APPENDICES

Preparation of reagents for various experiments are given below-

1. Preparation of chemicals for the estimation of Chlorophyll

a) 80% Acetone

- Acetone 80 ml
- DDW 20 ml

2. Preparation of chemicals for protein estimation

a) 50% Hydrochloric acid

- Conc. HCL 10 ml
- DDW 10 ml

b) 6 N Sodium hydroxide (NaOH)

- NaOH 24 gm
- DDW 100 ml

c) 80% Ethanol (EtOH)

- EtOH 80 ml
- DDW 20 ml

d) 5% Perchloric acid (PCA)

- PCA 13 ml
- DDW 270 ml

e) 1N NaOH

- NaOH 4 gm
- DDW 100 ml

f) Preparation of protein binding dye

- Coomassie Brilliant Blue G-250 10 mg
- 95% EtOH 5 ml
- 85% (w/v) Phosphoric acid 10 ml
- DDW 100 ml
Final concentration in the reagent were 0.01% (w/v) Coomassie Brilliant Blue G-250, 4.7% (w/v) ethanol and 85% (w/v) phosphoric acid.

3. Preparation of chemicals for the estimation of lipid peroxidation

a) 50 mM phosphate buffer (pH 7.0)

\[
\begin{align*}
100 \text{ mM } & \text{KH}_2\text{PO}_4 & 3.89 \text{ ml} \\
100 \text{ mM } & \text{Na}_2\text{HPO}_4 & 6.11 \text{ ml}
\end{align*}
\]

b) TBA-TCA solution (0.5% Thiobituric acid (TBA) in 20% TCA)

\[
\begin{align*}
\text{TBA} & 0.5 \text{ gm} \\
0.1 \text{ N HCL} & 2 \text{ ml} \\
\text{TCA} & 20 \text{ ml} \\
\text{DDW} & 78 \text{ ml}
\end{align*}
\]

4. Preparation of chemicals for estimating SOD activity

a) 300 mM Na₂CO₃ (Mol. Wt. 106)

\[
\begin{align*}
\text{Na}_2\text{CO}_3 & 0.795 \text{ mg} \\
\text{DDW} & 25 \text{ ml}
\end{align*}
\]

b) 1000 µM NBT

\[
\begin{align*}
\text{NBT} & 8.18 \text{ mg} \\
\text{DDW} & 10 \text{ ml}
\end{align*}
\]

c) 378 µM NBT

\[
\begin{align*}
1000 \mu \text{M NBT} & 9.45 \text{ ml} \\
\text{DDW} & 25 \text{ ml}
\end{align*}
\]

d) 1000 µM Riboflavin (Mol. Wt. 376.37)

\[
\begin{align*}
\text{Riboflavin} & 7.52 \text{ mg} \\
\text{DDW} & 20 \text{ ml}
\end{align*}
\]

e) 100 µM Riboflavin

\[
\begin{align*}
1000 \mu \text{M Riboflavin} & 0.5 \text{ ml} \\
\text{DDW} & 4.5 \text{ ml}
\end{align*}
\]
f) 7.8 µM Riboflavin
   
   100 µM Riboflavin 1.56 ml
   DDW 18.44 ml

g) 78 mM L- Metionine
   
   100 mM L- Metionine 19.5 ml
   DDW 5.5 ml

5. Preparation of chemicals for estimating GPOX activity

a) Guaiacol 20 mM
   
   Guaiacol 0.22 ml
   DDW 100 ml

b) H$_2$O$_2$ 10 mM
   
   H2O2 0.4 ml
   DDW 9.6 ml

6. Preparation of chemicals for estimating CAT activity

a) 100 mM HEPAS NaOH buffer of pH 7.6
   
   2.383 g of HEPAS buffer is dissolved in 100 ml DDW and adjust its pH to 7.6 with 0.1 N NaOH.

b) 50 mM HEPAS buffer
   
   Take 50 ml from HEPAS buffer and dilute to 50%

c) 100 mM H$_2$O$_2$
   
   1.124 pure H$_2$O$_2$ is added to 18.87641 ml 100 mM HEPAS buffer

7. Preparation of chemicals for the estimation of proline

a) 3% Sulfosalicylic acid (SSA)
   
   SSA 3 gm
   DDW 100 ml
b) 3% Glacial acetic acid (GAA)

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<th>Component</th>
<th>Volume</th>
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<tbody>
<tr>
<td>GAA</td>
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<tr>
<td>DDW</td>
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c) M Ortho-phosphoric acid (OPA)

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<tr>
<td>DDW</td>
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d) Acid ninhydrin

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<tr>
<td>Pure GAA</td>
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<tr>
<td>6 M OPA</td>
<td>10 ml</td>
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LIST OF PUBLICATIONS


PAPER PRESENTED IN CONFERENCES


