

Reliable, fast assays to

measure

your proteins and peptides

New Thermo Scientific™ Pierce Protein Assays

Detergent Compatible Bradford Assay Kit • Quantitative
Colorimetric and Fluorometric Peptide Assays



Thermo Scientific Pierce Detergent Compatible Bradford Assay Kit

The Thermo Scientific™ Pierce Detergent Compatible Bradford Assay Kit is a quick and ready-to-use modification of the well-known Bradford coomassie-binding, colorimetric method for total protein quantitation. The Bradford Reagent is compatible with many reducing agents, but it is incompatible with most detergents. Proprietary additives to the Bradford Reagent make it compatible with up to 1% or higher of detergents and lysis reagents that are commonly used in life science research, including Triton™ X-100 and NP-40 detergents. Similar to the Bradford method, coomassie dye binds protein in an acidic medium causing an immediate shift in absorption maximum from 465nm to 595nm with a concomitant color change from green to blue. In addition, the assay is complete in just 10 minutes.

Now, there's no need to switch your Bradford assay when your samples contain detergent. Use our Pierce Detergent Compatible Bradford Assay for protein samples containing reducing agents and detergents!



Table 1. The Thermo Scientific Pierce Detergent Compatible Bradford Assay provides advantages over the Bio-Rad DC™ Protein Assay.

Feature	Pierce Detergent Compatible Bradford Assay Kit	Bio-Rad DC Protein Assay Kit
Assay measurement (absorbance maximum)	595nm	750nm
Detergent-free standards	Yes	No
Test tube assay sample volume	50µL	100µL
Microplate assay sample volume	10µL	5µL
Assay working range	100-1,500µg/mL	200-1,500µg/mL
Absorbance range (sensitivity)	High**	Low
Number of reagents in kit	1 Reagent	3 Reagents
Set-up time	10 minutes	30 minutes
Incubation time	10 minutes	15 minutes
Total time	20 minutes	45 minutes

**The Pierce Detergent Compatible Assay absorbance range is four times broader than the range for the DC Protein Assay

Highlights:

- **Flexible** – compatible with both detergent-free samples and detergent-containing samples
- **Convenient** – detergent-free standard curve; standards can be used directly without preparing them in the same detergent found in the test samples
- **Minimal sample** – requires only 10µL for microplate procedure
- **Easy to use** – single reagent; no working reagent preparation required
- **Fast** – 10 minute incubation at room temperature
- **Broad range** – detects protein concentration in the ranges of 2 to 1500µg/mL

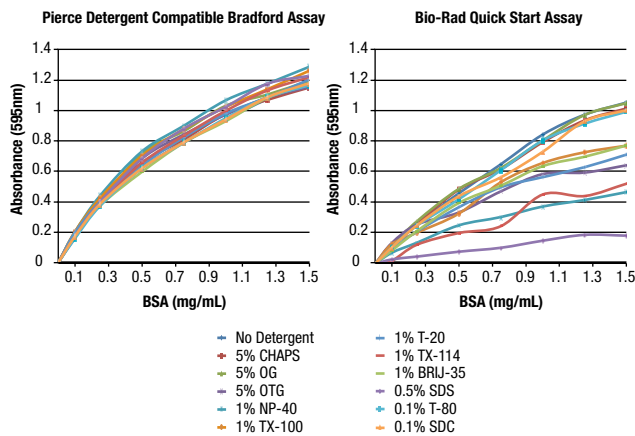


Figure 1. Superior detergent compatibility with the Thermo Scientific Pierce Detergent Compatible Bradford Protein Assay versus the Bio-Rad Quick Start Bradford Protein Assay. Assays were performed according to the microplate procedure using BSA standards spiked with detergent or water (control). Sample and reagent volumes used were according to the manufacturer's instructions. Each assay was incubated for 10 minutes and absorbance was measured at 595nm. The Pierce Detergent Compatible Bradford Assay is compatible with 11 commonly used detergents. Compatibility is defined as $\leq 10\%$ difference in absorbance between samples with and without detergent. In contrast, the Bio-Rad Quick Start™ Bradford Protein Assay is compatible with only three of the 11 detergents tested.

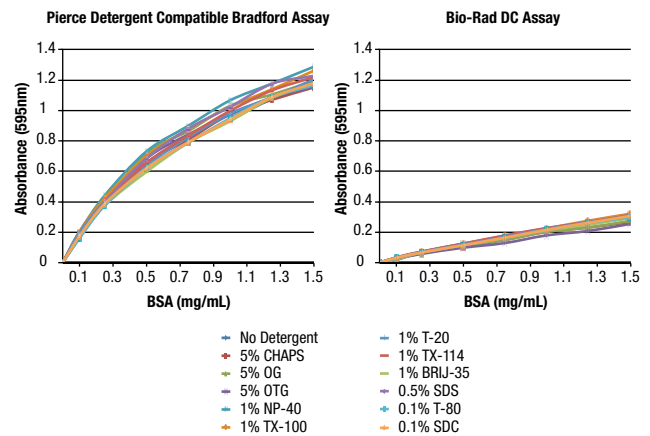


Figure 2. Better sensitivity with the Thermo Scientific Pierce Detergent Compatible Bradford Protein Assay versus the Bio-Rad DC Protein Assay. Each assay was performed in a microplate using BSA standards spiked with detergent or water (control), and followed the manufacturer's instructions. The range of the standard curve for the Pierce Detergent Compatible Bradford Assay is four times broader than the range for the Bio-Rad DC Assay.

Thermo Scientific Pierce Quantitative Peptide Assay Kits

The Thermo Scientific™ Pierce™ Peptide Assay Kits include easy-to-use, robust colorimetric and fluorescent quantitative assays specifically optimized for the measurement of peptides and peptide mixtures.

Our **Quantitative Colorimetric Peptide Assay** is a colorimetric microplate assay designed specifically for the quantitation of peptide mixtures. Our **Quantitative Fluorometric Peptide Assay** is the most sensitive peptide assay and is suitable for individual peptides and peptide mixtures. The colorimetric and fluorometric assays provide reliable and reproducible peptide quantitation results for discovery and targeted proteomic workflows where sample normalization is necessary. The increased sensitivity, low sample assay volume and included reference standards are essential for accurate and robust measurement of peptide digest samples, especially for mass spectrometry (MS) applications.

The **Pierce Quantitative Colorimetric Peptide Assay** is a modified BCA assay with a proprietary chelator optimized for the quantitation of peptide mixtures. In this reaction, the copper is first reduced by the amide backbone of peptides under alkaline conditions (Biuret reaction), followed by the proprietary chelator coupling with the reduced copper to form a bright red complex with absorbance at 480nm. The signal produced from this reaction is three- to four-fold more sensitive than the micro BCA protein assay for peptide analysis. This colorimetric peptide assay requires a small amount (20µL) of sample with a working peptide concentration range of 25-1000µg/mL and is best suited for peptide mixtures and complex digests (labeled and unlabeled).

The **Pierce Quantitative Fluorometric Peptide Assay** kit is designed for the sensitive quantitative measurement of peptide concentrations. In this assay, peptides are specifically labeled at the amino-terminus using an amine-reactive fluorescent reagent, and the fluorescently labeled peptides are detected using ex390/em475. This sensitive assay only requires 10µL of sample, produces a linear response with increasing peptide concentrations (5-1000µg/mL), results in a stable final fluorescence that can be detected in as few as five minutes and is suitable for the quantitative measurement of synthetic peptides as well as peptide digest mixtures.



Highlights:

- **Sensitive** – accurately detect as few as 5µg/mL of peptide mixtures using the fluorometric assay and 25µg/mL of peptide mixtures using the colorimetric format
- **Robust** – assay performance rigorously tested using peptide digest mixtures
- **Robust peptide standard** – kit includes a validated peptide digest standard for improved reproducibility of quantitation
- **Compatible** – compatible with many reagents, including MS sample preparation reagents (Table 1)
- **Convenient** – easy mix-and read-format; colorimetric signal is stable and may be read in as few as 15 minutes and fluorescent signal is stable and may be read in as few as 5 minutes up to several hours

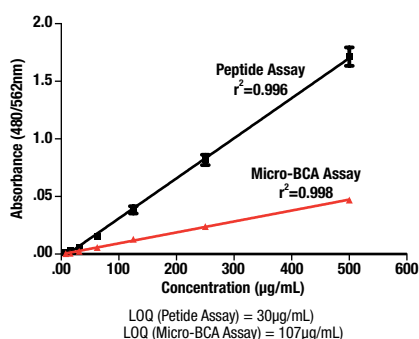


Figure 1. Sensitivity of Thermo Scientific Pierce Quantitative Colorimetric Peptide Assay. A BSA tryptic digest was serially diluted from 500µg/mL to 15µg/mL to compare assay sensitivities. Each sample was assayed in triplicate and error bars represent standard deviation. Limit of quantitation (LOQ) for the assay was determined by the following equation: $LOQ = 10 \times St. Dev of Blank + Average of Blank$.

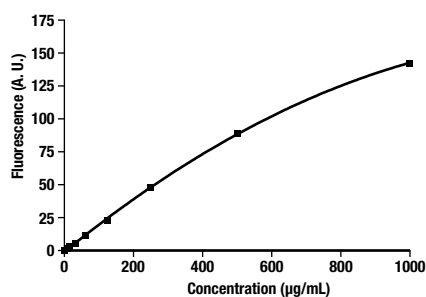


Figure 2. Standard curve for Thermo Scientific Pierce Fluorometric Peptide Assay using the Peptide Digest Assay Standard.

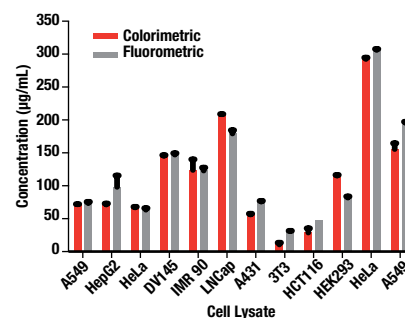


Figure 3. Quantitation comparison between colorimetric and fluorometric peptide assays. Tryptic peptide digests were prepared from 12 cell lines. Peptide digest concentrations were determined using both assay kits according to instructions. Each sample was assayed in triplicate, and the concentration of each digest was calculated with standard curve generated using the Protein Digest Assay Standard.

Table 1. Compatible substance concentrations in the Thermo Scientific Pierce Quantitative Colorimetric Peptide Assay.

Substance	Compatible Concentration
Acetone	50%
Acetonitrile	50%
Ammonium bicarbonate	50mM
DMSO	50%
DTT (dithiothreitol)	Not compatible
EDTA	5mM
Formic acid	0.5%
Guanidine	0.25M
Iodoacetamide	1M
Methanol	25%
SDS	1%
Sodium azide	1%
TCEP	Not compatible
TEA acetate	5mM
TEA bicarbonate	5mM
Trifluoroacetic acid	0.5%
Tris	100mM
Urea	1M

Table 2. Compatible substance concentrations in the Thermo Scientific Pierce Quantitative Fluorometric Peptide Assay.

Substance	Compatible Concentration
Acetone	25%
Acetonitrile	50%
Ammonium acetate	100mM
Ammonium bicarbonate	50mM
DMSO	50%
DTT (dithiothreitol)	10mM
EDTA	25mM
Formic acid	0.1%
Guanidine	1M
Iodoacetamide	100mM
Methanol	25%
SDS	1%
Sodium azide	1%
TCEP	10mM
TEA acetate	100mM
TEA bicarbonate	100mM
Trifluoroacetic acid	0.2%
Urea	1M

Note: It is possible to have a substance additive effect even though a single component may be present at a concentration below its listed compatibility. A sample buffer containing a combination of substances could potentially interfere with the assay. Combinations of reagents have not been examined in detail.

Ordering Information

Product #	Description	Pkg. Size
PI23246	Pierce Detergent Compatible Bradford Assay Kit <i>Sufficient for 300 test tube assays or 1500 microplate assays</i>	Kit
	Detergent Compatible Bradford Assay Reagent containing coomassie G-250 dye, methanol, phosphoric acid and solubilizing agents in water	450mL
	Albumin Standard Ampules, 2mg/mL	10 x 1mL
PI23246S	Pierce Detergent Compatible Bradford Assay Reagent <i>Sufficient for approximately 15 test tube assays or 80 microplate assays</i> Note: Standards are not included with this package.	25mL

Product #	Description	Pkg. Size
PI23275	Pierce Quantitative Colorimetric Peptide Assay <i>Sufficient for 500 assays</i>	Kit
	Colorimetric Peptide Assay Reagent A	50mL
	Colorimetric Peptide Assay Reagent B	2 x 25mL
	Colorimetric Peptide Assay Reagent C	2mL
	Peptide Digest Assay Standard (1mg/mL)	1.5mL
PI23290	Pierce Quantitative Fluorometric Peptide Assay <i>Sufficient for 500 assays</i>	Kit
	Fluorometric Peptide Assay Reagent	4 x 2.5mL
	Fluorometric Peptide Assay Buffer	50mL
	Peptide Digest Assay Standard (1mg/mL)	1.5 mL
PI23295	Peptide Digest Assay Standard	1.5mL

For information on the Thermo Scientific™ Total Protein Assay Reagent and Kit portfolio, visit thermoscientific.com/pierce and search on protein assays.

660 nm Protein Assay • BCA Protein Assay • BCA Protein Assay – Reducing Agent Compatible • Bovine Gamma Globulin (BGG) Standards • Bovine Serum Albumin (BSA) Standards • Compat-Able Protein Assay Kits • Coomassie (Bradford) Protein Assay • Coomassie Plus (Bradford) Protein Assay • Micro BCA Protein Assay • Modified Lowry Protein Assay

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