**Comparative analysis of catalase activity in various vegetable sources**

**Hypothesis:** Different vegetables sources in the catalase-hydrogen peroxide reaction will result in varying rates of oxygen release and therefore demonstrating that catalase activity differs among vegetables.

**Null hypothesis:** Different vegetable sources in the catalase-hydrogen peroxide reaction will not exhibit varying rates of oxygen release and therefore not show different catalase levels between them.

**Equipment**

* Celery, onion, mushroom, and carrot - used as the independent variables.
* Knives - used for dicing of vegetable to create maximal surface area.
* Kitchen scales - weighing out amount of vegetable source.
* Universal buffer (pH 7) - to maintain a constant pH and as a solution for the reaction to occur.
* Detergent - to visualise the capturing of oxygen from the reaction.
* Timers - to keep track of the reaction progress.
* 10% hydrogen peroxide - to react with the catalase enzyme within the vegetable sources.
* Autopipette - to measure out hydrogen peroxide.
* Measuring cylinders - to measure out universal buffer, as a container for the reaction, and to measure out the bubbles produced from the reaction.
* Room temperature (19-25°C) - a consistent temperature for use a control variable.

**Controls**

**Variables that will remain consistent:**

* The amount of pH 7 universal buffer (20mL),
* 3 drops of detergent,
* 2mL 10% hydrogen peroxide (excluding negative control),
* Consistent room temperature varying between 19-25°C, and
* 5g of a given vegetable source.

**Positive control:** Celery with hydrogen peroxide

Nearly all aerobic organisms all contain the catalase enzyme (Nandi A et. al; 2021), meaning we can say with a high degree of certainty that celery should produce oxygen when exposed to hydrogen peroxide.

**Negative control:** Celery without hydrogen peroxide. We will test the celery without the hydrogen peroxide as we know the catalase enzyme will not react without it present.

**Independent variable:** The vegetable source.

**Dependent variable:** The measured level of bubbles in millimetres produced from the trapping of oxygen produced in the catalase-hydrogen peroxide reaction.

**Replicates**

We will do each test with each vegetable source three (3) times to account for human error and to

create a mean production of O2 bubbles. This will equate to a total of 12 non-control experiments and an overall total of 14 experiments.

**Positive control:** No changes to treatment from the methodology.

**Negative control:** Not using hydrogen peroxide in the treatment.

**Changes to the methodology:** We chose to keep the methodology much the same as a previously written experiment.

* The vegetable source being cut into ‘fine pieces’ maximises the surface area to volume ratio meaning it does not have to be altered.
* 5 grams is plenty of vegetable source to run the reaction meaning any more than 5 grams is wasteful.
* 20mL of universal buffer is enough solution to be the medium for the reaction to take place.
* We do not want to have pH be an extraneous variable, meaning a relatively low hydrogen peroxide concentration of 10% is suitable for the purposes of the experiment.
* The primary change is the addition of step 10 where we added to repeat all the steps three times with each vegetable source.

**Method**

1. Using a razor blade, carefully cut the vegetable source into fine pieces (WARNING: Sharps; risk of cuts; use great care. RISK: Low).
2. Using the balance, weigh out 5 grams of the finely cut vegetable source.
3. Pipette 20 mL of Universal buffer (pH7) into a 100 mL measuring cylinder.
4. To the cylinder add 3 drops of detergent and gently swirl until buffer and detergent are mixed. Avoid creating bubbles.
5. To the cylinder add the 5g of vegetable source. If the pieces are stuck to the sides, push them down into the buffer solution.
6. Make a note of the volume in each cylinder and record it in your data sheet. This is essential to calculate the rate of reaction.
7. If you are completing more than one test, label each cylinder according to the test it belongs to, and set the timer for 15 minutes.
8. To the measuring cylinder add 2 mL 10% hydrogen peroxide (negative control excluded) and immediately start the timer.
9. Note the volume of foam created in the measuring cylinder and record it in your datasheet after 5, 10 & 15 minutes.
10. Repeat each step for each vegetable source, for a total of three tests for each source (excluding positive and negative controls).

**References**

Nandi A, Yan L-J, Jana C K, and Das N; (2019); ‘Role of Catalase in Oxidative Stress- and Age-Associated Degenerative Diseases’; *Oxidative Medicine and Cellular Longevity*; 2019:9613090;

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