

# Real-time PCR beer screening

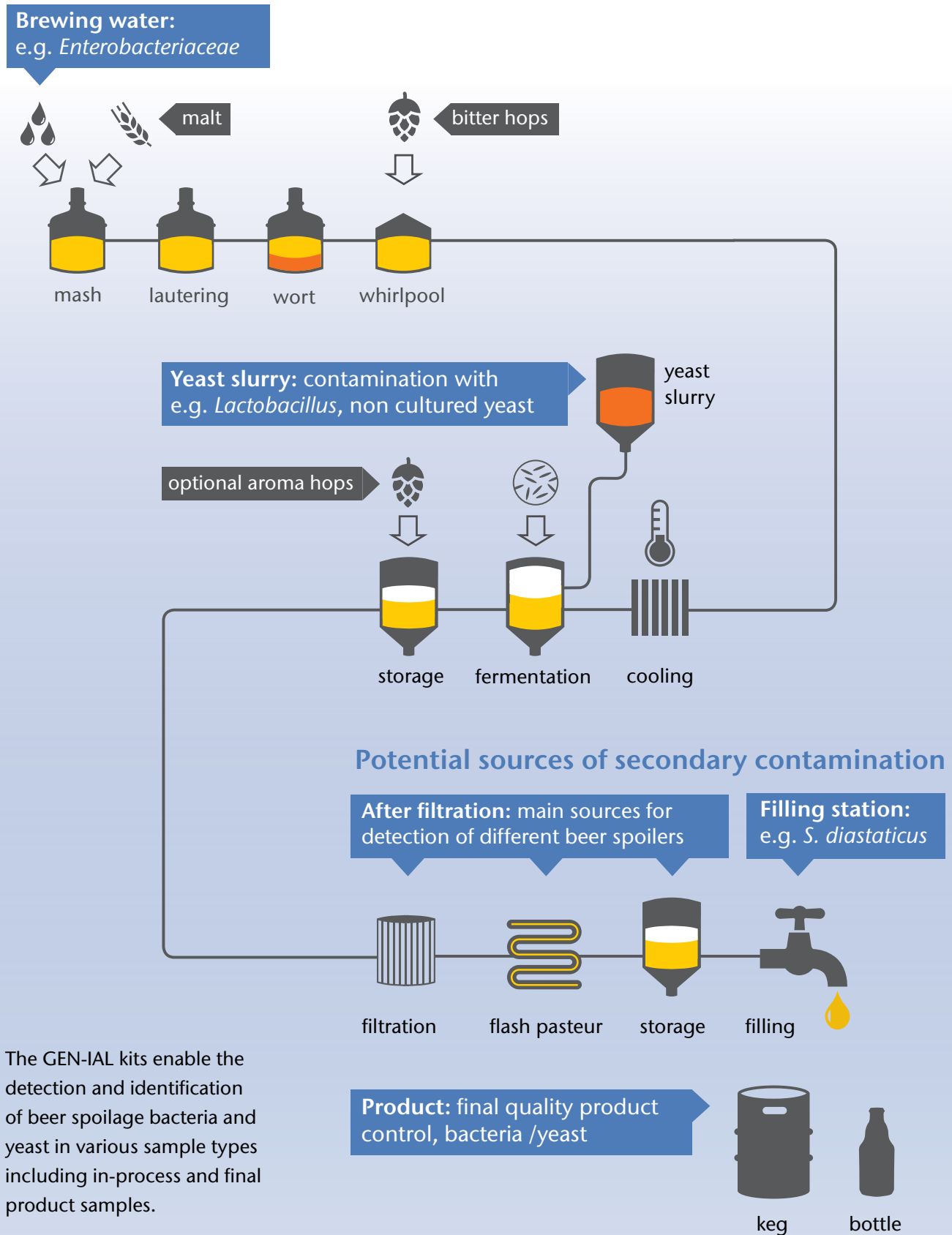
For a quick and GEN-IAL® result

- For yeast propagation, in-process control and/or for online final product control
- Avoid product recalls
- Reduce the risk of spoilage



## 5 potential sources of microbiological spoilage in beer brewing

### Potential sources of primary contamination



The GEN-IAL kits enable the detection and identification of beer spoilage bacteria and yeast in various sample types including in-process and final product samples.

## Faster and more sensitive results with real-time PCR – Customize your routine analysis

Either extremely fast or maximal sensitive detection or specific identification is possible by combining different methods of sample preparation and detection with real-time PCR.

### Approaches for microbiological analysis in beer

Time requirement: ~ 2 hrs

#### Centrifugation

- Centrifugation, qPCR: 2 hours from sample to result
- Fast screening approach for specific applications



- + Extremely fast
- + Screening and/or identification
- + Only two hours to result

- Volume maximum 30 mL
- Sensitivity maximum 10 - 100 cfu/30 mL (without enrichment)

Time requirement: ~ 2 hrs

#### Filtration and qPCR

- Filtration, qPCR: 2 hours from sample to result
- Fast screening approach for specific applications



- + Extremely fast
- + Screening and/or identification
- + Only two hours to result

- Volume maximum 1 L (depending on sample type)
- Sensitivity maximum 100 cfus

Time requirement: ~ 50 hrs

#### Enrichment and qPCR

- Pre-enrichment and qPCR: 2 days and 2 hours from sample to result
- Modern established approach for beverage analytic



- + Fast
- + Screening and/or identification
- + Ensure absence of low cfu / sample volume

Time requirement: ~ 50 hrs

#### Polymer binding and qPCR

- Sampling via polymer, pre-enrichment and qPCR
- Very sensitive approach for highest demands



- + Highest sensitivity
- + Up to 30 liter sample adsorption: kegs, in-process control, several bottles
- + Pre-enrichment of polymer ensures absence of few cfu/sample volume

## PolyBIND® – the new approach to adsorb and concentrate spoilage bacteria from large volumes

The separation and enrichment of microorganisms from large sample volumes or viscous liquids is still challenging in the beverage industry.

### Capturing bacteria with PolyBIND® (Art. No. PB 0050)

For the first time, the newly developed PolyBIND® particles enable the quick and easy isolation of

microorganisms and viruses from large sample volumes and highly viscous or solid-loaded liquids. PolyBIND® binds bacteria by adsorption. Using the sampling device Granusim 14.2, several bottles or one Keg up to 30 liters can be analyzed. Subsequent incubation in a medium of choice, allows state-of-the-art sampling for high demands/sensitivity of microbiological beverage testing.

The beer bottle with cleaned crown cap is placed in the chamber (bottom right) and the seal will be punched manually (top right) under sterile conditions. Carbon dioxide then flushes the sample in small fractions into a sample bottle (top left). Spoilage microorganisms bind to PolyBIND® during an incubation phase. The sample bottle will be removed and incubated with medium of choice.



### Matrices

#### Beers:

Pils, Kölsch, mixed beer drinks (e.g. Radler, Lemon), malt beer, IPA beers, Pale Ale, Ale, sour beer, stout, wheat beer, Kellerbier, Belgian beer, dark beers

#### Wines:

white wine, red wine

#### Soft drinks:

coke, apple juice, orange juice clear, milk

### Detection limit

The detection limit is 10 - 100 cells/analyzed volume.

### PolyBIND® binding capacity and efficiency

The binding capacity of different microorganisms is between  $10^5$  and  $10^6$  cfu / 100 mg PolyBIND®.

The binding efficiency depends on the pH-value of the liquid. The more acidic, the higher the binding efficiency. Optimal pH-values: 3 - 5. Sugar ingredients do not influence the binding efficiency.

### Microorganisms being validated

- *Lactobacillus brevis*
- *Lactobacillus buchneri*
- *Pectinatus cerevisiiphilus*
- *Saccharomyces cerevisiae* var. *diastaticus*
- *Lactobacillus lindneri*
- *Lactobacillus parabuchneri*
- *Megasphaera cerevisiae*
- *Candida parapsilosis*
- *Lactobacillus casei*
- *Lactobacillus collinoides*
- *Saccharomyces pasteurianus*
- *Dekkera bruxellensis*
- *Lactobacillus paracasei*
- *Lactobacillus damnosus*
- *Saccharomyces cerevisiae*

## Rapid and flexible detection of beer spoilage yeast and bacteria

Different combinations of screening and identification possibilities are available, thus allowing a cost effective and customized routine analysis.

### 1. Group Specific Screening

An initial screening gives a group specific answer of for example yeast and bacteria in one assay.

QTPYB0096\* In a mixture of groups and specific parameters, two samples can be run on one strip, therefore 24 samples per kit can be analyzed for:

Tube	Sample	FAM	HEX	ROX
1	1	<i>Enterobacteriaceae</i>	<i>Lactobacillus/Pediococcus</i>	<i>Pediococcus</i>
2	1	Wild yeast 1*	Bottom-fermented yeast	Internal control
3	1	Wild yeast 2*	Top-fermented yeast	Acetic acid bacteria
4	2	<i>Enterobacteriaceae</i>	<i>Lactobacillus/Pediococcus</i>	<i>Pediococcus</i>
5	2	Wild yeast 1*	Bottom-fermented yeast	Internal control
6	2	Wild yeast 2*	Top-fermented yeast	Acetic acid bacteria
7	NTC	–	–	Inhibition control
8	PTC	Positive Control	–	–

### 2. Screening and differentiation in one assay

The most relevant beer spoilage bacteria and yeast can be identified in one assay for example with QTPB 0096\* – 12 strips of 8 wells allows for the detection of the most relevant spoilage organisms for 12 samples in one assay.

Well	FAM	HEX	ROX
1	NTC	NTC	IAC
2	<i>Enterobacteriaceae</i>	<i>P. anomala</i>	<i>S. diastaticus</i>
3	<i>P. damnosus</i>	<i>P. acidilactici/pentosaceus/parvulus/inopinatus</i>	<i>P. clausenii</i>
4	<i>Pectinatus</i> spp.	<i>Megasphaera</i> spp.	<i>L. rossiae</i>
5	<i>L. brevis/L.parabrevis/L.brevisimilis</i>	<i>L. lindneri</i>	<i>L. cas ei/L.paracasei</i>
6	<i>L. buchneri/L. parabuchneri</i>	<i>L. collinoides/L. paracollinoides</i>	<i>L. perolens/L. harbinensis</i>
7	<i>L. plantarum/L. paraplantarum</i>	<i>L. coryniformis</i>	IAC
8	<i>L. acetotolerans</i>	<i>L. backii</i>	PTC

New

### 3. Detection and identification of yeast or bacteria only

Several kits are available for the detection of specific bacteria or yeast only.


QYDIF0096\* - 12 strips allow the identification of 12 yeast species.

Tube	FAM	HEX
1	NTC	Inhibition control
2	<i>Rhodotorula</i> spp.	<i>Saccharomyces exiguus</i>
3	<i>Candida</i> spp.	<i>Saccharomyces diastaticus</i>
4	<i>Saccharomycodes ludwigii</i>	<i>Debaromyces hansenii</i>
5	<i>Torulaspora delbrückii</i>	<i>Saccharomyces bayanus / pastorianus</i>
6	<i>Kluyveromyces marxianus</i>	<i>Hanseniaspora</i> spp.
7	<i>Dekkera</i> spp.	Inhibition control
8	<i>Pichia</i> spp.	PTC

\* Available for low, high and white tube profiles.



## GEN-IAL® – products for beer analysis

Product	Description	No. of tests/amount	Art. No.
<b>Beer</b>			
<b>DNA preparation</b>			
GEN-IAL® QuickGEN* Sample preparation filtration	DNA preparation of beverage samples, Filtration	100 preparations	FSE 0100
GEN-IAL® QuickGEN* Sample preparation centrifugation	DNA preparation of beverage samples, Centrifugation	100 preparations	CSE 0100
<b>Beer – bacteria &amp; yeast</b>			
<b>Qualitative multiplex real-time PCR</b>			
GEN-IAL® QuickGEN* First-Beer Differentiation PCR Kit	Multiplex detection (30 species) and identification (19 species) of relevant beer spoilers high profile: ABI 7500, Agilent MX3005P	96 reactions	QTPBD0096 high
GEN-IAL® QuickGEN* First-Beer Differentiation PCR Kit	Multiplex detection (30 species) and identification (19 species) of relevant beer spoilers low profile: Agilent Aria MX, Biorad CFX96, MyGo Pro	96 reactions	QTPBD0096 low
GEN-IAL® QuickGEN* First-Beer Differentiation PCR Kit	Multiplex detection (30 species) and identification (19 species) of relevant beer spoilers white strips: Biorad CFX96, LightCycler® 480	96 reactions	QTPBD0096 white
GEN-IAL® QuickGEN* First-Beer yeast and bacteria differentiation TaqMan™	Multiplex detection and identification of beverage spoiling bacteria and yeasts high profile: ABI 7500, Agilent MX3005P	96 reactions	QTPYB0096 high
GEN-IAL® QuickGEN* First-Beer yeast and bacteria differentiation TaqMan™	Multiplex detection and identification of beverage spoiling bacteria and yeasts low profile: Agilent Aria MX, Biorad CFX96, MyGo Pro	96 reactions	QTPYB0096 low
GEN-IAL® QuickGEN* First-Beer yeast and bacteria differentiation TaqMan™	Multiplex detection and identification of beverage spoiling bacteria and yeasts white strips: Biorad CFX96, LightCycler® 480	96 reactions	QTPYB0096 white
GEN-IAL® P1 Hyb Probe Screening LC 2.0 FRET	DNA screening and differentiation of bacteria and yeasts for LC 2.0	50 reactions	PP1H 0050 LC2.0
GEN-IAL® P1 Screening TaqMan™	DNA Screening and differentiation of beer spoiling bacteria and yeasts	50 reactions	PP1T 0050
GEN-IAL® QuickGEN* P1 Screening TaqMan™	DNA screening and differentiation of beer spoiling bacteria and yeasts	50 reactions	QPP1T 0050
 GEN-IAL® QuickGEN* P1 Screening TaqMan™	DNA screening and differentiation of beer spoiling bacteria and <i>Saccharomyces diastaticus</i>	50 reactions	QPP1SD 0050
GEN-IAL® QuickGEN* P1 Screening TaqMan™	DNA screening and differentiation of beer spoiling bacteria and hop resistance genes high profile: ABI 7500, Agilent MX3005P	48 reactions	QPP1HR0048 high
GEN-IAL® QuickGEN* P1 Screening TaqMan™	DNA screening and differentiation of beer spoiling bacteria and hop resistance genes low profile: Agilent Aria MX, Biorad CFX96, MyGo Pro	48 reactions	QPP1HR0048 low
GEN-IAL® QuickGEN* P1 Screening TaqMan™	DNA screening and differentiation of beer spoiling bacteria and hop resistance genes white strips: Biorad CFX96, LightCycler® 480	48 reactions	QPP1HR0048 white
GEN-IAL® P1 Screening Spartan DX-12 TaqMan™	DNA screening without differentiation of bacteria and yeasts	50 reactions	PP1T 0050 SP
GEN-IAL® P1OHScreening Spartan DX-12 TaqMan™	DNA screening without differentiation of bacteria	50 reactions	PP1TOH 0050 SP
GEN-IAL® QuickGEN* First-Biofilm TaqMan™	Specific DNA detection of <i>Lactococcus lactis</i> , <i>Leuconostoc mesenteroides</i> and <i>Pichia anomala</i>	50 reactions	QTPBF0050
GEN-IAL® First Yeast Hyb Probe Screening LC 2.0 FRET	DNA screening and differentiation of yeasts	50 reactions	PYHYB 0050 LC2.0



\* QuickGEN kits deliver a rapid, easy one-step DNA lysis and amplification method. Pre-enriched samples as well as centrifuged or filtrated samples may be used with CSE 0100 or FSE 0100 and subsequent QuickGEN detection kits.

## GEN-IAL® – products for beer analysis

Product	Description	No. of tests/amount	Art. No.
<b>Beer – bacteria</b>			
<b>Qualitative real-time PCR</b>			
GEN-IAL® Pectinatus spp./Megasphaera spp. TaqMan™	Specific DNA detection and differentiation of <i>Pectinatus</i> and <i>Megasphaera</i>	50 reactions	TPPMD 0050
<b>Beer – resistance genes</b>			
<b>Qualitative real-time PCR</b>			
GEN-IAL® QuickGEN* hop resistance genes horA and horC / hitA and orf5 TaqMan™	Specific DNA detection of hop resistance genes	50 reactions	QTPHR 0050
<b>Beer – yeast</b>			
<b>Qualitative real-time PCR</b>			
<b>New</b> GEN-IAL® QuickGEN First-Yeast differentiation PCR Kit	DNA screening and differentiation of 12 yeasts high profile: ABI 7500, Agilent MX3005P	96 reactions	QYDIF0096 high
<b>New</b> GEN-IAL® QuickGEN First-Yeast differentiation PCR Kit	DNA screening and differentiation of 12 yeasts low profile: Agilent Aria MX, Biorad CFX96, MyGo Pro	96 reactions	QYDIF0096 low
<b>New</b> GEN-IAL® QuickGEN First-Yeast differentiation PCR Kit	DNA screening and differentiation of 12 yeasts white strips: Biorad CFX96, LightCycler® 480	96 reactions	QYDIF0096 white
GEN-IAL® Dekkera anomala TaqMan™	Specific DNA detection of <i>Dekkera anomala</i>	50 reactions	TPYDA 0050
GEN-IAL® Pichia anomala TaqMan™	Specific DNA detection of <i>Pichia anomala</i> ( <i>Wickerhamomyces anomalus</i> )	50 reactions	TPYPA 0050
GEN-IAL® Saccharomyces diastaticus TaqMan™	Specific DNA detection of <i>Saccharomyces diastaticus</i>	50 reactions	TPYSD 0050
GEN-IAL® Pichia membranaefaciens TaqMan™	Specific DNA detection of <i>Pichia membranaefaciens</i>	50 reactions	TPYPM 0050
GEN-IAL® Bottom fermented yeast TaqMan™	Specific DNA detection of bottom fermented yeast	50 reactions	TPYUG 0050
GEN-IAL® Top fermented yeast TaqMan™	Specific DNA detection of top fermented yeast	50 reactions	TPYOG 0050
<b>GEN-IAL® accessories</b>			
<b>Real-time PCR</b>			
GEN-IAL® Dekkera bruxellensis Standards	DNA standards for <i>Dekkera bruxellensis</i> quantification	200.000 cfu	DBST 0100
Color Compensation Kit LightCycler® 480	Color compensation kit for multiplex assays	5 reactions	PP1TCC 0005
Color Compensation Kit LightCycler® 2.0	Color compensation kit for multiplex assays	5 reactions	CCFH 0005
Washing solution	Washing solution for SEW 0100	43 ml	WS 0100

\* QuickGEN kits deliver a rapid, easy one-step DNA lysis and amplification method. Pre-enriched samples as well as centrifuged or filtrated samples may be used with CSE 0100 or FSE 0100 and subsequent QuickGEN detection kits.



## Benefits

### + DNA Extraction – Easy one-step DNA lysis method

- One DNA extraction method for the detection of bacteria and yeast

### + Real-time PCR assays

- Ready to use consumables – tube stripes pre-coated with lyticase, primer and probes (Depending on the thermocycler, different versions of kits are available  
Further information is available on request)
- Simultaneous detection of the most relevant beer spoiling bacteria and yeast in one assay
- Internal amplification control (IAC)

### + New: qPCR program templates – no more programming of the thermocycler

- The qPCR thermocycler MyGo Pro is an affordable device which is suitable and recommended for the use with the GEN-IAL beverage kits
- Ready to download qPCR templates (settings for thermoprofile, dyes and samples) enable a most easy workflow, especially in combination with the new line of QuickGEN kits with pre-coated tubes

Contact your R-Biopharm sales representative for more information.