



Promega

Technical Bulletin

PowerPlex® 16 and PowerPlex® ES Monoplex Systems

INSTRUCTIONS FOR USE OF PRODUCTS DC6551, DC6561, DC6571,
DC6581, DC6591, DC6601, DC6611, DC6621, DC6631, DC6641, DC6651,
DC6661, DC6671, DC6681, DC6691, AND DC6751.



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PowerPlex® 16 and PowerPlex® ES Monoplex Systems

All technical literature is available on the Internet at: www.promega.com/protocols/
Please visit the web site to verify that you are using the most current version of this
Technical Bulletin. Please contact Promega Technical Services if you have questions on use
of this system. E-mail: genetic@promega.com

| | |
|---|---|
| 1. Description..... | 1 |
| 2. Product Components and Storage Conditions | 2 |
| 3. Amplification Protocols..... | 3 |
| 4. Detection Methods | 4 |
| A. Detection Using the ABI PRISM® 310, 3100 or 3100- <i>Avant</i> Genetic Analyzer and the Applied Biosystems 3130 or 3130 <i>xl</i> Genetic Analyzer ... | 4 |
| B. Detection Using the Hitachi FMBIO® II Fluorescence Imaging System. | 5 |

1. Description

The PowerPlex® 16 and PowerPlex® ES Monoplex Systems contain primer pairs that have the same sequence as those used in the PowerPlex® 16 (Cat.# DC6531), PowerPlex® 16 BIO (Cat.# DC6541) and PowerPlex® ES Systems (Cat.# DC6731). Refer to the *PowerPlex® 16 System Technical Manual #TMD012*, for locus-specific information, allele size range, allelic ladder components and genotypes of commonly used standard DNA templates. Refer to the *PowerPlex® ES System Technical Manual #TMD017* for SE33 locus-specific information. Technical Manuals are available upon request or at: www.promega.com/tbs/

The PowerPlex® 16 and PowerPlex® ES Monoplex Systems were developed for human identification applications including forensic analysis, relationship testing and research use.

2. Product Components and Storage Conditions

| Product | Size | Cat.# |
|---|---------------|--------|
| PowerPlex® 16 Monoplex System, Penta E (Fluorescein) ^(a,b,c) | 100 reactions | DC6591 |
| PowerPlex® 16 Monoplex System, Penta D (JOE) ^(a,b,c) | 100 reactions | DC6651 |
| PowerPlex® ES Monoplex System, SE33 (JOE) ^(a,b,d) | 100 reactions | DC6751 |

Not For Medical Diagnostic Use. Each system includes:

- 150µl Internal Lane Standard
- 300µl Gold ST★R Buffer
- 250µl 10X Primer Pair Mix
- 70µl Allelic Ladder

| Product* | Size | Cat.# |
|---|---------------|--------|
| PowerPlex® 16 Monoplex System, D3S1358 (Fluorescein) ^(a,b) | 100 reactions | DC6551 |
| PowerPlex® 16 Monoplex System, TH01 (Fluorescein) ^(a,b) | 100 reactions | DC6561 |
| PowerPlex® 16 Monoplex System, D21S11 (Fluorescein) ^(a,b) | 100 reactions | DC6571 |
| PowerPlex® 16 Monoplex System, D18S51 (Fluorescein) ^(a,b) | 100 reactions | DC6581 |
| PowerPlex® 16 Monoplex System, D5S818 (JOE) ^(a,b) | 100 reactions | DC6601 |
| PowerPlex® 16 Monoplex System, D13S317 (JOE) ^(a,b) | 100 reactions | DC6611 |
| PowerPlex® 16 Monoplex System, D7S820 (JOE) ^(a,b) | 100 reactions | DC6621 |
| PowerPlex® 16 Monoplex System, D16S539 (JOE) ^(a,b) | 100 reactions | DC6631 |
| PowerPlex® 16 Monoplex System, CSF1PO (JOE) ^(a,b) | 100 reactions | DC6641 |
| PowerPlex® 16 Monoplex System, vWA (TMR) ^(a,b) | 100 reactions | DC6661 |
| PowerPlex® 16 Monoplex System, D8S1179 (TMR) ^(a,b) | 100 reactions | DC6671 |
| PowerPlex® 16 Monoplex System, TPOX (TMR) ^(a,b) | 100 reactions | DC6681 |
| PowerPlex® 16 Monoplex System, FGA (TMR) ^(a,b) | 100 reactions | DC6691 |

Not For Medical Diagnostic Use. Each system includes:

- 150µl Internal Lane Standard
- 300µl Gold ST★R Buffer
- 250µl 10X Primer Pair Mix

Storage Conditions: Store all components at -20°C. The fluorescent 10X Primer Pair Mix is light-sensitive; therefore minimize light exposure and store in the dark.

*Items listed are available from Promega as “special order” items. A minimum order is not required, but items may not be immediately available for shipment. Contact Promega Customer Service for more information.

3. Amplification Protocols

Follow the protocols in the *PowerPlex® 16 System Technical Manual #TMD012* for the PowerPlex® 16 Monoplex Systems and in the *PowerPlex® ES System Technical Manual #TMD017* for the PowerPlex® ES Monoplex System. However, follow the directions below for the amount of AmpliTaq Gold® DNA polymerase to use per 25µl reaction.

1. Prepare the amplification mix as directed in Tables 1 and 2.
2. Add template DNA as directed in Tables 1 and 2. For optimal amplification results, we recommend 0.5–2ng of DNA per reaction.

Table 1. Amplification Mix for a Single Sample.

| PCR Component | Volume Per Sample |
|--|-------------------|
| Nuclease-Free Water | 17.4µl |
| Gold ST★R 10X Buffer | 2.5µl |
| 10X Primer Pair | 2.5µl |
| AmpliTaq Gold® DNA polymerase ¹ | 0.1µl (0.5 units) |
| Total amplification mix volume | 22.5µl |
| Template DNA ² (to be added) | 2.5µl |
| Total Reaction Volume | 25.0µl |

¹Assumes the AmpliTaq Gold® DNA polymerase is 5u/µl. If the enzyme concentration is different, adjust the volume of enzyme used accordingly.

²Assumes the template DNA volume is 2.5µl.

Table 2. Preparation of Amplification Mix for Multiple Samples.

| PCR Component | Volume Per Sample | × Number of Reactions | = Final Volume (µl) |
|---|-------------------|-----------------------|---------------------|
| Nuclease-Free Water | µl | | |
| Gold ST★R 10X Buffer | 2.5µl | | |
| 10X Primer Pair | 2.5µl | | |
| AmpliTaq Gold® DNA polymerase ¹ | 0.1µl (0.5 units) | | |
| Total amplification mix volume² | µl | | |
| Template DNA (to be added) | µl | | |
| Total Reaction Volume | 25.0µl | | |

¹Assumes the AmpliTaq Gold® DNA polymerase is 5u/µl. If the enzyme concentration is different, adjust the volume of enzyme used accordingly.

²The volume of template DNA plus the volume of amplification mix should equal 25.0µl.

4. Detection Methods

Note: Allelic ladders are only included with the PowerPlex® 16 Monoplex System, Penta E (Fluorescein), PowerPlex® 16 Monoplex System, Penta D (JOE) and PowerPlex® ES Monoplex System, SE33 (JOE). Allelic ladders for the other systems are available by custom order. Allelic ladder options are included in Tables 3 and 4.

4.A. Detection Using the ABI PRISM® 310, 3100 or 3100-*Avant* Genetic Analyzer and the Applied Biosystems 3130 or 3130*xl* Genetic Analyzer

Specific directions for detection using the ABI PRISM® 310, 3100 or 3100-*Avant* Genetic Analyzers and the Applied Biosystems 3130 or 3130*xl* are given in the *PowerPlex® 16 System Technical Manual #TMD012* and the *PowerPlex® ES System Technical Manual #TMD017*. Please follow these protocols when using the PowerPlex® Monoplex Systems. For additional questions, contact Promega Technical Services.

Table 3. Allelic Ladder Options for the ABI PRISM® 310, 3100 and 3100-*Avant* Genetic Analyzers and the Applied Biosystems 3130 or 3130*xl* Genetic Analyzer.

| PowerPlex® 16/ES Monoplex Loci | Allelic Ladder Options^{1,2} |
|--|---|
| D18S51, D21S11, TH01, D3S1358, FGA, D8S1179, vWA | PowerPlex® 16 or PowerPlex® ES |
| Penta E | Penta E or PowerPlex® 16 |
| Penta D | Penta D or PowerPlex® 16 |
| CSF1PO, D16S539, D7S820, D13S317, D5S818, TPOX | PowerPlex® 16 |
| SE33 | SE33 or PowerPlex® ES |

¹The PowerTyper™ Macros (Cat.# DG3470) can be used for data analysis.

²All allelic ladders are available by custom order.

4.B. Detection Using the Hitachi FMBIO® II Fluorescence Imaging System

For detection using the Hitachi FMBIO® II Fluorescence Imaging System, see the *PowerPlex® 16 BIO System Technical Manual #TMD016*. The filter set used for detection of the PowerPlex® 16 and PowerPlex® ES Monoplex loci is shown in Table 4.

Table 4. Allelic Ladder Options for the Hitachi FMBIO® II Fluorescence Imaging System. The filter set used for detection of each PowerPlex® 16 or PowerPlex® ES Monoplex locus is shown in bold in the “Filters Needed for Detection” Column.

| PowerPlex® 16/ES Monoplex Locus | Allelic Ladder Options¹ | Filters Needed for Detection (nm) | PowerPlex® Filter Set |
|--|---|--|------------------------------|
| D18S51, D21S11, TH01, D3S1358 | PowerPlex® 2.1 | 505, 585, 650 | 1.1/2.1 |
| | PowerPlex® 16 BIO ² | 505, 577, 598, 665 | 16 BIO |
| CSF1PO, D16S539, D7S820, D13S317, D5S818 | PowerPlex® 16 BIO ² | 505, 577, 598, 665 | 16 BIO |
| | JOE Ladder Mix | 585, 650 | 1.1/2.1 |
| | | 577, 665 | 16 BIO |
| FGA, TPOX, D8S1179, vWA | PowerPlex® 2.1 | 505, 585, 650 | 1.1/2.1 |
| | | 505, 577, 665 | 16 BIO ³ |
| SE33 | SE33 Allelic Ladder | 585, 650 | 1.1/2.1 |
| | | 577, 665 | 16 BIO |
| Penta E | PowerPlex® 2.1 | 505, 585, 650 | 1.1/2.1 |
| | PowerPlex® BIO | 505, 577 598, 665 | 16 BIO |
| | Penta E Allelic Ladder | 505, 650 | 1.1/2.1 |
| 505, 665 | | 16 BIO | |
| Penta D | PowerPlex® BIO | 505, 577 598, 665 | 16 BIO |
| | JOE Ladder Mix | 585, 650 | 1.1/2.1 |
| | | 577, 665 | 16 BIO |
| | Penta D Allelic Ladder | 585, 650 | 1.1/2.1 |
| | | 577, 665 | 16 BIO |

¹All allelic ladders are available by custom order.

²The Matrix 16 BIO is required for use with the PowerPlex® 16 BIO Allelic Ladder Mix. The Internal Lane Standard 600, labeled with CXR, can be detected with the 665nm filter.

³When using the PowerPlex® 16 BIO filter set for TMR-labeled loci, do not include the data from the 598nm filter in the FMBIO® Analysis Project or do not scan using the 598nm filter. Both TMR and CXR will be detected with the 598nm filter making color separation impossible. The TMR-labeled loci will be detected in the 577nm scan (JOE channel).

^(a)The purchase of this product does not convey a license to use AmpliTaq Gold® DNA polymerase. You should purchase AmpliTaq Gold® DNA polymerase licensed for the forensic and human identity field directly from your authorized enzyme supplier.

^(b)STR loci are the subject of U.S. Pat. No. RE 37,984, German Pat. No. DE 38 34 636 C2 and other patents issued to the Max-Planck-Gesellschaft zur Förderung der Wissenschaften, e.V., Germany. The development and use of STR loci are covered by U.S. Pat. No. 5,364,759, Australian Pat. No. 670231 and other pending patents assigned to Baylor College of Medicine, Houston, Texas.

^(c)U.S. Pat. No. 6,238,863, Chinese Pat. No. ZL99802696.4, European Pat. No. 1058727, Japanese Pat. No. 4494630 and other patents pending.

^(d)Use of the SE33 locus is licensed under U.S. Pat. Nos. 5,468,610 and 5,721,100.

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