

Simultaneously measure live and dead cells in the same well

A variety of assay chemistries are currently available for cell viability and cytotoxicity determination. Viability assays usually measure metabolic activity or ATP levels. Cytotoxicity assays usually assess cell membrane integrity. Although existing technologies provide useful data and are cost effective, they have limitations when it comes to combining assays in order to get more information from samples. For example, ATP assays, which are lytic endpoint assays, are incompatible with reporter gene assays or luminescent caspase assays.

The MultiTox-Fluor Assay allows the relative measurement of live and dead cells in culture wells. This assay gives ratiometric, inversely proportional values of viability and cytotoxicity that are useful for normalising data to cell number (Figure 1). In addition, MultiTox-Fluor is compatible with additional fluorescent and luminescent chemistries.

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- Another new application for Maxwell[®] 16: RNA purification
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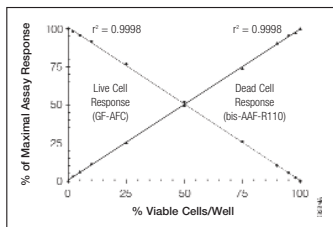


Figure 1. Viability and cytotoxicity measures are inversely correlated and ratiometric.

Assay Design

The MultiTox-Fluor Assay measures two distinct and independent protease activities as markers of cell viability and cytotoxicity (Figure 2). The live-cell protease activity is restricted to intact viable cells and is measured using a cell permeable, fluorogenic peptide substrate (Gly - Phe-AFC). The substrate enters intact cells, where it is cleaved to generate a fluorescent signal proportional to live cell number. This live-cell protease activity marker becomes inactive upon loss of membrane integrity and leakage into the surrounding culture medium. A second, cell impermeable, fluorogenic peptide substrate ([Ala-Ala-Phe]₂-R110) is used to measure dead-cell proteases that have been released from cells with compromised membranes.

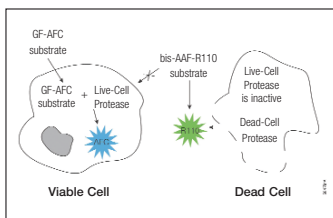


Figure 2. The MultiTox-Fluor Assay measures two distinct and independent protease activities.

Sensitivity

With the MultiTox-Fluor assay, as few as 10 live cells and 40 dead cells in a sample can be detected.

Correlation with conventional assays

Data from the MultiTox-Fluor assay correlates well with data obtained from enzyme release, resazurin reduction, ATP quantitation or dye exclusion assays.

Additional multiplexing possibilities

The MultiTox-Fluor Assay can be used with other Promega cell-based assays, such as those for measuring caspase activation or reporter gene expression (Figure 3).

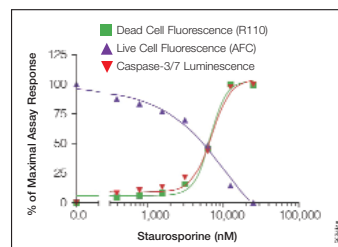


Figure 3. The MultiTox-Fluor Assay multiplexed with the Caspase-3/7 luminescence assay.

Conclusion

MultiTox-Fluor is a single reagent, homogeneous, fluorescent assay that simultaneously measures live and dead cells in culture wells. MultiTox allows data to be normalized, making results more comparable, well-to-well, plate-to-plate, day-to-day. It also enables more information to be obtained from samples, owing to its compatibility with additional assay chemistries.

1 For more information please request a MultiTox Information Pack using the Faxback form on page 4, or visit www.promega.com/uk/multitoxfluor

Total RNA purification now available for Maxwell® 16

The purification and analysis of RNA is one of the most important techniques used to monitor the expression of genetic information within cells.

Purified RNA is routinely used in applications such as quantitative Real Time PCR (qRT-PCR) or microarray analysis.

These applications require the isolation of high-quality RNA, which can be tedious and labour intensive.

The new Maxwell® 16 total RNA Purification Kit provides high yields of RNA while effectively removing contaminating genomic DNA (gDNA) from the RNA preparation in about 30 minutes of hands-free instrument operation.

Features

- **Enjoy confidence in your application results:** No detectable contaminating genomic DNA means fewer repeated experiments or unexplained variable results
- **Obtain application-ready RNA:** For most sample types, the eluted RNA concentration is ready for qRT-PCR. No post-purification concentration required
- **Achieve High Yield:** Processing is highly efficient, and sample capacity is higher than comparable systems

i For more information, or to request a demonstration, please visit www.MeetMaxwell.com

The Maxwell® 16 Integrated System combines instrumentation, optimised automated methods, pre-filled reagent cartridges, service and support to save time, enhance productivity and improve consistency in results.

Maxwell® 16 processes up to 16 samples in approximately 30 minutes. The purified nucleic acid is of high quality and at high yield and concentration, suitable for direct use in a variety of downstream applications.

The compact size ensures that it fits on every bench top.

Figure 4. A maximum instrument at a minimum size.



The new Total RNA Purification Kit expands the capabilities of the Maxwell® 16 purification system, giving hands free, automated purification of total RNA from virtually any sample type without detectable gDNA contamination.

The Maxwell® 16 Total RNA system purifies total RNA from a wide variety of sample types. For more detail please refer to **Table 1** below.

The presence of contaminating DNA in RNA samples leads to complications in downstream applications. Highly sensitive amplification based methods such as qRT-PCR rely on DNA-Free RNA since even minute levels of gDNA will lead to variability in your results.

The Maxwell® 16 Total RNA Purification Kit uses a novel method to remove gDNA. Our experiments showed that the Maxwell® 16 Total RNA Purification Kit results in a 100- to 1000-fold lower gDNA contamination than DNase removal based methods.

Ordering Information

Product	Size	Cat. No.	Price
Maxwell® 16 Total RNA Isolation Kit	48 preps	AS1050	£180

Also available:

- Maxwell® 16 Blood DNA Purification Kit
- Maxwell® 16 Cell DNA Purification Kit
- Maxwell® 16 Tissue DNA Purification Kit
- DNA IQ® Reference Sample Kit for Maxwell® 16

Table 1

	Amount processed	Typical Yield (µg)	Typical $A_{260}/280$
Mouse Liver	25-50 mg	100-233	2.12
Mouse Brain	50 mg	29	2.08
Mouse Heart	50 mg	23	2.11
Mouse Intestine	50 mg	128	2.09
Mouse Kidney	50 mg	83	2.09
Mouse Lung	50 mg	22	2.12
PAXgene® Blood sample	1 tube	5	1.8
HeLa Cells	5x10 cells	47	1.8
Tomato Leaf	100 mg	33	2.19

PCR Cloning Systems-Simplify and speed up achieving your results



Cloning is one of the most basic procedures in molecular biology, used to insert DNA sequences into vectors for further study.

Unfortunately, cloning is often a bottleneck, consuming precious time before you can start the more interesting part of your research project: Analysing your results.

It doesn't have to be that way.

Promega provides solutions without the hassle so you can get on with the interesting stuff.

Four steps to simple cloning

1 **Amplify** your gene/sequence of interest.

GoTaq® Green Master Mix-premixed and ready-to-use, offers the easiest way to reliably amplify your target sequence and give you a high yield.

2 **Purify** your PCR product either directly or from a gel slice in under 15 minutes using **Wizard® SV Gel and PCR Clean up System**.

Save time and achieve high yield and concentration. Purified DNA is suitable for automated fluorescent sequencing, cloning, labelling, restriction enzyme digestion or *in vitro* transcription/translation without further manipulation.

3 **Clone** your PCR product into the **pGEM®-T Easy Vector System**.

The multiple cloning site is flanked by restriction enzyme sites for *BstZ I*, *NotI* and *EcoRI* allowing you to remove the insert with a single digest.

Now you have your clone, you can start to study the functionality of your construct with the **Promega Flexi® Vector System**.

Once your protein-coding region is sub-cloned into a Flexi® Vector, you can easily shuttle it into many other Flexi® Vectors, to compare various expression systems and obtain your best yields and results.

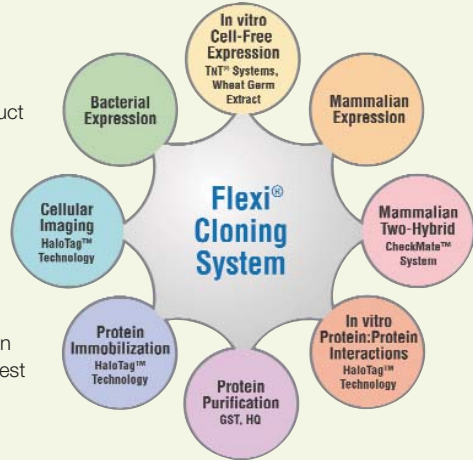


Figure 6. Functional protein analysis capabilities provided by the Flexi® Vectors.

Products	Size	Cat. No.	Price
GoTaq® Green Master Mix	100 rxn	M7112	£56
	1000 rxn	M7113	£387
Wizard® SV Gel and PCR Clean Up System	50 preps	A9281	£54
	250 preps	A9282	£244
pGEM®-T Easy Vector System II	20 rxn	A1360	£132
pGEM®-T Easy Vector System II-with High Efficiency JM109 Competent Cells	20 rxn	A3610	£211
JM109 Competent Cells 108cfu/µg	1ml	L2001	£88
Flexi® Vector System	For more information visit www.promega.com/uk/flexi		

4 **Transform** our **JM109 High Efficiency** competent cells with your construct.

Promega's **JM109 High Efficiency competent cells** reliably show transformation efficiencies higher than 10⁸cfu/ g.

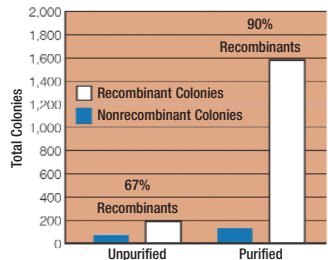


Figure 5. Purification of PCR products enhances cloning success.

Customer testimonial:

Cloning PCR fragments has never been easier and faster with the pGEM-T Easy Kit.

Victoria Workman,
Cardiff School of Biosciences

For more information please visit www.promega.com/uk/flexi for a complete overview of the range of Flexi® Vectors available.

For more information on any of the products mentioned above please fill in the faxback form found on page 4.

Promega Young Life Scientist Awards

Every year we make awards to some of the country's most promising young life scientists, working with the Biochemical Society, the Genetics Society and the British Society for Immunology and with the support of Science Magazine.

We are happy to announce that we have recently presented this year's awards in Genetics and



Bernhard Payer, Promega Young Geneticist 2006, Cancer Research UK Gurdon Institute.

Functional analysis of germ cell specification in mice.

Biochemistry. **Bernhard Payer** of Cancer Research UK Gurdon Institute won the Genetics award for his work on Functional analysis of germ cell specification in mice and **Jaclyn Long** was presented with the Biochemistry award for her work on Lipid phosphate phosphatase-1 regulation of p42/p44 MAPK activation and cell migration.



Jaclyn S Long, Promega Young Biochemist 2006, University of Strathclyde.

Lipid phosphate phosphatase-1 regulates lysophosphatidic acid - and platelet-derived growth factor-induced p42/p44 MAPK activation and cell migration. Presented at Biochemistry Annual Society Meeting, Bioscience 2006 in Glasgow in July.

My Team!

Are you part of a team or club linked to a department or organisation working in the life sciences? If so you can apply for one of Promega's 10 team sponsorships, worth £300 for each team.

Any sport or competitive activity is OK, from Tiddlywinks to Tennis and Rowing to Rugby. Sponsorship will be awarded annually at the start of the playing season for your sport and the most successful team will automatically receive sponsorship the following year.

i For more information and to apply online, please visit www.promega.com/uk/myteam



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- MultiTox cell viability systems
- Maxwell® 16 Purification instrument
- GoTaq® Green Master Mix
- Wizard® SV Gel and PCR clean up system
- pGEM®-T Easy Vector Systems
- Flexi®-Vectors

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