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Product datasheet

Hydroxyproline Assay Kit (Colorimetric) ab222941

4 Abreviews 26 References 2 Images

Overview

Product name	Hydroxyproline Assay Kit (Colorimetric)
Detection method	Colorimetric
Sample type	Urine, Serum, Other biological fluids, Tissue Homogenate
Assay type	Quantitative
Range	0.1 µg - 1 µg
Assay time	3h 00m
Species reactivity	**Reacts with:** Mammals, Other species
Product overview	Hydroxyproline Assay Kit (Colorimetric) ab222941 provides a quick and convenient method to quantify hydroxyproline in tissue lysates and biological fluids such as urine and serum. The classical hydroxyproline assay protocol is based on the oxidation of hydroxyproline to a pyrrole intermediate followed by reaction with Ehrlich's reagent dissolved in concentrated perchloric acid. Perchloric acid is a hazardous material that is both toxic and highly reactive, requiring special handling and waste-disposal protocols. This hydroxyproline assay protocol employs a proprietary acidic developer solution to accurately measure hydroxyproline in hydrolysates without the use of hazardous perchlorates. It is a quick and convenient protocol where hydroxyproline gets oxidized to form a reaction intermediate, which further in reaction forms brightly-colored chromophore that can be easily detected at OD 560 nm. The assay can detect as low as 0.05 µg hydroxyproline/well.
Notes	This product is a replacement for Hydroxyproline Assay Kit K555; this kit does not contain the hazardous Perchloric acid which is present in K555. The components in this product are exactly the same as in K555, except that the Perchloric acid/Isopropanol Solution component in K555 has been replaced with the Developer component. If you would prefer to continue to use a kit in the format of K555, then use this product and: 1) a) purchase 70% Perchloric Acid (**ab291263**) and Isopropanol (**ab291264**), or b) separately purchase 70% Perchloric Acid and 99% Isopropanol through another supplier. For safety reasons, Perchloric Acid will not be shipped by Abcam in the same shipment as either Isopropanol or this product.

2) In a fume cupboard, mix 1 ml of 70% Perchloric Acid and 5 ml of Isopropanol to produce the Perchloric acid/Isopropanol Solution component previously provided in K555.

3) Then follow the protocol for K555 available on **www.biovision.com/documentation/datasheets/K555.pdf**, using the components from this product, and the Perchloric acid/Isopropanol Solution that you produced.

This product is manufactured by BioVision, an Abcam company and was previously called K226 Hydroxyproline Assay Kit (Perchlorate-Free). K226-100 is the same size as the 100 test size of ab222941.

Hydroxyproline assay protocol summary:
- add 10 N concentrated NaOH to samples and hydrolyze at 120ºC for 1 hr
- cool on ice
- neutralize with 10 N concentrated HCl, centrifuge and collect supernatant
- add samples and standards to wells
- evaporate wells to dryness by heating at 65ºC
- add oxidation reagent mix to dissolve crystalline residue, and incubate at room temp for 20 min
- add developer and incubate at 37ºC for 5 min
- add DMAB concentrate and incubate for 45 min at 65ºC
- analyze with microplate reader

We are currently not able to offer K555 Hydroxyproline Colorimetric Assay Kit from BioVision and apologize for the inconvenience that this may cause. We recommend ab222941 as an alternative.

Platform Microplate reader

Properties

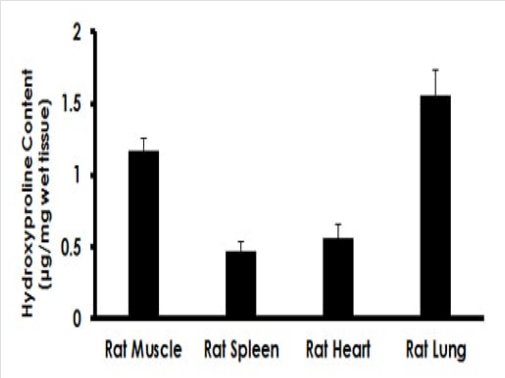
Storage instructions Store at -20°C. Please refer to protocols.

Components	100 tests
Chloramine T Concentrate	1 x 600µl
Developer Solution I	1 x 5ml
DMAB Concentrate	1 x 5ml
Hydroxyproline Standard	1 x 100µl
Oxidation Buffer	1 x 10ml
AlumaSeal® Film	1 unit

Relevance Hydroxyproline, a non-essential amino acid derived from proline, with no known therapeutic use. Hydroxproline is used as a major component of structural protiens such as collagen, connective tissues, plant cell walls, tendons and ligaments and provides skin elasticity. Vitiman C is required for the conversion process from proline to hydroxyproline, a deficincy in vitiman C can lead to defects in collagen synthesis, thus, resulting in easy bruising, internal bleeding, breakdown of connective tissue of the ligaments and tendons, and increased risk to blood vessel damage. An unusual feature of this amino acid is that, it is not incorporated into collagen during biosynthesis at the ribosomal level, but is formed from proline by a posttranslational modification by an enzymatic

hydroxylation reaction.

Images



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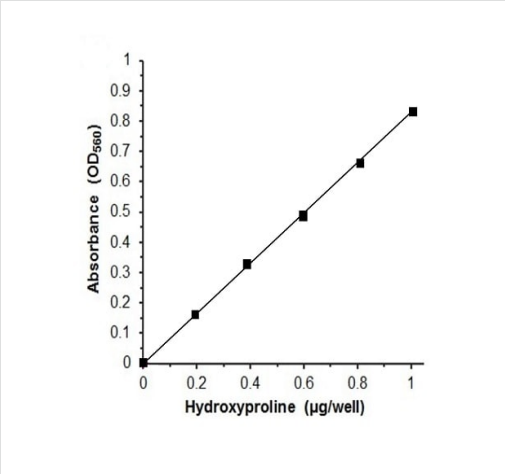

Typical Hydroxyproline standard calibration curve.

Hydroxyproline Assay Kit (Colorimetric) (ab222941)

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