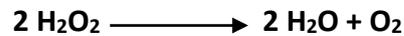


T.P. n°4 : Study of the enzymatic activity of catalase extracted from turnips (Effect of enzyme concentration)

Principle :

The principle is based on the disappearance of H₂O₂ in the presence of the enzyme source. In fact, catalases are present in a large number of tissues. They are cytoplasmic enzymes. They are tetrameric, with each unit carrying one heme molecule and one NADPH molecule. These enzymes help defend cells against oxidative stress by eliminating active oxygen derivatives and accelerating the spontaneous reaction that decomposes hydrogen peroxide (H₂O₂) into water and oxygen.

Catalase decomposes hydrogen peroxide into water and oxygen according to the reaction :



Equipment :

- ✓ Mixer ;
- ✓ Erlenmeyer flask + beaker ;
- ✓ Test tubes ;
- ✓ Pipettes and micropipettes ;
- ✓ Agitator ;
- ✓ Vortex agitator ;
- ✓ Filter or strainer ;
- ✓ 1 paper towel ;
- ✓ 1 fine-tipped clamp ;
- ✓ 10 filter paper discs cut out with a hole punch for binder sheets ;
- ✓ Chronometer.

Reagents :

- ✓ Turnip ;
- ✓ Oxygenated water ;
- ✓ Distilled water.

Procedure :

To prevent oxidation, use only plastic or silicone utensils (no metal).

1. Extraction of catalase from turnips :

- ✓ Cut a fresh turnip into pieces (without peeling it) and then blend in a mixer.
- ✓ Add distilled water (10 mL of distilled water for every 10 mL of turnip juice).
- ✓ Filter and store the mixture in a cool place.

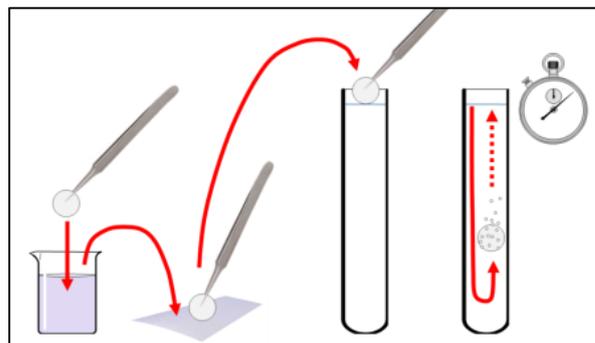
2. Tubes preparation :

- ✓ Label 6 test tubes, numbered 1 to 6.
- ✓ Fill the tubes with 20 mL of the 3% H₂O₂ solution :

Tube	1	2	3	4	5	6
Dilution of the mixture	0,1	0,2	0,4	0,6	0,8	1
mixture (mL)	1	2	4	6	8	10
Distilled water (mL)	9	8	6	4	2	0

3. Demonstration of catalase activity :

- ✓ Using fine-tipped clamp, dip a filter paper disc into the turnip mixture ;
- ✓ Briefly drain the excess on absorbent paper ;
- ✓ Place the disc vertically in the liquid at the top of the test tube. Immediately start the chronometer ;
- ✓ Measure the time it takes for the disc to return to the surface after sinking ;
- ✓ Record the value in the measurement table ;
- ✓ Repeat step 3 to test each tube.

**Work to do :**

- ✓ Trace the curve : round-trip time of the disc = f(mixture dilution).
- ✓ Interpret.