

Range:  $\geq 2$  (BU) Bitter units / Accuracy:  $\pm 0.5$  BU

LCK241

Scope and application: For beer and wort.



## Test preparation

### Test storage

Storage temperature: 15–25 °C (59–77 °F)

### Items to collect

Description	Quantity
1-cm quartz-glass cuvette (LZP332)	1
Pasteur pipet (150 mm) with suction pipet (LZC141)	1

### Before starting

Clean the quartz-glass cuvette thoroughly after each series of measurements. First, clean the quartz-glass cuvette with alcohol (isopropyl or pure ethanol), second, clean with plenty of water, third, clean with more alcohol and allow to air-dry. For sample batches with widely varying bitter units, it is recommended to clean the quartz-glass cuvette between samples with iso-octane.

UV-active substances can cause inconclusive findings. These substances include, among others, plasticisers. The iso-octane phase must be transferred to the quartz-glass cuvette using a glass pipet. When transferring the iso-octane, make sure that no iso-octane gets into the suction pipet.

Absolute absorption levels are measured when determining the bitter units. This means that any fluctuations between different devices and/or cuvettes and changes in the iso-octane quality have a direct impact on the measurement result. Therefore, a unique zero value must be set on the photometer for each series of measurements. The same quartz-glass cuvette must be used for determining the zero value and measuring the sample.

Any samples containing carbon dioxide must be degassed before analysis (e.g., stir with a magnetic stirrer LYW 977); Do not filter the samples, as the foam will also contain bitter substances. Clear the wort and cloudy beer by means of a centrifuge prior to analysis.

Before taking a series of measurements, the zero value must be set one time on the photometer.

Several samples can be handled in parallel.

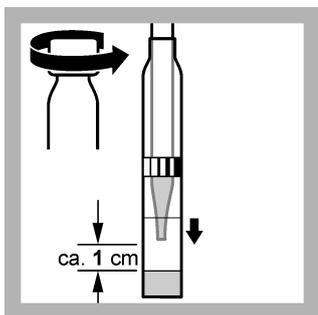
This method is applicable on DR5000/DR6000 only.

Review safety information and expiration date on the package.

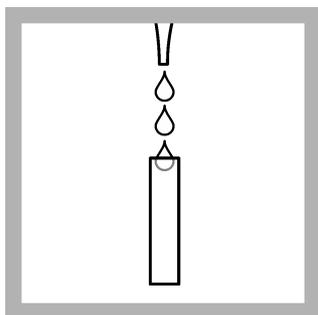
Review the Safety Data Sheets (MSDS/SDS) for the chemicals that are used. Use the recommended personal protective equipment.

Dispose of reacted solutions according to local, state and federal regulations. Refer to the Safety Data Sheets for disposal information for unused reagents. Refer to the environmental, health and safety staff for your facility and/or local regulatory agencies for further disposal information.

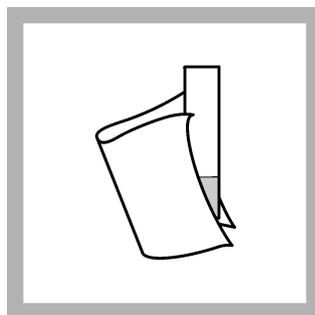
## Procedure—Preparation of the Zero Value



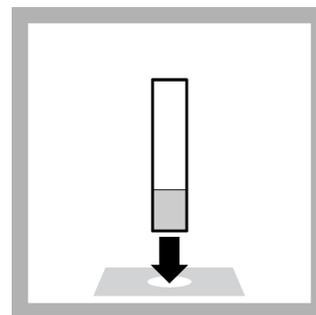
1. Open the cuvette. Remove the iso-octane phase from an unused sample cuvette using a clean Pasteur pipet up to approximately **1 cm** clearance from the aqueous phase.



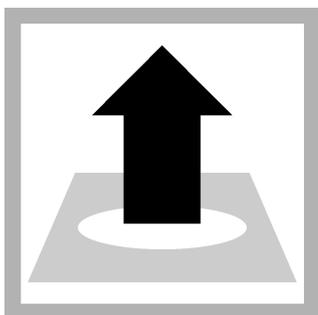
2. Pipet the iso-octane into the clean quartz-glass cuvette.



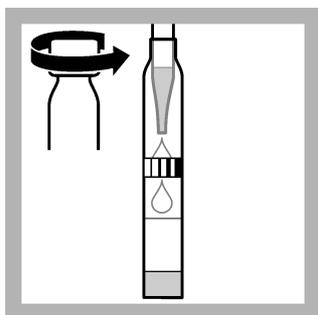
3. Thoroughly clean the outside of the quartz-glass cuvette.



4. Insert the quartz-glass cuvette into the cell holder. DR5000/DR6000: Go to Stored programs. Select the test, push **ZERO**.



5. Remove the cuvette.

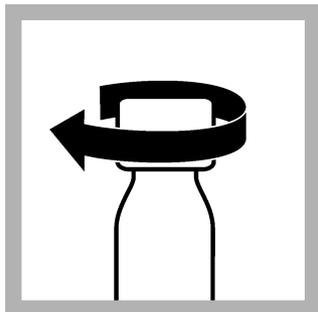


6. Transfer all of the iso-octane back into the sample cuvette using the Pasteur pipet. Now the cuvette can be used to extract samples.

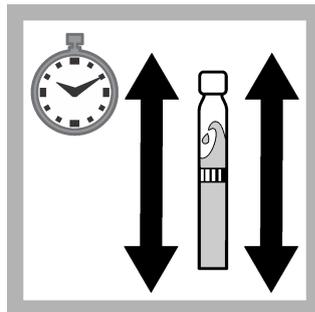
## Procedure—Sample Extraction



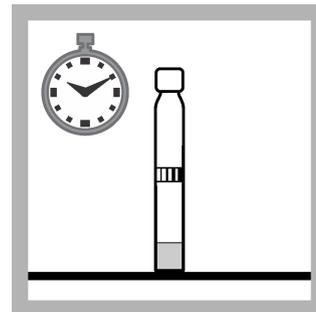
1. **Dispense iso-octane at bottom:** Run a 1-mL sample slowly along the length of the cuvette wall.



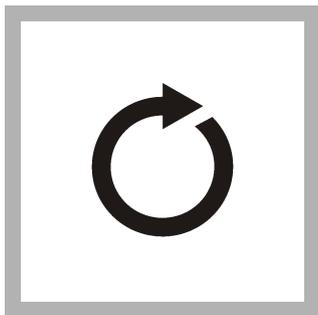
2. Close the cuvette.



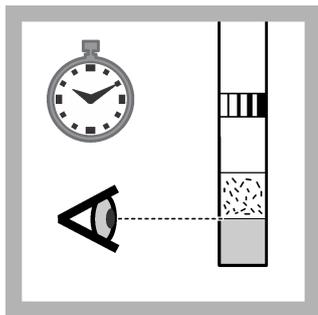
3. Shake the vial vigorously for **10 seconds**.



4. Let the contents of the cuvette settle for **20 seconds**.

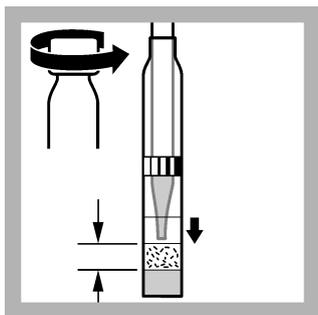


5. Repeat steps 3–4 **two** times.

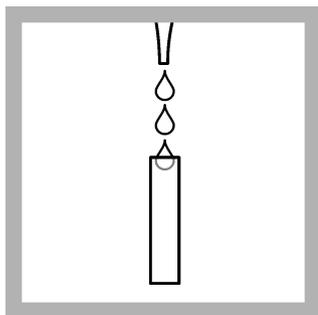


6. Let the contents of the cuvette settle for **1 minute** to ensure that the phase has fully separated.

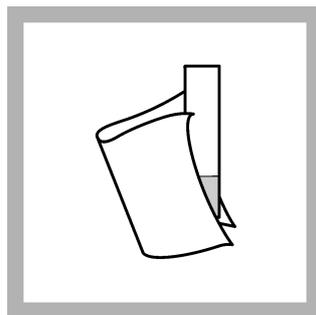
### Procedure—Evaluation



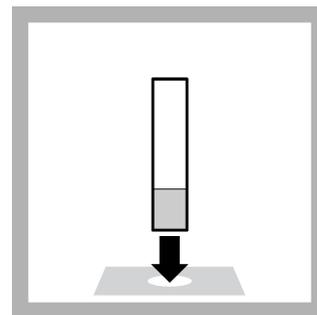
1. Remove the organic phase with up to **1 cm** clearance using a Pasteur pipet.



2. Use the pipet to dispense the organic phase into the quartz-glass cuvette.



3. Thoroughly clean the outside of the quartz-glass cuvette.



4. Insert the quartz-glass cuvette into the cellholder. DR 5000/DR 6000: Push **READ**.

### Summary of method

The bitter substances or recyclable materials in hops—primarily iso- $\alpha$ -acids—are extracted from the acidified sample with iso-octane. The concentration contained in the iso-octane extract is measured spectrophotometrically.



HACH LANGE GMBH  
Willstätterstraße 11  
D-40549 Düsseldorf

Tel. +49 (0) 2 11 52 88-0  
Fax +49 (0) 2 11 52 88-143

info-de@hach.com  
www.hach.com