A high-performance (sheet) solution for TAB

Juice and concentrate filtration with depth filter sheets – a solution for alicyclobacillus spp.

| Alicyclobacillus Spp. | Concentrates | Filter Sheets | Filtration | Fruit Juices | Microorganisms | TAB |

In today's fruit juice industry, the concept of safety is closely associated with a company's product quality. Here, the product quality depends largely on the fruit and processing method, because microorganisms that enter the juice — for instance, through soiled fruits during the harvest — from the process environment or during storage and transportation can spoil the product. However, this exposure does not necessarily result in an infection. The various selection factors in fruit juice, such as pH values and osmotic pressure have a growth-inhibiting impact on the majority of the naturally occurring microflora and thereby prevent a mass proliferation of these organisms naturally. Nevertheless, there are microorganisms whose proliferation cannot be prevented, which in turn can mean a negative change to the taste or spoilage of the product.

pH value and heat – the tools against bacteria

Most fruit juices have an acidic pH value of between 3.0 and 4.0. Since the majority of bacteria prefer neutral or alkaline habitats, this provides a natural protection for most juices. In support of this and to improve shelf life, clear fruit juices and fruit juice concentrates usually undergo a pasteurization process, which should prevent the spores from proliferating. In this process, the fruit juice is heated up to 194 to 203 °F (90 to 95 °C) for a period of 15 to 20 seconds, which kills off most bacteria and thereby stabilizes the microbiological status. In most cases, the hot-fill-hold process is used to continue the preservation, during which the newly pasteurized juice is filled in sterile bottles or carton boxes at about 180 to 198 °F (82 to 92 °C). Here, the temperature is then maintained for about two or more minutes before the final packaging is sealed and cooled.

***Alicyclobacillus spp.* – a particular challenge**

One exception is the endospores of the *alicyclobacillus acidoterrestris* species, which are registered with the International Fruit Juice Union (IFU) under the acronym TAB (thermo acidophilic bacteria). The representatives of the *alicyclobacillus spp.* species are large, immobile, spore-creating bacteria. Contrary to most other bacteria, TABs are both thermophile and acidophil: the duration, temperature and holding periods of the pasteurization are not sufficient to kill off the spores (resistant dormant body) of the bacterium. Nevertheless, one practical approach to deactivating the spores is to extend the heat treatment several fold (between one, and five and a half minutes), but this has two major downsides: Firstly, the juice/concentrate quality suffers the longer the juice/concentrate is heated. Secondly, the success is not guaranteed if the heat treatment lasts longer, meaning that the high temperatures can stimulate the germination of the spores in the thermophile microorganism. In this case, it has a heat-shock treatment effect on the spores.[1]

The high acid resistance of the spores also contributes to a constant or an increasing spore count in contaminated juices, even in the event of longer storage (> 40 days).[2] A negative impact on the vitality of the spores by the product being linked to a low pH value has not been proven in the laboratory to date. In products stored without cooling and particularly at higher ambient temperatures, germs grow rapidly after the germination of the spores.[3] In doing so, very small initial germ contents of 1 spore/ml can be sufficient to cause vegetative cells to germinate and proliferate. Since this ultimately means that the affected products will go bad, there are repeated calls in the beverage industry for zero tolerance.

Since the alicyclobacillus spp. is not a health hazard, going bad in this case means the formation of off-flavors.[4] The contamination of fruit juices and fruit juice concentrates with alicyclobacillus spp. is most apparent with the off-flavors guaiacol, 2.6-dichlorophenol and 2.6-dibromphenol. These off-flavors are described as medical, disinfection agent-like, antiseptic, phenolic or smoky.[5]

However, not all *alicyclobacillus spp.* can create off-flavors. Overall, 18 different species of the *alicyclobacillus spp.* are known. Nevertheless the off-flavor creation was

proven most prevalent with *alicyclobacillus acidoterrestris*, along with other species such as *alicyclobacillus acidophilus* and *alicyclobacillus herbarius*.[5] *Alicyclobacillus acidoterrestris* is the most significant in regards to the frequency and the ability to create off-flavors. In the event of a contamination with this bacterium, the above metabolism products make the juice undrinkable, but not health hazard. The change in quality cannot be seen, since there is neither a change in color nor any formation of gas.

Solution: Depth filter sheets – separation of spores

Traditional pasteurizing and ultrafiltration are not sufficient to stop the alicyclobacillus acidoterrestris in juice production. In the search for alternatives, Lippe University has conducted tests on spore suspensions on the instruction of the service laboratory of Eaton Technologies in Langenlonsheim, Germany. The initial aim was to show the effectiveness of depth filter sheets and then to make recommendations for the safe removal of TAB from fruit juices, or their semi and full concentrates.

To achieve both objectives, the germ retention rates LRV (logarithm reduction value) must be determined for the TAB spores in the depth filter sheets. Verification of the LRV is a cumbersome five day process. In this test arrangement, Eaton Technologies defines filtration with a depth filter sheet and a LRV of ≥ 5 as sterilizing for TAB. This means that with a LRV of ≥ 5 and an initial germ contamination of ≥ 105 cfu (colony forming units) per ml in the unfiltered liquid no more spores can be proven in the filtrate.

The test – structure and results

Laboratory filtrations with spore suspensions of *alicyclobacillus acidoterrestris* were carried out in the test. In this process, a total of five depth filter sheets with three different retention rates were tested:

- two depth filter sheets for sterile filtration (BECOPAD® 170 of Eaton as well as a depth filter sheet A of a competitor),
- two depth filter sheets for microbial reduction (BECOPAD 270 and depth filter sheet B of a competitor), and
- BECODPAD 350 depth filter sheet for fine filtration.

It must be noted that the definition of "sterile" in the beverage industry means exclusively the retention of germs harmful to beverages. Depth filter sheets A and B were selected to be as similar to BECOPAD depth filter sheets as possible to offer a fair comparison. All test filtrations were carried out in a double determination. The filtration flow corresponded to the industry standard of 12.3 gal/ft/h (500 l/m²/h).

Initially, a wild stem of *alicyclobacillus acidoterrestris* selected from fruit nectar was cultivated on BAT Bouillon (*bacillus acidoterrestris bouillon*) in accordance with IFU method 12 (2012) to determine the spore concentration. This was between 1.1 and 5.6 * 105 cfu/ml. Subsequently, the germ retention rate of the various depth filter sheets was determined.

The test results (*Fig. 1*) were extremely surprising. The two depth filter sheets BECOPAD 170 and A for sterile filtration

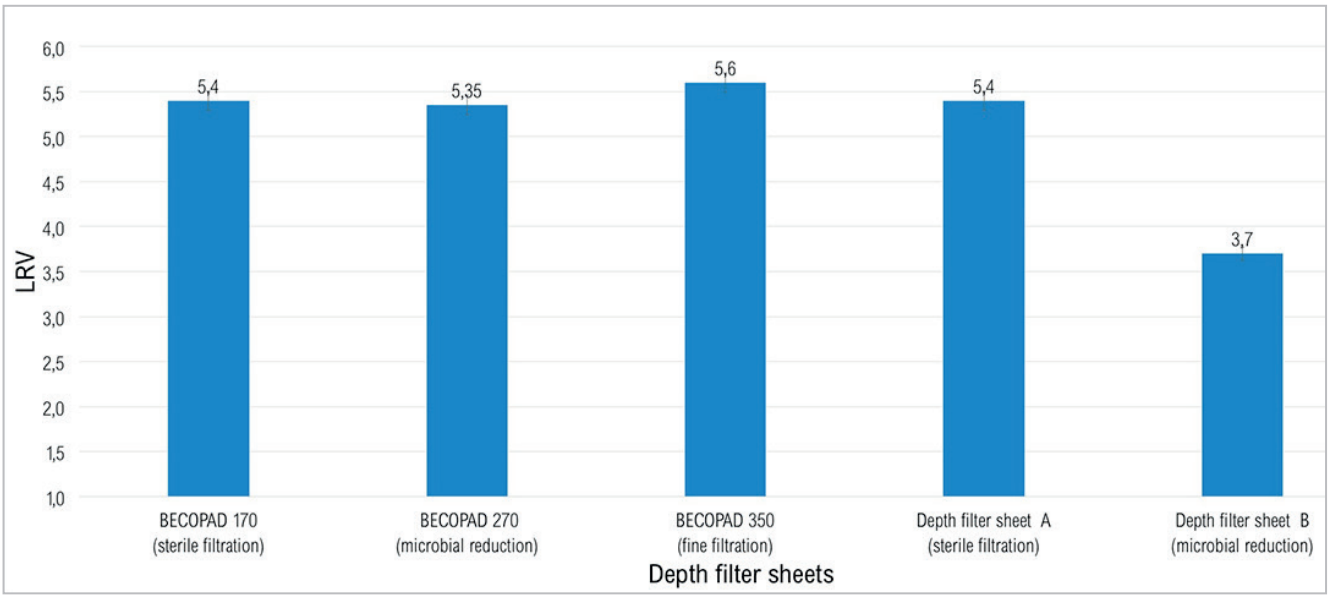

Fig. 1: Comparison of LRV of alicyclobacillus acidoterrestris spore solutions.

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and even the more open BECOPAD 270 (microbial reduction) and BECOPAD 350 (fine filtration) showed LRV considerably above 5. Only depth filter sheet B for microbial reduction performed worse, with a LRV of 3.7. These results back up earlier series of tests, during which the more open BECOPAD 270 and 350 already achieved high LRV and up to 100-fold higher germ retention rates compared to depth filter sheets with filtration-active components, such as diatomaceous earth.[6] These results are particularly interesting for industrial use, since they show that TAB spores can be removed with sheet filtration and allow one to suggest that it is not necessarily essential to resort to depth filter sheets for sterile filtration in order to achieve effective TAB filtration. According to the test, more open depth filter sheets such as BECOPAD 270 and 350 can also be used, which enable a higher flow.

The considerable difference in microbial reduction performance between depth filter sheet B (LRV 3.7) and BECOPAD 270 (LRV 5.35) is striking. It indicates that the composition of the depth filter sheets, its processing and structure also play a relevant role in separating *alicyclobacillus acidoterrestris* spores.

Sheet for sheet against TAB – recommendations for practical application

Eaton's BECOPAD depth filter sheets produced without addition of mineral components, such as diatomaceous earth, have been used successfully for more than ten years in the beverage industry – and also to separate TAB, as the laboratory tests demonstrate.

To make the laboratory results usable in practice, the test structure is guided by the usual industry standards and practical experience of Eaton for safe juice and concentrate production: A filtration flow of maximum 12.3 gal/ft/h (500 l/m/h), a filtration temperature of maximum 176 °F (80 °C) and a maximum pressure difference of 22 psi (1.5 bar). Initially, the determination of the spore concentration is vital. At an initial concentration of ≥ 105 cfu/ml *alicyclobacillus acidoterrestris* spores, no residues can be detected any longer after the filtration with the tested BECOPAD 170, 270 and 350 depth filter sheets (LRV ≥ 5), which means they are perfectly suitable for this application.

At a higher initial spore concentration of > 5.6 * 105 cfu/ml, there are two options: either the filtration flow can be reduced to separate more spores, or a two-level filtration process can be carried out – the latter being the safe option. In this process, a depth filter sheet is used for pre-filtering, with the aim of reducing the haze and the concentration to the maximum value of 105 cfu/ml. For costs and efficiency reasons, here—depending on the degree of contamination—pre-filtration is recommended with, for instance, BECOPAD 550 (coarse filtration), BECOPAD 450 (clarifying filtration) or even BECOPAD 350 depth filter sheets (fine filtration). After a successful haze reduction to 1 NTU (nephelometric turbidity units), one of the tested BECOPAD filter sheets with a LRV of ≥ 5 can be used. Regardless of the approach, it must be ensured that the filtered juice or the filtered concentrate is not exposed to any further contamination in the downstream process steps in order to avoid any reinfection during storage or transportation.

BECO COMPACT PLATE A600 filtration system

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BECOPAD® depth filter sheet

The BECOPAD® depth filter sheet is a premium depth filter medium made from high-purity cellulose, with no mineral components added. For more than ten years, it has satisfied customers with its gentle filtration, value preservation, microbiological safety and ability to increase performance and environmental protection. Apart of being fully biodegradable and it only requires up to 50 % less water for neutral rinsing and backwashing. Used in the BECO COMPACT PLATE A™ filtration system, it separates haze-causing, fruit juice-damaging and spore-forming germs such as alicyclobacillus acidoterrestris/TAB safely. The filtration system is a high-quality, multi-sheet filter in different sizes and with comprehensive equipment options. The flexible filter area can be adapted to an individual application. The hydraulic tightening unit with automatic subsequent pressing reduces drip loss to a minimum: a perfectly coordinated combination for the filtration of clear and stable fruit juices.