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"TO STUDY THE EFFECT OF pH ON ENZYME ACTIVITY"

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ABTRACT :-
This experiment was performed in order to find a better understanding of whether or not pH will affect the enzyme activity. To test this, we had a different time frame to mix together substances with different pHs in order to test whether or not that specific pH was ideal for the tube. The results were as predicted, the pH increased the reaction rate if the solution was more neutral, whereas the pH that was more acidic or basic caused the reaction rate to decrease almost completely. All enzymes have an ideal pH value, which is called optimal pH. Under the optimum pH conditions, each enzyme showed the maximum activity. When the pH value deviates from the ideal conditions, the activity of the enzyme slows down and then stops. The enzyme has an active site at the substrate binding site, and the shape of the active site will change with the change of pH value. When the pH value of the reaction medium changes, the shape and structure of the enzyme will change. Depending on the extreme extent of the enzyme and pH changes, these changes may permanently destroy the enzyme, or once the conditions return to the desired range of the enzyme, the enzyme will return to normal. My hypothesis, pH affects enzyme activity because enzymes are proteins that get their molecular structure via interactions between the charges of the amino acids that form the protein chains. These interactions are in the form of a hydrogen bond which is affected by the pH level.

KEYWORDS: pH, Enzyme, Enzyme Activity, Enzyme Extract, Effect.

1. INTRODUTION:-

"Enzymes may be defined as biocatalysts synthesized by living cells. They are protein in nature (exception – RNA acting as ribosome), colloidal and thermolabile in character, and specific in their action."[3]

- **Factors Affecting Enzyme Activity:-**

1. Contact between Enzyme and Substrate
2. Concentration of Enzyme
3. Concentration of substrate
4. Order of reaction
5. Effect of temperature
6. Effect of product concentration
7. Coenzyme and Activators
8. Effect of time
9. Effect of Radiation
10. Oxidation
11. Water effect[3,4,5]
12. Effect of Ph on enzyme action

- **Effect of pH on enzyme action**

- Each enzyme works within quite a small pH range. There is a pH at which its activity is greatest (the optimal pH).
- This is because changes in pH can make and break intra- and intermolecular bonds, changing the shape of the enzyme and, therefore, its effectiveness.
- Most enzymes have optimum pH in the range of 4 to 9.

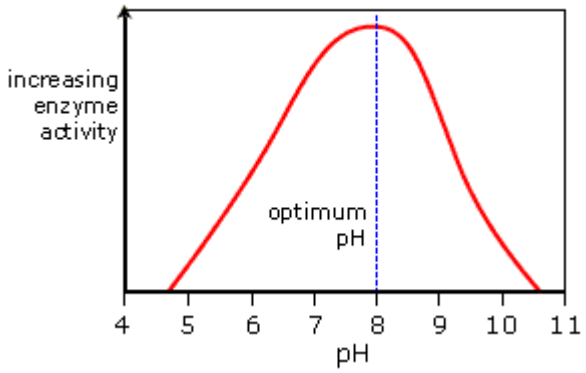


fig. no. 1 effect of ph on enzyme action.

2. AIMS AND OBJECTIVES:-

- **Aim:-** To study the effect of pH on enzyme activity.

- **Objectives:-**
  - i. To study and look at the review of literature on enzyme activity.
  - ii. To elaborate the activity of enzyme on various pH.
  - iii. To prepare different enzyme extract and to study them on different pH
  - iv. To compute the data of pH and absorbance.
  - v. To correlate the experimental and theoretical evidences of enzyme activity.

3. REVIEW OF LITERATURE:-

i. **Robytet.al (1967)** Three conceptual action patterns of α-amylase hydrolysis of amylose have been considered: single chain, multichain, and multiple attack. To test these concepts, curves were obtained relating the drop in amylose-iodine color to the increase in reducing value for amylolysis by human salivary (HS), porcine pancreatic (PP), Aspergillusoryzae (AO) α-amylases, and 1 m H2SO4. Robyt, John F., and Dexter French. "Multiple attack hypothesis of α-amylase action: action of porcine pancreatic, human salivary, and Aspergillusoryzae αamylases." Archives of biochemistry and biophysics 122.1 (1967): 88-16.

ii. **Hayashidaet.al(1988)** A newly isolated bacterium, identified as Bacillus subtilis 65, was found to produce raw-starch-digesting α-amylase. The electrophoretically homogeneous preparation of enzyme (molecular weight, 68,000) digested and solubilized raw corn starch to glucose and maltose with small amounts of maltooligosaccharides ranging from maltotriose to maltoheptaose. This enzyme was different from other amylases and could digest raw potato starch almost as fast as it could corn starch, but it showed no adsorbability onto any kind of raw starch at any pH. Hayashida, Shinsaku, Yuji Teramoto, and Takehiro Inoue. "Production and characteristics of raw-potato-starch-digesting α-amylase from Bacillus subtilis 65." Applied and Environmental Microbiology 54.6 (1988): 1516-1522.

iii. **Witt et.al(1996)** An α-amylase (EC 3.2.1.1) was purified to apparent electrophoretic homogeneity from potato (Solanumtuberosum L.) tubers by affinity chromatography on a starch granule column, Q-Sepharose chromatography, and gel filtration.The results suggest that this starch hydrolase is involved in the initiation of reserve starch dissolution in potato tubers. Witt, Wolfgang, and Jõrg J. Sauter. "Purification and properties of a starch granule-degrading α-amylase from potato tubers." Journal of experimental botany 47.11 (1996): 1789-1795.

iv. **Ceciet.al(2002)** The aim of this paper was to evaluate the effects of pH on the α-amylase activities in a commercial amylolytic enzyme used for apple juice processing. Kinetics of thermal inactivation was studied in acetate and citrate/phosphate buffers at different temperatures (55–70 °C) and enzyme concentrations (0.276 and 0.552 mL/100 mL). Maximum α-amylase activity was observed at pH=3.4 in both buffers. Ceci, Liliana N., and Jorge E. Lozano. "Amylase for apple juice processing: Effects of pH, heat, and Ca2+ ions." Food Technology and Biotechnology 40.1 (2002): 33-38.

v. **Thomas et.al(2003)** Climate change due to increased [CO2] and elevated temperature may impact the composition of crop seed. This study was conducted to determine the potential effects of climate change on composition and gene expression of soybean [Glycine max (L.) Merr. Cv. 'Bragg'] seed. Soybean plants were grown in sunlit, controlled environment chambers under diel, sinusoidal temperatures of 28/18, 32/22, 36/26, 40/30, and 44/34°C (day/night, maximum/minimum), and two levels of [CO2], 350 and 700 μmol mol−1, imposed during the entire life cycle.These results confirm previous studies indicating that high temperature alters soybean seed composition, and suggest possible mechanisms by which climate change may affect soybean seed development and composition. Thomas, J. M. G., et al. "Elevated temperature and carbon dioxide effects on soybean seed composition and transcript abundance." Crop Science 43.4 (2003): 1548-1557.

vi. **Liu, Wei et.al(2010)** A commercial cellulase was first assessed to be effective in hydrolyzing glycosyl at the C-3 and C-26 positions in steroidal saponins from yellow ginger (Dioscoreazingiberensis C. H. Wright) to diosgenin, a very important chemical in the pharmaceutical industry. The effect of different parameters on enzyme hydrolysis was further investigated by systematically varying them. Liu, Wei, et al. "Production of diosgenin from yellow ginger (Dioscoreazingiberensis CH Wright) saponins by commercial cellulase." World Journal of Microbiology and Biotechnology 26 (2010): 1171-1180.

vii. **Kumariet.al(2010)** Starch hydrolyzing amylase from germinated soybeans seeds (Glycine max) has been purified 400-fold to electrophoretic homogeneity with a final specific activity of 384 units/mg. SDS–PAGE of the final preparation revealed a single

protein band of 100 kDa, whereas molecular mass was determined to be 84 kDa by MALDI–TOF and gel filtration on Superdex-200 (FPLC). The enzyme exhibited maximum activity at pH 5.5 and a pI value of 4.85. The energy of activation was determined to be 6.09 kcal/mol in the temperature range 25–85 °C. Kumari, Arpana, et al. "α-Amylase from germinating soybean (Glycine max) seeds–Purification, characterization and sequential similarity of conserved and catalytic amino acid residues." Phytochemistry 71.14-15 (2010): 1657-1666.

viii. **Kumariet.al(2011)** Alpha amylase from soybeans was immobilized on two different matrices, Chitosan beads and Amberlite MB-150. Maximum immobilization of 62% and 70.4% was obtained with Chitosan and Amberlite MB-150, respectively. The optimum pH obtained was 8.0 and 7.0 for the α-amylase immobilized on Chitosan beads and Amberlite MB-150, respectively; free enzyme showed an optimum pH of 5.5. Kumari, Arpana, and Arvind M. Kayastha. "Immobilization of soybean (Glycine max) α-amylase onto Chitosan and Amberlite MB-150 beads: Optimization and characterization." Journal of Molecular Catalysis B: Enzymatic 69.1-2 (2011): 814.

ix. **Niveditaet.al(2011)** The detailed kinetic study of the immobilized enzyme is usually important and pertinent for the better management of the enzyme preparation. Therefore, the suitable comparison of the various kinetic parameters of the immobilized αamylase (on gelatin) with that of the soluble enzyme has been conducted. The immobilized α-amylase could be considered as a potential candidate for use as a cleaning additive in detergents in order to facilitate the removal of starch stains due to which it may find potential application in laundry detergents. Nivedita, Jaiswal, and Prakash Om. "Immobilization of soybean α-amylase on gelatin and its application as a detergent additive." Asian Journal of Biochemistry 6.4 (2011): 337-346.

x. **Cristina et.al(2012)** This article presents two integrated laboratory exercises intended to show students the role of α-amylases (AAMYs) in saliva and detergents. These laboratory practicals are based on the determination of the enzymatic activity of amylase from saliva and different detergents using the Phadebas test (quantitative) and the Lugol test (qualitative) under different conditions (e.g. variations in temperature and alkalinity). Valls, Cristina, et al. "Characterization of the activity and stability of amylase from saliva and detergent: Laboratory practicals for studying the activity and stability of amylase from saliva and various commercial detergents." Biochemistry and Molecular Biology Education 40.4 (2012): 254-26.

xi. **Izmirliogluet.al(2012)** Bio-ethanol is one of the energy sources that can be produced by renewable sources. Waste potato mash was chosen as a renewable carbon source for ethanol fermentation because it is relatively inexpensive compared with other feedstock considered as food sources. However, a pretreatment process is needed: specifically, liquefaction and saccharification processes are needed to convert starch of potato into fermentable sugars before ethanol fermentation. In this study, hydrolysis of waste potato mash and growth parameters of the ethanol fermentation were optimized to obtain maximum ethanol production. Izmirlioglu, Gulten, and Ali Demirci. "Ethanol production from waste potato mash by using Saccharomyces cerevisiae." Applied Sciences 2.4 (2012): 738-753.

xii. **Shahzad, et.al(2016)** The present study is concerned with the production and characterization of αamylase by Aspergillusniger in solid-state fermentation using food waste as substrate. Various cultural conditions such as incubation period, incubation temperature, pH of the medium, moisture level and inoculum size were optimized for maximum αamylase yield. The maximum activity of enzyme (1262.27 ± 2.11 U/g) was recorded after 72 h of incubation at 30°C temperature, pH 5 with 5% moisture level and inoculum size. Among different nitrogen and carbon sources evaluated, peptone (1.5%), NH4NO3 (0.75%) and soluble starch (1.25%) gave maximum α-amylase production under optimized conditions. Mahmood, Shahzad, et al. "Production and optimization of α-amylase from Aspergillusniger using potato peel as substrate." Pakistan Journal of Biotechnology 13.2 (2016): 101-109.METHOD.

xiii. **Hester et.al(2019)** The purpose of this study was to determine the impact of thermal treatment of garlic, ginger, and turmeric on total phenolic content (TPC), total flavonoid content (TFC), scavenging activity, ferric reducing antioxidant potential (FRAP), trolox activity (TEAC), lipase, a-amylase, and a-glucosidase inhibition. Conventional stovetop heating of selected spices was performed followed by methanolic and aqueous extractions (1 – 5 minutes; 70°C – 130°C). Overall methanolic extracts had higher phytochemical, antioxidative, and anti-diabetic potential. However, aqueous garlic extracts exhibited higher phytochemical and antioxidative potential over methanolic garlic extracts. Hester, Fredreana, et al. "A comparison of the antioxidative and anti-diabetic potential of thermally treated garlic, turmeric, and ginger." Food and Nutrition Sciences 10.2 (2019): 207-219.

xiv. **Faleseet.al(2021)** Human salivary α-Amylase (HSAmy) non-catalytic but cellular interaction with sanguinarine (SG), a bioalkaloid with a high therapeutic promise, was sought through multiple spectroscopic approaches and molecular docking under simulated salivary physiological conditions. Falese, Babatunde A., et al. "Probing the interaction of iminium form of sanguinarine with human salivary α-amylase by multi-spectroscopic techniques and molecular docking." Journal of Molecular Liquids 334 (2021): 116346.

xv. **Dalsassoet.al(2022)** The work carried out in this review based on the relation between ginger processing and their bioactive compounds, focusing especially on gingerols and shogaols, as well as the main processes that increase the content of 6-shogaol without compromising other phenolic compounds to produce highly functional extracts for future applications in the food packaging sector. It include broad overview of how these methods affect the composition and functionalities of ginger extracts. Dalsasso, Raul Remor, Germán Ayala Valencia, and Alcilene Rodrigues Monteiro. "Impact of drying and extractions processes on the recovery of gingerols and shogaols, the main bioactive compounds of ginger." Food Research International (2022): 111043.

4. MATERIALS AND METHODS:-

- **Materials :-** Extract of Soyabean seeds, Apples, Ginger, Potatoes and Saliva sample solution were prepared and used.

- **Chemicals and Reagent:-** DNS(Dinitrosalicyclic acid) reagent, 5% Starch solution, 0.5% NaCl solution, 0.5N NaOH, Sodium Acetate Buffer (pH= 4, 4.4, 4.6, 4.7, 5) were used.

- **Instruments :-**

a. UV – Visible spectrophotometer:
Make : SHIMADZU
Model no. : UV – 1800-shimadzu

b. PHOTOCOLOROMETRE
Make : ESICO
Model no. : 1313

- **Preparation Of Solution:-**

i. DNS Reagent:- 1gm DNS, 30gm of Sodium potassium tartarate and 1.6gm of NaOH dissolve in water and making up the volume upto 100ml with distilled water.
ii. 5% Starch solution:- 5gm of starch is dissolved in 100ml distilled water.
iii. 0.5% NaCl solution:- 0.5gm of NaCl is dissolved in 100ml distilled water.
iv. 0.5M NaOH:- 2gm of NaOH is dissolved in 100ml distilled water.
v. Acetate Buffer (pH-4.6):- Dissolve 5.4gm of sodium acetate in 50ml of distilled water, add 2.4ml of glacial acetic acid and dilute with distilled water to 100ml. Adjust the pH, if necessary.

- **Preparation Of Buffer Solution:-**

i. Acetate Buffer pH 4:-
Place 0.286 ml of glacial acetic acid and 0.1 ml of a 5% w/v solution of sodium hydroxide in 100 ml volumetric flask, add distilled water to volume and mix. Adjust the pH, if necessary.
ii. Acetate Buffer pH 4.4:-
Dissolve 13.6 gm of sodium acetate and 7.7 gm of ammonium acetate in distilled water and dilute with distilled water to 100 ml. Add 25 ml of glacial acetic acid and mix.
iii. Acetate Buffer pH 4.6:-
Dissolve 5.4 gm of sodium acetate in 50 ml of distilled water, add 2.4 ml of glacial acetic acid and dilute with distilled water to 100 ml. Adjust the pH, if necessary.
iv. Acetate Buffer pH 4.7:-
Dissolve 0.84 gm of sodium acetate and 0.335 ml of glacial acetic acid in sufficient distilled water to produce 100 ml. Adjust the pH, if necessary.
v. Acetate Buffer pH 5:-
Dissolve 1.36 gm of sodium acetate and 0.6 ml of glacial acetic acid in sufficient distilled water to produce 100 ml. Adjust the pH, if necessary.

- **Preparation of Enzyme Extract:-**

a. Enzyme extract from potato:-
i. Take 2-3 potatoes. Use cold water to rinse these off. The goal is to remove surface dirt.
ii. Peel the raw potatoes by using peeler and cut into small pieces. Grind them in a mortar and pestle with sufficient water.
iii. Collect the potatoes homogenate into a beaker and add enough water.
iv. Then filter the homogenate through a muslin cloth to remove the particles. Allow the filtration to settle. Starch rapidly settles at the bottom.
v. Collect the starch free supernatant solution carefully.
vi. Again filter it by using buchner funnel.

b. Enzyme extract from soyabean seeds:-
i. Take handful of soyabean seeds, washed with distilled water and kept immersed overnight.
ii. Grind the soaked seeds in a mortar and pestle with sufficient water.
iii. Collect the homogenate into a beaker and add enough water.
iv. Then filter the homogenate through a muslin cloth to remove the particles. Uniform slurry was prepared.
v. After preparing the slurry, filter it by using buchner funnel.

c. Enzyme extract from ginger:-
i. Take a ginger and use cold water to rinse these off.
ii. Peel the raw ginger by using peeler and cut into small pieces. Grind them in a mortar and pestle with sufficient water.
iii. Collect the potatoes homogenate into a beaker and add enough water.
iv. Then filter the homogenate through a muslin cloth to remove the particles.
v. Collect the slurry in a beaker and filter it by using buchner funnel.

d. Enzyme extract from apple:-
i. Take 2-3 apples, wash the apples with water. The goal is to remove pesticides and surface dirt.
ii. Peel the apples by using peeler and cut into small pieces. Grind them in a mortar and pestle with sufficient water and turned into apple mash.

iii. Collect the apples homogenate into a beaker and add enough water.
iv. Then filter the homogenate through a muslin cloth to remove the particles.
v. Collect the slurry in a beaker and again filter it by using buchner funnel to obtain uniform slurry.

e. Collection and preparation of saliva solution.

i. Rinse your mouth with clean water. Tale a mouthful of water and hold it in your mouth for two minutes, moving it with the tongue. Collect it in a beaker.

➢ **Procedure For Determining The Effect Of pH On Enzyme Activity:-**

i. Prepare series of test tube containing 1ml starch solution, 1ml pH (different pH), 1ml 0.5% NaCl solution and 1ml of enzyme extract.
ii. Incubate all test tube at 37°C for 15min.After incubation add 1ml DNS reagent and 0.5N NaOH.
iii. Heat all the test tube in boiling water for 10mins and allow it to cool and dilute 10 times with distilled water.
iv. Mix the content thoroughly and take absorbance at 540nm.
v. Plot the graph of pH v/s absorbance.

5. OBSERVATION TABLE

Table No. 1. Enzyme Extract From Potato.

Test Tube	Starch (Ml)	Buffer (Each Of 1 Ml)	Nacl (Ml)	Enzyme (Ml)	Dns (Ml)	0.5 N Naoh (Ml)	Absorbance
1	1	Ph 4	1	1	1	1	0.115
2	1	Ph 4.4	1	1	1	1	0.276
3	1	Ph 4.6	1	1	1	1	0.008
4	1	Ph 4.7	1	1	1	1	O.095
5	1	Ph 5	1	1	1	1	0.001

Table No. 2. Enzyme extract from Soyabean Seeds.

Test Tube	Starch (ml)	Buffer (each of 1 ml)	NaCl (ml)	Enzyme (ml)	DNS (ml)	0.5 N NaOH (ml)	Absorbance
1	1	PH 4	1	1	1	1	0.876
2	1	PH 4.4	1	1	1	1	0.036
3	1	PH 4.6	1	1	1	1	0.546
4	1	PH 4.7	1	1	1	1	O.619
5	1	PH 5	1	1	1	1	0.013

Table No. 3. Enzyme extract from Ginger.

Test Tube	Starch (ml)	Buffer (each of 1 ml)	NaCl (ml)	Enzyme (ml)	DNS (ml)	0.5 N NaOH (ml)	Absorbance
1	1	PH 4	1	1	1	1	0.876
2	1	PH 4.4	1	1	1	1	0.036
3	1	PH 4.6	1	1	1	1	0.546
4	1	PH 4.7	1	1	1	1	O.619
5	1	PH 5	1	1	1	1	0.013

Table No. 4. Enzyme extract from Apple.

Test Tube	Starch (ml)	Buffer (each of 1 ml)	NaCl (ml)	Enzyme (ml)	DNS (ml)	0.5 N NaOH (ml)	Absorbance
1	1	PH 4	1	1	1	1	0.621
2	1	PH 4.4	1	1	1	1	0.113
3	1	PH 4.6	1	1	1	1	0.543
4	1	PH 4.7	1	1	1	1	O.754
5	1	PH 5	1	1	1	1	0.717

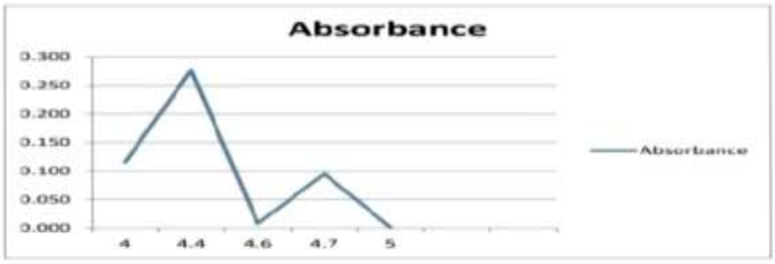
Table No. 5. Enzyme extract from Saliva Solution.

Test Tube	Starch (ml)	Buffer (each of 1 ml)	NaCl (ml)	Enzyme (ml)	DNS (ml)	0.5 N NaOH (ml)	Absorbance
1	1	PH 4	1	1	1	1	0.106
2	1	PH 4.4	1	1	1	1	0.308
3	1	PH 4.6	1	1	1	1	0.214
4	1	PH 4.7	1	1	1	1	O.195
5	1	PH 5	1	1	1	1	0.005

6. RESULTS

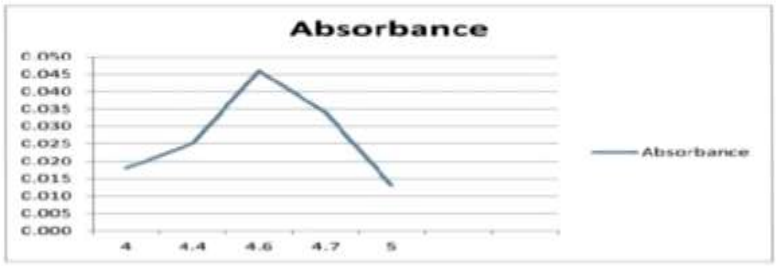
Enzyme Extract Potato

pH	Absorbance
4	0.115
4.4	0.276
4.6	0.008
4.7	0.095
5	0.001



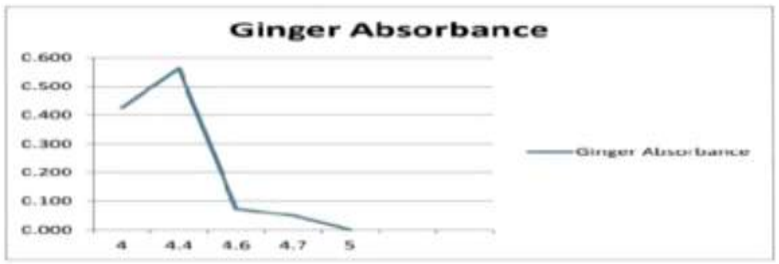
Enzyme Extract Soyabean

pH	Absorbance
4	0.018
4.4	0.025
4.6	0.046
4.7	0.034
5	0.013



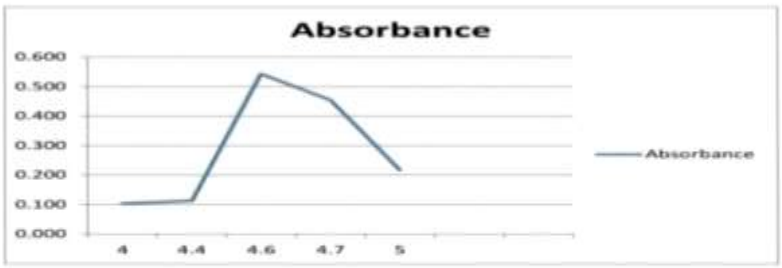
Enzyme Extract Ginger

pH	Absorbance
4	0.426
4.4	0.563
4.6	0.073
4.7	0.049
5	0.001



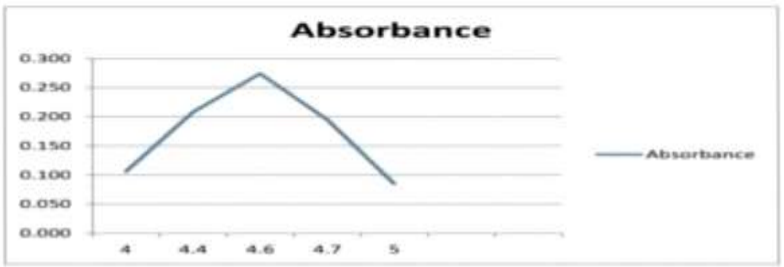
Enzyme Extract Apple

pH	Absorbance
4	0.104
4.4	0.113
4.6	0.543
4.7	0.454
5	0.217



Enzyme Extract Saliva

pH	Absorbance
4	0.106
4.4	0.208
4.6	0.274
4.7	0.195
5	0.085



7. DISCUSSION

The present investigation was carried on to check the effect of pH on enzyme Activity. The activity of enzyme was determined after incubation of the crude enzyme Preparation, during assay conditions, under different PH ranging from 4 to 5.

The results obtained in **Graph.1:** while plotting the graph of absorbance against PH of enzyme extract of potato, showed that a gradual increase in enzyme activity from 4 to 4.6.The optimum PH for maximum enzyme activity was recorded at 4.4.Beyond this range there is decline in the activity at 4.6.

The activity of crude enzyme extract from soybean seeds at PH variations is Shown in **Graph.2:** It shows the optimum PH at 4.6.Gradual increase in enzyme Activity from 4 to 4.6,it shows least enzyme activity at 4 and maximum activity at 4.6.As the PH rises more than optimum PH 4.6 to 5,the enzyme Activity gets lowers. The protein nature of enzyme makes them extremely sensitive; so after Optimal PH the rate starts to drop that is because the protein starts to denature so It loses its shape and don't function well.

The results obtained in **Graph 3:** while plotting the graph of absorbance against PH Of enzyme extract of ginger, it shows the optimum PH at 4.4.Till 4.4 it was In increasing order and at 4.4 it has highest enzyme activity and after 4.4 it is in Declining phase, because the enzyme activity gradually lowers as the PH rises more than the optimum PH.

The results obtained in **Graph 4:** while plotting the graph of absorbance against PH Of enzyme extract of apple, showed that a gradual increase in enzyme activity from 4 to 4.6. The optimal PH for maximum enzyme activity was recorded at 4.6.Beyond this range there is decline in the activity at 5 .

The results obtained in **Graph 5:** while plotting the graph of absorbance against PH Of saliva. The enzyme activity is slowly increased namely at PH 4 to 4.6.The optimal PH for maximum enzyme activity was recorded at 4.6.These Results indicate the sensitive nature of the enzyme. Increasing the reaction incubation PH namely from 4.6 to 5 led to exponential decrease in enzymatic activity. These results collectively indicated that the enzyme is a sensitive and showed a Complete stability at 4.6 .

8. CONCLUSION

Enzyme activity is at its maximum value at the optimum pH. As the pH value is increased above or decreased below the optimum pH the enzyme activity decreases.All enzymes have an ideal pH value, which is called optimal pH. Under the optimum pH conditions, each enzyme showed the maximum activity. For example, the optimum pH of an enzyme that works in the acidic environment of the human stomach is lower than that of an enzyme that works in a neutral environment of human blood. When the pH value deviates from the ideal conditions, the activity of the enzyme slows down and then stops. The enzyme has an active site at the substrate binding site, and the shape of the active site will change with the change of pH value. Depending on the extreme extent of the enzyme and pH changes, these changes may permanently "destroy" the enzyme, or once the conditions return to the desired range of the enzyme, the enzyme will return to normal.PH not only affects the activity of the enzyme, but also affects the charge and shape of the substrate, so that the substrate cannot bind to the active site, or cannot be catalyzed to form a product. In a narrow range of pH, the structural and morphological changes of enzymes and substrates may be reversible. However, if the level of pH changes significantly, the enzyme and substrate may be denatured. In this case, the enzyme and the substrate do not recognize each other, so there will be no reaction.The rate of enzymatic reaction depends on pH of the medium. Each enzyme has an optimum pH, where the rate of enzymatic reaction is maximum. At higher or lower pH, the rate of an enzymatic reaction decreases. For most enzymes, the optimum pH lies in the range from pH 5 to pH 9.There are several factors that affect the speed of an enzyme's action, such as the concentration of the enzyme, the concentration of the substrate, temperature, hydrogen ion concentration (pH), and the presence of inhibitors.Because enzymes are proteins, their molecular structure is determined by interactions between the charges of the amino acids that compose the protein chains. These interactions take the form of hydrogen bonds, which are altered by pH. These positive charges impact the charges of the amino acids inside the protein, making the enzyme more or less active depending on the enzyme's optimum pH.

9. BIBLIOGRAPHY

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