



Performance and tools

Microbiology

The ecotoxicological assessment of environmental samples requires some tests where bacteria colonies on petri dishes are counted, if necessary separately in colour and size. It has long been possible to count homogenous colonies of similar size with only some few intersections using colony counting tools. For the benefit of modern image processing today it is possible to count and classify inhomogeneous and coloured colonies. Even colonies growing side by side and touching can be separated and then counted with the help of the image processing system developed by LemnaTec:

Colonies of different colour, size and shape may be counted and assigned to separate categories. This classification may be adapted to any specific application. The evaluation of colonies on agar plates spreads the range of use of the LemnaTec Scanalyzer to become a multifunctional tool for the efficient evaluation of bio tests.

Concept

The main goal of the LemnaTec system in the scope of microbiology is to give a comprehensive and reproducible evaluation of colonies. But the Scanalyzer is far beyond a mere measuring instrument. It supports every step of the testing-routine, from preparation to statistics. On demand, LemnaTec installs the unit or instructs the users. Although being a system ready to use the Scanalyzer still remains flexible. The user may freely design the test and define many parameters of evaluation. Thus he remains in total control of aim and style of his ecotoxicological research.

Details in function guarantee such comfort in working with the Scanalyzer.

It is possible to optimise the quality of the images of different types of agar and colonies by illuminating with backlight or dark-field and diffuse light from above.

Performance of analytic software

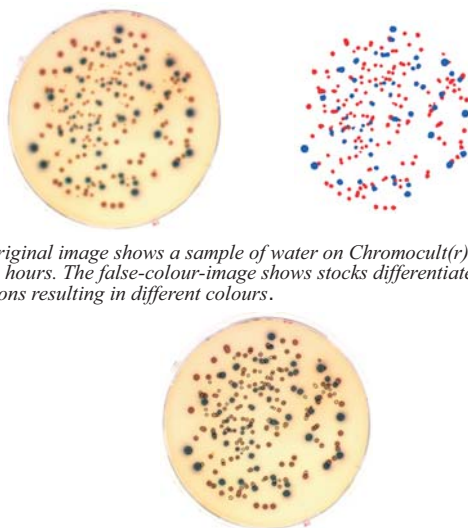
Identification of single colonies

The LemnaTec Scanalyzer separates complex structures of colonies into single colonies and identifies them by colour. The power and flexibility of the program guarantees perfect recognition of colonies and a high correspondence to the laborious manual counting. The methods of recognition are delivered with the Scanalyzer, ready to use, but may be modified by the user. Single mistakes may be corrected manually using an easy drawing tool. This tool allows to exclude, include, merge and separate objects.

Besides evaluating the number of the colonies and their total area, also size distribution is quantified. A classification of the colonies to colour classes and shapes completes the range of measurement.

Removal of background objects

To analyse petri-dishes all objects visible other than the colonies (sediment particles, blisters, faults of the agar, colour gradients) are removed automatically from the processed image. With the help of a dialog-based software assistant it is possible to optimise the methods offered. Single, unwanted objects may be removed easily in a second, correction-window.



The original image shows a sample of water on Chromocult(r)-agar incubated for 24 hours. The false-colour-image shows stocks differentiated by enzymatic reactions resulting in different colours.

On the basis of the outline image colonies are counted and the classification into colours is performed. In this case 154 red and 56 blue colonies were detected.

Colour classification

In an intuitive user interface the user can extract colours out of a representative image and name colour-classes (e.g. red, yellow, blue, etc.) so generating a false colour image. Any colour scheme may be saved for further reproducible assessment of colours. While creating a new colour-scheme original image and false colour image are displayed side by side. So the LemnaTec Scanalyzer differentiates even small but significant differences in colour.

Counting takes place automatically and the colonies are sorted into colour classes reproducibly, fast and comfortably.

Data evaluation of a single image

Because flexible working is very important in research, single images may be analysed. On this basis the Scanalyzer supports extremely heterogeneous test samples and special applications needing more manual assistance than usual.

Test series management and evaluation

In general the LemnaTec Scanalyzer is used in a routine of larger scale series of dilutions or screening samples. Every single step of a whole test is supported by the LemnaTec Scanalyzer. So, all data that are necessary to record for a test in compliance with standards may be entered and printed later. A dilution assistant helps with the preparation of a series of concentration and dilution (also LID-values referring to ISO) and the labeling of the samples. Identification of petri dishes can be comfortably automated with a barcode scanner. Pictures then can be taken without following a prescribed sequence but in the report all sample are displayed in a clear order. Results may be saved, printed

or exported.

The test series assistant serves to the necessary transparency in the quality control.

Specific application

Ames-Test

In the classical Ames-assay colonies of *Salmonella typhimurium* are counted with the Scanalyzer fast and efficiently in dark-field illumination despite their natural slight colouring. The test series management allows the organised recording of whole dilution series. The results are tabled clearly and the reversion coefficient is calculated. This allows a fast evaluation of screening test or sized dilution series to promote follow up tests.

Colony forming units

The determination of colony forming units in wastewater requires the recognition of very varying colonies and eliminating particles of pollution. This is achieved by an extensive classification of all recognised objects. Colonies can be differentiated to colour, form and size. That simplifies the evaluation of samples of wastewater, because types of colonies characteristically found there can be assigned morphologically to their varying stocks without repeated physiological characterisation. The user can determine the classification to colour and form by himself.

Chromogene nutrient-agar

A firm and secure colour evaluation is of particular importance if chromogene nutrient-agar gives a specific colouring to the varying types of bacteria. Using the effective LemnaTec image processing the differently coloured colonies are reproducibly classified and counted. The classification may be changed according to any user's interests and samples.

Documentation

All images, raw data, evaluation methods and results are saved accordingly to GLP within unchangeable raw data files and extra analysis files. Results may be easily tracked, reconstructed and reanalysed at a later time. So highest control of quality according to GLP is guaranteed.

In addition, all data are transferable to data bases. By marking the evaluated colonies within the image the counting process is documented extensively. So all steps are accessible permanently to the quality control. The application of barcodes simplifies the data recording and excludes mistakes.

Perspectives

Quantification of areas, shapes and colours is necessary and possible for a lot of other biotests. Please ask for the actual state of further developments.

