**4-Method:**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  | B | 1 | 2 | 3 | 4 | 5 | S1 |
| Standard | -- | 0.2 | 0.4 | 0.6 | 0.8 | 1 | ------ |
| Sample | -- | -- | -- | -- | -- | -- | 1 |
| H2O | 1 | 0.8 | 0.6 | 0.4 | 0.2 | 0 | ------ |
| Formic acid | 0.5 ml | | | | | | |
| Ninhydrine | 2 ml | | | | | | |
| * Mix throughly after each addition . * Boiling water bath for 10 min and then allow to cool at room temperature for 10 min. * (a deep red colour should develop). * Add 10 ml. of the 2-propanol-water solution (1:1) were added to each tube * Mix well using Vortex * Measure the absorbance at 520 nm. | | | | | | | |

**5-Result:**

|  |  |  |
| --- | --- | --- |
| Proline concentration mg/dl | Abs. At 520 nm | Tubes |
|  |  | **1** |
|  |  | **2** |
|  |  | **3** |
|  |  | **4** |
|  |  | **5** |
|  |  | **6** |
|  |  | **S1** |

**6-Calculations:**

………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………………