MUTAGENESIS



Reference : M/MAT



Material

Les mutations et leurs effets phénotypiques sur le site régulateur de l'expression d'un gène

Objectives:

Approach to the Genotype-Phenotype relationship.

Mutations introduce a variability of genetic information.

To study the mutagenic effect of UV on yeasts, to deduce that they act directly on DNA by introducing mutations: appearance of revertants, decrease in the number of clones =lethal effect ...

Show that the phenotypic characters, directly related to the genotype, are transmitted from generation to generation.

Proposed activity:

Each pair of students has five boxes of medium specific to the growth of Ade2 yeast .

 Students display a defined concentration of yeast suspension (prepared by the teacher) on the five boxes.
Four of the five boxes are then placed in an irradiation box where the yeasts will be subjected to UV action. The fifth will not be exposed.

The four boxes correspond to four increasing exposure times.

Attention: The irradiation doses recommended in the protocol have been established with our products (strain, medium and irradiation box).

3. The boxes will be incubated 5 days at 28° C or one week at room temperature.

Results obtained:

The strain used carries a mutation that affects the Ade2 gene involved in the adenine biosynthetic chain. This mutation causes the accumulation of a compound (AIR) that turns into a red pigment: On agar medium, Ade2 yeasts form red colonies. If these yeasts are subjected to UV action, the introduction of mutations in their DNA may lead to a return to a "white" phenotype



(we call these revertants.) The longer the action of the UV is, the more the number of colonies decreases =lethal effect. The proportion of whites to red also increases with the exposure time.

Composition (for 20 pairs of students): STRAIN: Saccharomyces cerevisae Ade2 on Petri dish MEDIUM: Equivalent to 2 litres specific to the growth of our yeast Ade2, packaging according to the chosen option. MATERIAL:

- 70 sterile droppers
- 10 5ml sterile pipettes
- 1 KOVA numbering slide
- Sterile tubes: 45 of 5ml, 2 of 50ml and 4 of 10ml
- 20 inoculators
- 45 sterile spreaders
- 1 sterile bottle of 250 ml Technical and educational instructions available on our website.

Conservation:

3-4 weeks at 4° C. Attention, the strain blanches in the refrigerator, arrange for transplanting if you keep the strain for more than two weeks.

