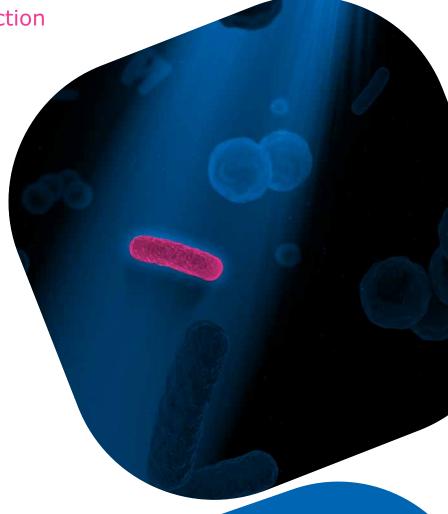


# The HybriScan® Molecular Screening System

For fast and accurate detection

of microorganisms





The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.

Millipore®

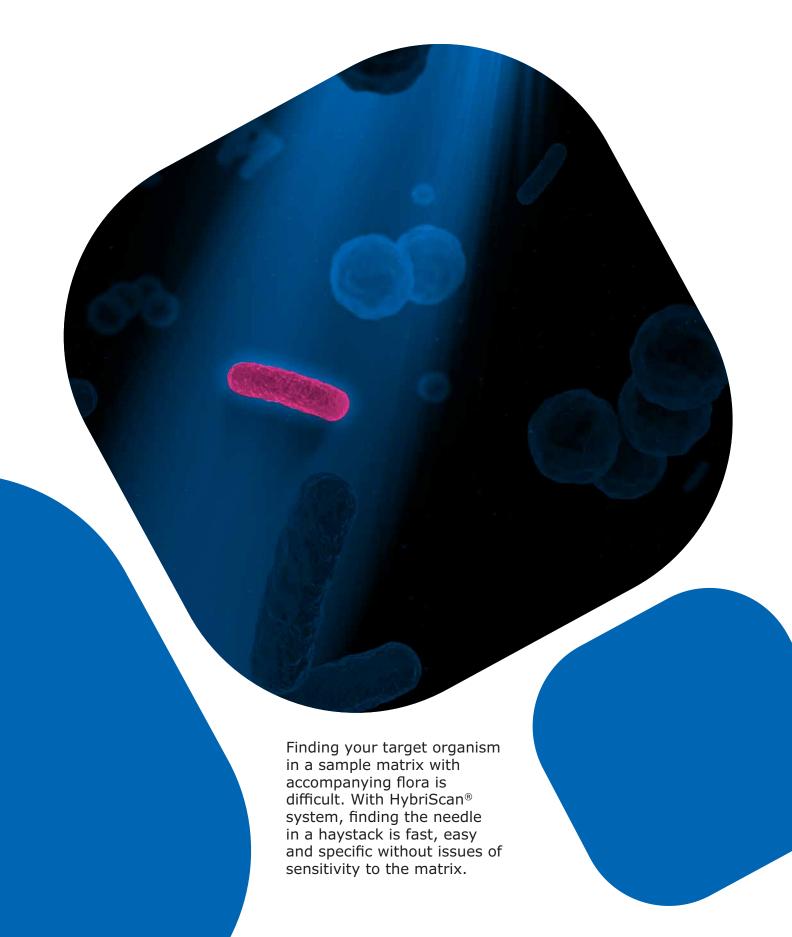
Preparation, Separation, Filtration & Monitoring Products

## Simple molecular screening for microorganism detection and identification—without PCR

When you need reliable routine control of microbial contamination in food production, from raw materials to finished goods, conventional standard cultivation-based methods are time consuming and often take from 2 to 15 days to get results. Ideal for the safety and quality control of beverages, water and food, our HybriScan® assays provides fast and accurate molecular detection of spoilage organisms and pathogens, without the need to use expensive PCR methods.

The HybriScan® system is based on the detection of microbe-specific rRNA using sandwich hybridization. The signal read-out is triggered optically by an enzymatically-generated color change. Results are available after only 2 hours of testing with a pre-enrichment step. Using two different specific probes for detection of microbial rRNA minimizes the possibility of a false positive.

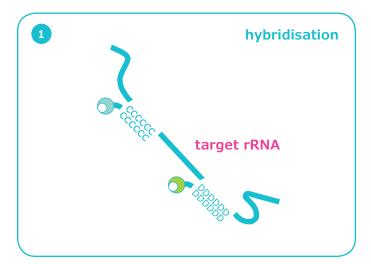
- Detects living cells only
- Not sensitive to sample matrix
- · High specificity
- Cost-efficient read-out using a standard plate reader (by eyes for qualitative results)
- Quantitative and qualitative results detect by organism group or specific species
- High sample throughput
- Can be used to detect non-culturable microbes
- Results in just 2 hours

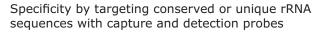


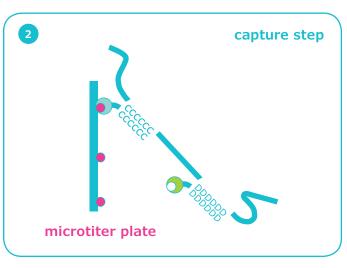
#### **Principle**

The HybriScan® method is based on the detection of rRNA via hybridization events and specific capture and detection probes. Sandwich hybridization is very sensitive, detecting attomoles of the respective target rRNA molecules providing sensitivity in crude biological samples because it is not susceptible to matrix interference.

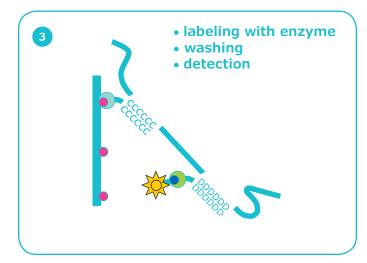
A capture probe is used to immobilize the target sequence on a solid support plate. A digoxigenin-labeled detection probe provides an enzyme-linked optical signal read-out. Detection of the desired targets results from application of anti-DIG-horseradish peroxidase Fab fragments. The bound complex is visualized by adding horseradish peroxidase substrate (TMB) finally results in a yellow color that can also be measured at 450 nm using a microplate reader.



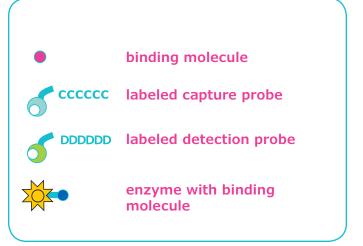




Biotin-labeled capture probe immobilizes the target sequence on streptavidin-coated microtiter plate



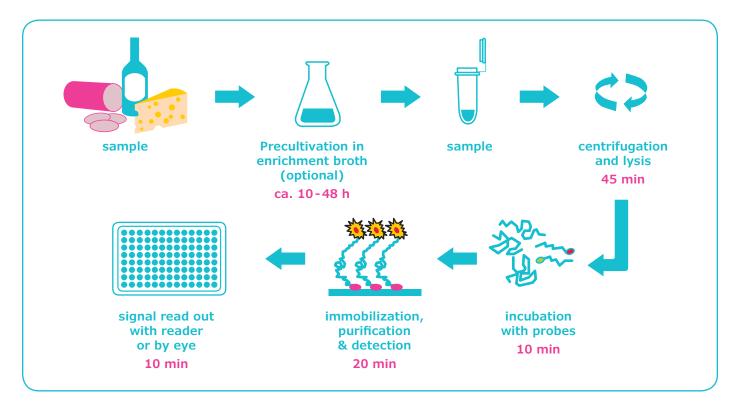
- Digoxigenin-labeled detection probe interacts with a DIG-horseradish peroxidase Fab fragment.
- The desired labeled complex is visualized using horseradish peroxidase substrate TMB (tetramethylbenzidine) giving a blue color which is then stopped with sulfuric acid and turns then to yellow and can be measured at 450 nm.



#### Workflow

The method simply needs a centrifuge, a thermal mixer and an optional a microplate reader. The complete analysis can be achieved in 2 hours for the HybriScan $^{\otimes}D$  kit (quantitative kits) and about 1 hour for HybriScan $^{\otimes}I$  kit (identification kits).

Qualitative results can be determined by eye or, for quantitative measurement, use with a microplate reader.



Two types of kits are available:

- Detection (D-kits) contain 96 reactions (12 strips with 8 wells) for quantitative results
- Identification (I-kits) contain 48 reactions (6 strips with 8 wells) for qualitative results

#### **Ordering Information**

Detection (quantification) kits	Cat. No.	Specifity	Beverage	Brewery	Wine	Water	Bioburden	Other Applications	Sensitivity
HybriScan® <b>D</b> Beer	62533	Beer spoiling organisms							1 -10 CFU/L (after
		<ul> <li>Lactobacillus, Pediococcus, Pectinatus, Megasphera</li> </ul>		Х					pre-enrichment)
HybriScan® <b>D</b> Drinks	68301	Beverage spoiling organisms							
		<ul> <li>amongst others yeast of the genera Saccharomyces, Zygosacchromyces, Brettanomyces, Torulaspora, Pichia, Candida</li> </ul>	x						1 -10 CFU/L (after pre-enrichment)
		<ul> <li>bacteria of the genera Lactobacillus, Acetobacteraceae and Alicyclobacillus</li> </ul>							
HybriScan® <b>D</b> E. coli	96343	E. coli				х		х	5,000 CFU (without pre-enrichment), 1-10 CFU after pre-enrichment
HybriScan® <b>D</b> Lactobac	59744	Lactobacilli (Lactobacillus and Pediococcus)	х	Х	х				1 -10 CFU/L (after pre-enrichment)
HybriScan® <b>D</b> Legionella	16593	Legionella				х			1,000 CFU (per assay), 1-10 CFU/L after pre-enrichment
HybriScan® <b>D</b> Legionella pneumophila	07190	Legionella pneumophila				х			1,000 CFU (per assay), 1-10 CFU/L after pre-enrichment
HybriScan® <b>D</b> Total Bacterial Count	02349	Total Bacterial Count	х	х	х	х	х		5,000 CFU (without pre-enrichment)
HybriScan® <b>D</b> Waste Water Microthrix parvicella	04447	Waste Water <i>Microthrix</i> parvicella				х			
HybriScan® <b>D</b> Waste Water Total Bacterial Count	78436	Waste Water Total Bacterial Count				х	х		
HybriScan® <b>D</b> Yeast	61397	Yeasts (including genera Zygosaccharomyces, Saccharomyces, Candida, Dekkera, Torulaspora and Pichia)	х	х	х				500 CFU/mL, 1 -10 CFU/L (after pre-enrichment)

Identififcation kits	Cat. No.	Specifity	Beverage	Brewery	Wine	Water	Bioburden	Other Applications	Sensitivity
HybriScan® <b>I</b> Brettanomyces	79742	Brettanomyces (Brettanomyces bruxellensis)	х	х	х				1000 CfU/assay
HybriScan® <b>I</b> Candida albicans	19503	Candida albicans						х	1000 CfU/assay
HybriScan® <b>I</b> E. coli	96343	E. coli				x		х	5,000 CFU (without pre-enrichment), 1-10 CFU after pre-enrichment
HybriScan® <b>I</b> Lactobacillus brevis	75724	Lactobacillus brevis	х	х	х				1000 CfU/assay
HybriScan® <b>I</b> Lactobacillus buchneri	80065	Lactobacillus buchneri	х	х	х				1000 CfU/assay
HybriScan® <b>I</b> Lactobacillus lindneri	86827	Lactobacillus lindneri	х	х	х				1000 CfU/assay
HybriScan <sup>®</sup> <b>I</b> Legionella pneumophila	49417	Legionella pneumophila				х			1000 CfU/assay
HybriScan® <b>I</b> Leuconostoc	77007	Leuconostoc	х	х	х				1000 CfU/assay
HybriScan® <b>I</b> Megasphaera	42875	Megasphaera (Megasphaera cerevisiae)		х					5,000 CFU (without pre-enrichment), 1-10 CFU after pre-enrichment
HybriScan® <b>I</b> Pectinatus cerevisiiphilus	89384	Pectinatus cerevisiiphilus	х	х					1000 CfU/assay
HybriScan® <b>I</b> Pectinatus frisingensis	73582	Pectinatus frisingensis	х	х					1000 CfU/assay
HybriScan® <b>I</b> Pediococcus damnosus	67289	Pediococcus damnosus	х	х	х				1000 CfU/assay

HybriScan® kits, their specificity and application area: HybriScan® $\boldsymbol{D}$  kits for quantitative testing,  $\boldsymbol{D}$  stands for detection. These kits contain 96 tests. HybriScan® $\boldsymbol{I}$  kits are for identification, the  $\boldsymbol{I}$  stands for identification and means it is a qualitative test and includes 48 tests (a half microtiter plate).

To learn more and for an actual and complete product list visit SigmaAldrich.com/HybriScan

#### **Equipment needed for HybriScan® kits**

Equipment	Cat. No.
Thermomixer Comfort, Eppendorf	Z605271
Exchange unit for 24 x 2 mL reaction tubes; Eppendorf	Z605670
Exchange unit for microtiter plate; Eppendorf	T3942
Centrifuge for 2 mL reaction tubes	Z605220 Z606235
Microplate reader (e.g. Multiskan FC)	n/a

### Millipore®

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