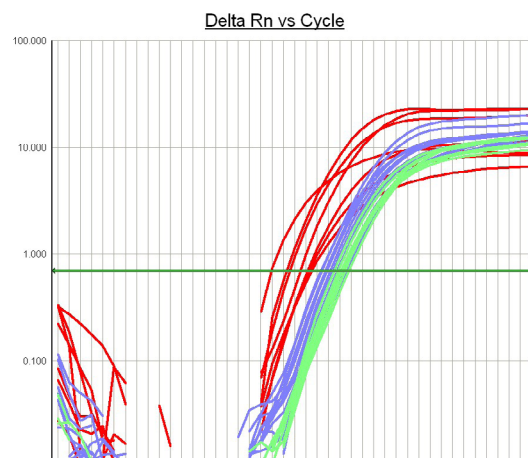
phytoGEM for detection of plant pathogens

Early detection of pathogens is vital for successful crop management. It allows proactive control measures to be applied well before the establishment and spread of a disease. The *phytoGEM* system offers a simple hands-off method for DNA extraction and is ideal for studying endophytic plant pathogens.

So far, *phytoGEM* has been validated for the detection of *Pseudomonas syringae* pv. *actinidiae* (Psa) detection in kiwifruit, *Phytophthora* spp. in kauri, pine and oak and *Phytoplasma* spp. in grapevines.



A kiwifruit leaf infected with *Pseudomonas syringae* pv. *actinidiae*.



qPCR plots showing the presence of *Psa*-V DNA in leaf infected under controlled conditions. Leaf samples directly on infected brown spots; leaf samples adjacent to spots; plots from crushes made on clear area of infected leaf.

phytoGEM for RNA?

We are almost there. We expect to have an RNA solution for *phytoGEM* in late 2018





Description	Product code
PDQeX Machine	XMC
PDQeX phytoGEM Machine (includes two sampling tools, a punch and samples of <i>phytoCards</i>)	XPC

Description	Quantity	Product code
<i>phytoGEM</i> Cartridges	100	PPP0100
	500	PPP0500
	1000	PPP1000
<i>phytoCards</i>	500	PCA0500
	1000	PCA1000
Sampling tool	1	PCR



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