PIKA FastOrange® B Bouillon

Concentrated culture medium for the detection of beer spoiling bacteria

SKU #2036-1	FastOrange® B Bouillon	12 x 240 mL
SKU #2036-1-6	FastOrange® B Bouillon 6-Pack	6 x 240 mL

Description	Storage
Concentrated culture medium for the detection of all beer spoiling bacteria, *Lactobacillus* and *Pediococcus* species and also the anaerobic bacteria *Megasphaera* and *Pectinatus*, and *Dekkera* (*Brettanomyces*) yeasts.	In the dark at ambient temperature Cool storage is NOT necessary

Warning! Read the manual and the Safety Data Sheets before starting the analysis. Safety Data Sheets are available in the download area from www.pika-weihenstephan.com. All handling steps should be performed in sterile conditions. Wear appropriate protective clothing.

For *in vitro* use only.

Product description

PIKA FastOrange® B Bouillon is a concentrated culture medium which specifically detects beer spoiling bacteria in all stages of the brewing process from yeast to finished beer.

Lactic acid bacteria Lactobacilli and Pediococci as well as the anaerobes *Pectinatus* and *Megasphaera* are easily detected by a color change of the broth from violet to yellow.

Detectable microorganisms

Microorganism	Growth conditions
Lactic acid bacteria (*Lactobacillaceae*) - *Lactobacillus* sp. - *Pediococcus* sp.	aerobic and anaerobic
Acetic acid bacteria (*Acetobacteriaceae*) - *Acetobacter* sp. - *Gluconobacter* sp.	aerobic
Gram negative beer spoiling bacteria - *Pectinatus* sp. - *Megasphaera* sp.	anaerobic
Beer spoiling yeast - *Dekkera* sp. (*Brettanomyces*)	aerobic and anaerobic

Instructions for Use

Follow the user manual according to your sample type.

A. Clear samples and samples with low yeast concentration (e.g. beer, water, filtered samples)

For optimal color change, add an equal volume ratio of FastOrange® B Bouillon to the sample and mix. The final concentration of the medium is then 50%. The medium has not to be accurately measured, it is sufficient to pour it and visually judge the volume.

For example, mix app. 50 mL of sample with app. 50 mL of broth.

Incubate at 25 ± 2 °C (ambient temperature) for 3-7 days.

Note: Broth concentrations less than 50% may be used for the analysis of clear samples, but this will result in a decreased visibility of color change. Below a broth concentration of 30%, color change may not occur, but growth can still be monitored by turbidity and/or sediment formation. Compared to 50% broth concentration, time to visible growth may be prolonged.

B. Samples with high yeast concentration as fermenter samples or other yeast-turbid samples

For thick yeast analysis, serial enrichment gives best results.
We also recommend serial enrichment when the color of broth changes to yellow immediately upon mixing sample and medium.

Important! We strictly do NOT recommend using less than 50% final broth concentration for samples with high yeast concentration. Otherwise growth of yeast cells is not sufficiently inhibited and might lead to yellow color change, too.

For serial enrichment, first do enrichment as described in A. Clear Samples. For the second enrichment, choose one of the two recommended methods which are described in the following table.

Second enrichment	Method – use **either** option A, **or** option B
EITHER Option A	a. After 3 days of first enrichment, transfer half of the enriched sample into a fresh sterile tube or jar b. Add an equal amount of FastOrange® B Bouillon to the fresh tube/jar containing the first enrichment c. Incubate at 25 ± 2 °C for 3-5 more days
OR Option B	a. After 3 days of first enrichment, add an equal volume of fresh FastOrange® B Bouillon direct into the jar with the sample and broth (from first enrichment = 1. above) b. Incubate at 25 ± 2 °C for 3-5 more days

C. Swab samples and membrane filter analyses

1. Dilute FastOrange® B Bouillon with an equal volume of sterile water or sterile beer to reach 50% final.

Diluent	Application	Advantage
Sterile beer	Process samples and product relevant swabs (e.g. from filler area)	The mixture of beer and FastOrange® B broth provides a selective medium for growth of those beer spoiling bacteria which can grow especially in the beer type used
Sterile water	Swab samples, hygiene monitoring, surfaces	The FastOrange® B broth and water mixture provides growth of a broader range of microorga–nisms, as the inhibitory effect of hops is lacking. Grows even E. coli/coliforms, but without a color change to yellow

2. Fill a sterile tube or jar with the diluted FastOrange® B Bouillon and submerge the swab or membrane filter, then incubate.

Incubation conditions

Incubate at 25 ± 2°C for the following time, and analyze:

Analysis method	Incubation time
real time PCR	2-3 days
Visual inspection	3-7 days

Note

- Besides the beer spoiling bacteria, also *Dekkera* (*Brettanomyces)* yeasts may be detected. For specific detection of *Brettanomyces* yeasts, use FastOrange® BRETT Bouillon (SKU #2037-1) or Agar (SKU #2037-2).

- Growth of brewer's yeast and most wild yeast species with the exception of *Brettanomyces* is suppressed.

Result after incubation

Sample type	Samples are regarded as **positive** if showing one or more of the following
Clear sample	In 50% of broth – as recommended 1. Color change from violet-brown to yellow 2. Haze 3. Sediment
	In broth concentration 20 - 50%, monitoring will show ○ Decreased visibility of color change ○ Increased time to visible growth Below a broth concentration of 20% ○ no visible color change will occur ○ growth can be monitored by haze and sediment
Sample with yeast, as from fermentors or green beer	Bacteria 1. Color change from violet-brown to yellow 2. Increasing turbidity; often together with a yellowish sediment at the bottom
	Yeasts 1. If a high yeast concentration is present, a color change to yellow may occur directly after addition of broth. In this case, use serial enrichment as described in B. and monitor for increasing sediment and haze formation
Swabs and membrane filters	1. Color change from violet-brown to yellow 2. Haze and/or sediment formation
All sample types	In some cases, other microorganisms including hygiene indicator bacteria and accompanying flora might grow besides the typical beer spoi–lers. These are detected by turbidity and potential sediment formation, and usually show no color change to yellow

We recommend

- Microscopic examination or PCR analysis of positive enrichments to verify the presence of beer spoiling bacteria.
- Serial enrichment for thick yeast and also for samples which cause an immediate color change to yellow after the sample is mixed with broth.

General information

Store the product in the dark at room temperature (max. 25°C). Cooling below 25°C is NOT necessary.
Due to manufacturing, slight differences in color of culture medium may occur between bottles. This is NOT influencing product quality.
Best before date for unopened product is given on the outer label. After opening we cannot guarantee for shelf life.
The product is not suitable for human or animal consumption. It must not be used for the direct propagation of microorganisms which later are used for food production or might get into contact with food.

FastOrange® B Products

B Bouillon	(12 x 240 mL)	SKU #2036-1
B Bouillon 6-Pack	(6 x 240 mL)	SKU #2036-1-6
B Agar	(12 x 170 mL)	SKU #2036-2
B Agar 6-Pack	(12 x 170 mL)	SKU #2036-2-6
B Ready to Use Tubes 48-Pack (48 x 5 mL) with 48 Swabs		SKU #2036-3
B Tubes 48-Pack	(48 x 5 mL)	SKU #2036-10
B Enrichment Bottles	(15 x 50 mL)	SKU #2036-11

PIKA 4everyone™ Detection Kits for PCR Analysis

Acetics Screening	SKU 2401-15
Lactobacillaceae Screening	SKU 2401-32
LP Identification The real Beer Spoilers	SKU 2401-37
LP Screening The real Beer Spoilers	SKU 2401-38
Megasphaera sp. Screening	SKU 2401-41
Pectinatus sp. Screening	SKU 2401-44
Superattenuator Yeasts Screening	SKU 2402-58
S. cerevisiae var. diastaticus Screening	SKU 2402-49
Dekkera (Brettanomyces) sp. Screening	SKU 2402-20
Dekkera (Brettanomyces) anomala	SKU 2402-55
Dekkera (Brettanomyces) bruxellensis	SKU 2402-54
Dekkera (Brettanomyces) naardenensis	SKU 2402-56

More PCR tests are available, please check our website www.pika-weihenstephan.com/shop

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Notes: The relevant antibiotics/fungicides contained in the medium fall short of critical values that require monitoring or declaration according to regulation (EG) 1907/2006 (REACH).
When properly applied, the medium may be disposed of through the normal sewage system.
It is strongly recommended to inactivate the live microorganisms in any enriched sample by heating to 121°C/250°F for 20 min (autoclave) to exclude a release of live microorganisms.
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