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detects a much later stage in the genotoxic process, requiring a longer time and more manipulations to complete. In the MUTATEST each sample is processed separately and large-scale screening poses a problem. Figure 1 clarifies the principles of the two tests and the differences between them. Figure 2 shows the various results that can be obtained.

Note that the Mutatest uses a mutant strain for histidine and that as a result natural compounds such as foodstuffs must be freed of this essential amino acid before analysis. The duration of the test is such that only sterile solutions can be used.

The S.O.S. CHROMOTEST, to the contrary, is not influenced by either histidine or bacterial contamination, which makes it ideal for foodstuff analyses.

The S.O.S. CHROMOTEST X-Gal as a ß-Galactosidase substrate which can be read either visuallyas various shades of blue - semi quantitative determination - or by an ELISA-reader - quantitative determination - at 600-605 nm.

METABOLISATION

Certain products must be metabolised before they become genotoxic. This is true for the most potent procarcinogens and the use of rat liver extract - S-9 - induced with Arocolor is recommended by Ames.

Toxicity and Genotoxicity

Some products are toxic to cells. To determine that a negative answer obtained by using the S.O.S. CHROMOTEST is not caused by cell death, a parallel measurement of cell death is indispensable. This is accomplished by measuring the enzymatic activity of alkaline phosphatase, an enzyme which is not under the control of S.O.S. functions and which is also detected by color reaction. Figure 2 will show you the differences obtained with different products.

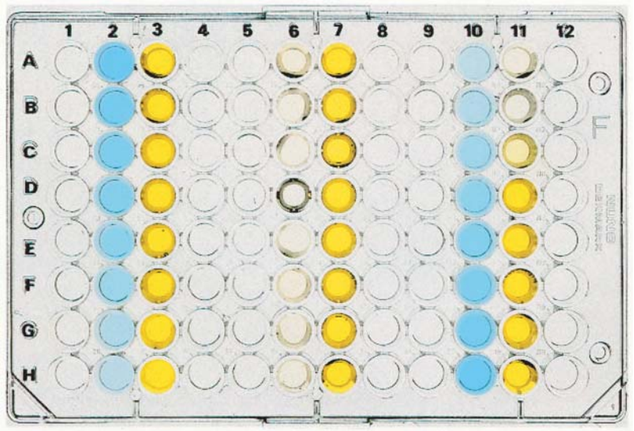

Figure 2

Columns 2-3: possible carcinogen; ß-Galactosidase activity (2) decreases with carcinogen dilution. Alkaline phosphatase activity (3) is optimal. Columns 6-7: no carcinogenic activity; no ß-Galactosidase activity (6). But the cells are alive; as can be determined by pronounced alkaline phosphatase activity (7).

Columns 10-11: toxic carcinogen. At high concentrations no alkaline phosphatase activity (11), indicating cell damage due to toxicity. ß-Galactosidase activity (10) appears at non-toxic concentrations of the tested material.

USES

The S.O.S. CHROMOTEST kit can be used to detect genotoxic activity in raw materials, cosmetics, pharmaceuticals, foodstuffs, water, soils, sediments.

EBPI offers complete technical support and will help to select the appropriate sample preparation procedure for a particular need.

If necessary, a collaborative study will be initiated for quick development of a specialized procedure.

THE S.O.S. CHROMOTEST

Fast and Economical Colorimetric Detection of Genotoxicity

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Adapted from M. Hofnung. Biofuture no.11, 1983

S.O.S. FUNCTIONS:

The cell's first response to genotoxic assault.

Genotoxic agents (carcinogens and mutagens, e.g. chemicals, irradiations) cause lesions in the DNA. Immediately thereafter the cell tries to restore the DNA to its original, native condition by activating a repair system called S.O.S. The result of S.O.S. repair efforts will determine the future of the cell.

- In a successful complete repair, the cell will resume its normal cycle and activities.
- In the case of an impossible repair, the damage will be too extensive and the cell will die.
- An incomplete repair will cause permanent changes in the genetic structure of the cell and may result in transmissible mutation or cancerous transformation of the cell.

The S.O.S. regulating system can be used for the detection of genotoxic agents.

The S.O.S. CHROMOTEST bacterial strain was originally patented by the INSTITUT PASTEUR & the French CHRs was restructured by genetic engineering methodology. An unrelated enzyme gene, ß-Galactosidase, normally absent from these bacteria, was linked to an S.O.S. operator gene. When the S.O.S. system is activated by genotoxic assault, the enzyme is produced and easily detected by a simple color reaction. In the S.O.S. CHROMOTEST the activity of the ß-Galactosidase is the result of genotoxic assault. Even cells that do not divide (and therefore do not give rise to colonies) would give a positive result in the S.O.S. CHROMOTEST.

The relationship between cellular events and the measured entities in the "S.O.S. Chromotest" and the "Mutatest"

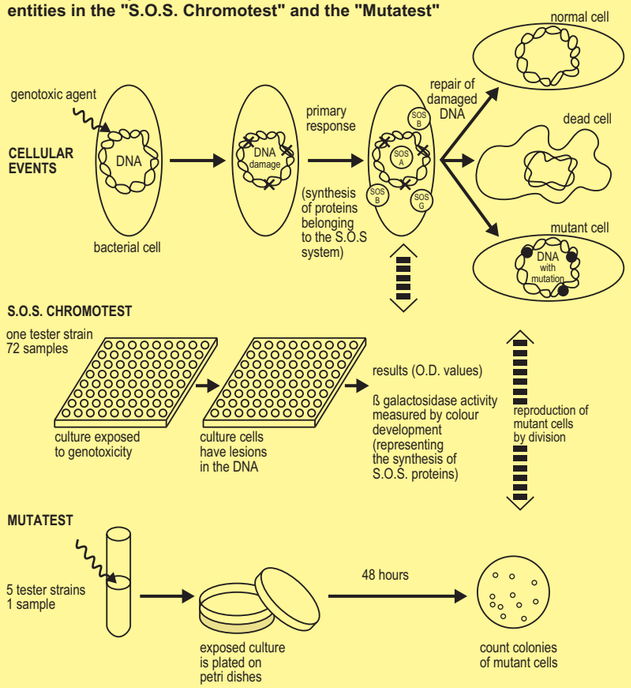

Figure 1

The S.O.S. CHROMOTEST Kit

The S.O.S. CHROMOTEST measures the primary response of a cell to genotoxic damage. In a few hours the EBPI S.O.S. CHROMOTEST Kit provides a clear, completely objective measurement of the genotoxicity of a compound by a simple appreciation of the color obtained compared to a color scale included in the kit or by spectrophotometry using a microplate reader.

The S.O.S. CHROMOTEST allows the examination of multiple samples all processed at once using only one cell tester strain. As was determined by testing more than 100 genotoxic compounds, the sensitivity of the S.O.S. CHROMOTEST is similar to that of the MUTATEST originated by Dr. Bruce Ames of the University of California at Berkley. The MUTATEST and its variations use Salmonella strains. This classical approach for measuring genotoxicity

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The S.O.S. CHROMOTEST simplifies the detection of genotoxic agents (carcinogens and mutagens) utilizing recombination techniques. The S.O.S. CHROMOTEST was inspired by the current knowledge of the cell's response to existence of lesions in its own genetic material - the DNA. The development into a kit form presents the simplest testing method for known carcinogens. The S.O.S. CHROMOTEST allows for testing genotoxcity in new chemicals raw materials, cosmetics and drugs and for monitoring waters sediments and industrial effluents.