



Reference : 01-674

Scharlau Microbiology - Technical data sheet

Product :
K AGAR

Specification

Selective solid medium used for the isolation of *Alicyclobacillus acidoterrestris*, in fruit juices according to IFU Standard Method No. 12.

Formula * in g/L

Yeast extract.....	2,50
Soy peptone.....	5,00
Dextrose.....	1,00
Polysorbate 80.....	1,00
Agar.....	20,00

Final pH 3,7 ±0,2 at 25 °C

* Adjusted and /or supplemented as required to meet performance criteria

Directions

Suspend 29,5 g in 1 L of distilled water and bring to the boil, to dissolve. Distribute in suitable containers and sterilize in the autoclave at 121°C for 15 minutes. Cool to 45-50°C and adjust the pH to 3,7 ± 0,2 with sterile 25% L-Malic acid solution. Mix well to homogenize and pour into sterile Petri dishes. Avoid heating or remelting the medium after the pH adjustment.

Description

Since the early 1980s, when spoilage of fruit juices by acid dependent thermotolerant spore-forming bacteria was recognised (Cerny *et al.*, 1984) members of the genus *Alicyclobacillus* have emerged as food spoilage organisms of major significance to the fruit juice industry (Baumgart & Menje, 2000). Spoilage is generally manifested as the formation of off-flavours and odours from compounds such as guaiacol and the halogenated phenols. To date, no human risk are known to be associated with the consumption of juices and other food products containing *Alicyclobacillus* bacteria, but the economic impact can be very high.

The low pH-value of the media, in combination with the high incubation temperature inhibits the growth of contaminating flora. Such is the case of some media such as BAT Agar (Ref 01-675) or YSG Broth (Ref 02-673). However, Agar K incubated at 45 ° C, supports the growth of predominantly *A. acidoterrestris* and limited growth of other species of the genus (*A. acidocaldarius*, *A. cycloheptanicus* and *A. hesperidium*). Therefore, K Agar can be used to detect predominantly *A. acidoterrestris* strains. This medium complies the Standard IFU Method on the detection of taint producing *Alicyclobacillus* in fruit juices.

Technique

The IFU Standard provides three methods of detection depending on the sample composition and the time since processing:

1. Raw materials (including process water): A heat shock treatment is prescribed followed by direct plating (optional), filtration or enrichment in liquid medium of the heated material.
2. End products sampled directly after (heat) processing where an additional heat shock is unnecessary: Pre-incubation of the sample in liquid medium is recommended.
3. End products taken from the market: Pre-incubation of the sample, and heat shock treatment is carried out. If spoilage is suspected and no *Alicyclobacilli* are detected after direct plating, a heat shock and enrichment treatment follow up is recommended.

In all the methodologies, incubation for 3-5 days at 45 ± 1°C is recommended. Count all colonies growing on the K Agar as presumptive *Alicyclobacillus acidoterrestris*. Confirm these colonies by further testing.

Quality control

Incubation temperature: 45°C ±2,0

Incubation time: 72 h-5 days

Inoculum: Pure culture. Inoculation by tried.

Microorganism	Growth	Remarks
<i>Alicyclobacillus acidoterrestris</i> ATCC® 49025	Good	-
<i>Bacillus subtilis</i> ATCC® 6633	Inhibited	-



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References

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- BAUMGART, J. & S. MENJE (2000) The impact of *Alicyclobacillus acidoterrestris* on the quality of juices and soft drinks. *Fruit Processing* 7:251-254.
- CERNY, G., W. HENNLICH & K. PORALLA (1984) Fruchtsaftverderb durch Bazillen: Isolierung und Charakterisierung des Verderberregers. *Z. Lebens. Unter Forsch.* 179:224-227.
- IFU STANDARDS (2004) Method No. 12 on the detection of taint producing *Alicyclobacillus* in fruit juices. March 2007 revision.
- ISO/TS 11133-1: 2009 Microbiology of food and animal feeding stuffs.- Guidelines on preparation and production of culture media. Part 1: General guidelines on quality assurance for the preparation of culture media in the laboratory.
- ISO/TS 11133-2: 2003 Corr. 2004 Microbiology of food and animal feeding stuffs.- Guidelines on preparation and production of culture media. Part 2: Practical guidelines on performance testing of culture media.
- LEE, S.Y., S.S. CHANG, J.H. SHIN & D.H. KANG (2007) Membrane filtration method for enumeration and isolation of *Alicyclobacillus* spp. from apple juice. *Letters in Appl. Microbiol.* 45:5: 540-546.
- WITTHUHN, R.C., W. DUVENAGE & P.A. GOUWS (2007) Evaluation of different growth media for the recovery of the species of *Alicyclobacillus*. *Letters Appl. Microbiol.* 45:224-229.

Storage

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).