

Quantitative determination of high molecular weight  $\beta$ -glucan from barley in malt-mash and wort  
 Art. No. E3500 test combination for 40 determinations  
 Art. No. E3550 test combination for 160 determinations

For *in vitro* use only  
 Store between 17 - 30 °C

**Approval:** EBC 4.16.3, MEBAK 3.1.4.9.2, ASBC

**1. Test principle**

Colorimetric assay for the quantitative determination of high molecular weight  $\beta$ -glucan from barley in malt-mash and wort.

The test has been recognized under EBC 4.16.3 (high molecular weight  $\beta$ -glucan content of malt wort: spectrophotometric method), MEBAK 3.1.4.9.2 (high molecular  $\beta$ -Glucan colorimetric method) and ASBC ( $\beta$ -glucan in wort – spectrophotometric method).

**2. Reagents**

**2.1. Content & composition**

The test is suitable for manual processing. With manual processing, the reagents are sufficient for 40 or 160 determinations.

**E3500** test combination for 40 determination:

- Solution A: 1 x 125 mL
- Calibrator set: 6 x 1.0 mL (0, 100, 200, 300, 400, 500 mg  $\beta$ -glucan/L)

**E3550** test combination for 160 determination:

- Solution A: 4 x 125 mL
- Calibrator set: 6 x 1.0 mL (0, 100, 200, 300, 400, 500 mg  $\beta$ -glucan/L)

**2.2. Reagent preparation**

The reagents are ready-to-use and be allowed to reach room temperature (17 - 30 °C) before use. Do not interchange components between kits of different batches.

**2.3. Storage & stability**

The reagents are stable until the expiration date printed on the label (see label) even after opening at 2 - 8 °C if handled properly. Do not freeze reagents.

**2.4. Safety & disposal**

The general safety rules for working in chemical laboratories should be applied. Do not swallow! Avoid contact with skin and mucous membranes.

This kit may contain hazardous substances. For hazard notes on the contained substances, please refer to the appropriate safety data sheets (SDS) for this product. After use, the reagents can be disposed of with the laboratory waste. Packaging materials may be recycled.

**3. Sample preparation**

- The samples should be brought to room temperature before measurement.
- The method for sample preparation has been described under EBC 4.5.1 (extract of malt: congress mash). It is mandatory to follow it if the present  $\beta$ -glucan kit should be used according to EBC 4.16.3.
- For the determination of the whole content of  $\beta$ -glucans (soluble and insoluble parts), it is recommended to heat the samples for 15 min at 80 °C to solubilize gel-formed fractions of  $\beta$ -glucans; after cooling down to room temperature, a centrifugation or filtration step might be necessary before testing the sample.

**4. Assays performance**

Wavelength: 550 nm ( $\pm$  5 nm)  
 Light path: 1.00 cm (glass; plastic)  
 Temperature: 18 - 30 °C (temperature dependent calibration necessary)  
 Method: end-point measurement  
 Incubation time: 30 minutes  
 Measurement: against air (without cuvette) or water  
 Measuring range: 100 - 500 mg/L  $\beta$ -glucan

Pipette into cuvette*:	Calibrator (1 cuvette each)	Sample
Calibrator set (1 - 6)	0.200 mL	-
Sample	-	0.200 mL
Solution A	3.000 mL	3.000 mL
Mix, incubate for 30 min at room temperature. Measure absorption ( $A_1$ ) at 550 nm.		

\* Before pipetting a sample or calibrator into the cuvette, first rinse the pipette tip once or twice with the calibrator / sample.

**5. Calculation of results**

**Calculation of sample solutions – total concentration of  $\beta$ -glucan**

Plot the calibration curve using the absorbance of the calibrators 1 to 6. The  $\beta$ -glucan content of the samples can be directly read from the curve or calculated from the resulting equation (linear regression). An excel sheet is available on request.

Example with typical absorbance values:

Calibrator	$\beta$ -glucan (mg/L)	$A_1$	minus blank
1	0	0.850	0.000
2	100	0.973	0.123
3	200	1.081	0.231
4	300	1.184	0.334
5	400	1.280	0.430
6	500	1.377	0.527

The specific values of each lot can be seen in the corresponding certificate of analysis.

**6. Performance data**

**Linearity, measuring range & sensitivity**

Linearity is given up to 500 mg/L  $\beta$ -glucan. The limit of detection (LoD) and limit of quantification (LoQ) were determined according to method DIN 32645:2008-11, using buffered aqueous solutions. This results in an LoD of 7 mg/L and an LoQ of 19 mg/L.

**7. Supporting documents**

On request, we offer the following document:

- Enzytec™ Liquid Excel template for result calculation

Safety data sheets (SDS) und certificates of analysis (CoA) are available in digital form under the following link <https://eifu.r-biopharm.com/>



**8. Services & technical support**

On request, we offer the following services:

- Customized troubleshooting
- Data & results analysis
- Customer workshops & webinars
- Automation: application support and technical service

**9. Disclaimer**

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