Collagen exists as many forms (27 types in vertebrates with 42 distinctive polypeptide chains). It is difficult to purify and to analyze, partly due to the extensive network that is formed by collagen molecules via different types of crosslinking which makes the collagen molecules insoluble.

Analysis of collagen is classically performed by different assays:
- ELISA for specific types of collagen, or specific pro-domains of collagen
- Sirius Red based assays for soluble collagen
- Western blotting using protein stain (for parafin-embedded tissues) or specific collagen antibodies
- Tissue hydrolysis followed by analysis of Hydroxyproline residues (either by a colorimetric method or by HPLC)

Following is a complete set of assays for quantitation of collagen and total proteins. It provides fast and easy solutions, especially for collagen and proteins in fresh, frozen or formalin-fixed paraffin-embedded tissues, without the need for HPLC or other special equipment.

**QuickZyme Soluble collagen assay**
This assay recognizes soluble or (acid/pepsin) solubilized collagen. The assay is colorimetric, has a 96-well plate format and is based on precipitation of collagen with Sirius-Red. This dye can bind the side-chain groups of basic amino acid residues. The dye is released from the precipitated complex at high pH followed by colorimetric detection. Sirius Red based assays recognize collagen, but also some other matrix related proteins may co-precipitate.

*Application*: The assay is used for the measurement of (soluble) collagen in e.g. cell culture media, and (acid or acid/pepsin) solubilized collagens e.g. from cellular extracts.

**QuickZyme Total collagen assay**
This assay recognizes all types of collagen irrespective of its form (mature, immature, procollagen, degraded collagen, cross-linked collagen, collagen from various sources). The assay is colorimetric, has a 96-well plate format, and is based on the quantification of hydroxyproline, an amino acid exclusively occurring in collagen. Hydroxyproline is released from collagen upon acid hydrolysis of the collagen containing sample. Hydrolysis is carried out at 95 OC, and the product can directly be used for hydroxyproline analysis, without further washing or drying steps. The analysis is based on established Chloramine T/DMBA methodology.

*Application*: The assay is used for the measurement of total collagen. Since the first step is complete hydrolysis of the sample, difficulty in extraction of collagen plays no role. The assay is applicable for all types of samples, including tissue.

**QuickZyme Hydroxyproline assay**
This assay is similar to the total collagen assay, with the difference that no protocols and materials are included for collagen hydrolysis and instead of a collagen standard a hydroxyproline standard is provided.

*Application*: This assay has the same application area as the total collagen assay, but is intended for customers who have their own hydrolysis method, or have a collection of hydrolyzed samples to be analyzed.

**QuickZyme Protein assay**
This assay measures free amino acids resulting from hydrolysis of protein. The assay is colorimetric and can directly be performed on acid hydrolyzates.

*Application*: This assay is intended to measure total protein in acid hydrolyzates and provides normalization of the Total collagen and Hydroxyproline values when weight based normalization is not possible, inconvenient or too variable.
**Total Protein Assay**

A robust protein assay measuring free amino acids resulting from hydrolysis of proteins

The QuickZyme total protein assay is used for the quantitative determination of all types of protein in hydrolyzed samples. The assay is based on the formation of a blue colored product from genipin with free amino acids. It measures the total amount of amino acids present in the hydrolyzed sample, with exception of proline and hydroxyproline. The amount of amino acids is a good measure for the amount of protein.

**Assay specifications**

- Quantitative measurement of protein in acid hydrolyzates
- Species independent
- Colorimetric read out at 570 nm
- Broad dynamic range: 0.05 to 3.0 mg/ml. Sensitivity: 0.007 mg/ml
- This kit does not require special equipment
- Ideally suited to normalize collagen analysis in hydrolyzed samples

<table>
<thead>
<tr>
<th>QuickZyme Total Protein Assay Kit</th>
<th>One 96 well plate kit</th>
<th>QZBtotprot1</th>
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<tbody>
<tr>
<td></td>
<td>Two 96 well plate kits</td>
<td>QZBtotprot2</td>
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<tr>
<td></td>
<td>Five plate bulk kit</td>
<td>QZBtotprot5</td>
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Store at RT. [Technical sheet](#), [Price](#).

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**Total Collagen Assay**

Now collagen can be determined by any biochemical laboratory!

The total collagen assay kit is based on the quantitative colorimetric determination of hydroxyproline residues, obtained by acid hydrolysis of collagen. Usually for this analysis special equipment is needed (speedvac with special modifications) for removal of the HCl. The QuickZyme total collagen assay does not need this laborious step, resulting in a fast and simple assay (< 2 hrs) following the hydrolysis step. This assay makes collagen determination available for every lab.

**Assay specifications**

- Quantitative measurement of all types of collagen, species independent.
- Samples: tissue culture supernatants, cellular extracts, tissue homogenates, tissues.
- Colorimetric measurement of hydroxyproline content
- Range: 6 to 300 μg/ml. Sensitivity: 2.9 μg/ml
- Uses rat tail collagen as standard
- Ease-of-use: Equivalent to ELISA

<table>
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<td>Five plate bulk kit</td>
<td>QZBtotcol5</td>
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</tbody>
</table>

Store at RT. [Technical sheet](#), [Price](#).
Hydroxyproline Assay

A robust protein assay measuring hydroxyproline resulting from hydrolysis of proteins

The QuickZyme hydroxyproline assay is used for the quantitative determination of hydroxyproline. Hydroxyproline results from (acid) hydrolysis of collagen. Usually for this analysis special equipment is required (speedvac with special modifications) for removal of the HCl. The QuickZyme hydroxyproline assay kit does not need this laborious step. It has a colorimetric read-out and a fast and simple assay format (< 2 hrs).

Hydroxyproline Assay specifications

- Quantitative measurement of hydroxyproline.
- Samples: acid hydrolized tissue culture supernatants, cellular extracts, tissue homogenates, tissues.
- Colorimetric measurement
- Range: 6 to 300 μM. Sensitivity: 2.4 μM
- Uses hydroxyproline as standard
- Ease-of-use: Equivalent to ELISA

QuickZyme Hydroxyproline Assay Kit

| One 96 well plate kit | QZBhypro1 |
| Two 96 well plate kits | QZBhypro2 |
| Five plate bulk kit | QZBhypro5 |

Store at RT. Technical sheet, Price.

This kit does not contain tubes and a protocol for collagen hydrolysis. If you need these items please order the total collagen assay

Soluble Collagen Assay

Assay specifications

- Measures all types of fibrillar collagen, species independent.
- Samples: tissue culture supernatants, cellular extracts and tissue homogenates.
- Quantitative. Range/sensitivity: 0.5 to 10 μg/ml. Sensitivity: 0.69 ng/ml.
- Ease-of-use: Equivalent to ELISA.
- <3 h protocol

QuickZyme Soluble Collagen Assay Kit

| One 96 well plate kit | QZBcol1-1 |
| Two kits | QZBcol1-2 |
| Five kits | QZBcol1-5 |

Store at +4°C. Technical sheet, Price.

Related products/documents

- Back to all Protein Assays page, i.e. :

  BC proteins assay 40840A, 1Kit (up to 4000 tests)
  Based on bicinchoninic acid (modified Lowry method). For very reproducible and accurate results, thanks to highly linear and large dynamic range, sensitive (down 10 and 0.5 μg/ml) and low protein/protein variations, large chemical tolerance.

  Coo protein Assay UPF86400A, 1Kit (up to 4000 tests)
  A modified version of the Bradford assay.

  Products Highlights Overview
  Biosciences innovation catalog (on line)
Information inquire

Reply by Fax: +33 (0) 4 70 03 82 60 or email at interbiotech@interchim.com

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