Acta Scientific Pharmaceutical Sciences (ISSN: 2581-5423)

Volume 8 Issue 4 April 2024

Research Article

Determination and Comparison of Antioxidant Activity of *Phyllanthus emblica*, *Ziziphus mauritiana*, *Mangifera indica* Samples Collected Locally from Banshkhali, Chattogram, Bangladesh

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DOI: 10.31080/ASPS.2024.08.1046

Received: February 27, 2024

Published: March 07, 2024

Abstract

Fruits and vegetables are an ample source of a vital group of compounds named antioxidants, which are crucial for maintaining sound health. Although antioxidants are prolifically available in almost every fruit and vegetable species, the levels of antioxidants in these species may vary depending on the location and on different climate conditions, even if grown in the same soil. The goal of this research is to quantify the total antioxidant content of some locally available fruits and compare them. Samples were collected from three different locations. Mango (*Mangifera indica*), Indian gooseberry (*Phyllanthus emblica*) and Indian jujube (*Ziziphus mauritiana*) was selected for this research as they are readily available and cheap in price. Methanolic extracts of these samples were used for the analysis. To determine the antioxidant capacity, DPPH assay was used. This research also reveals same sample shows different activity level in different place in the same region due to geographical distribution and climate conditions.

Keywords: Antioxidant Capacity; Methanolic Extracts; DPPH Assay; Indian Gooseberry; Indian Jujube; Mango

Introduction

To maintain sound health, we need fruits and vegetables in our daily diet. Fruits contain different phytochemicals and vitamins that are important for our physiological operations to maintain a sound health. Fruits can prevent many serious diseases like cardiovascular disease, diabetes, obesity, etc. It can be clearly noticed if any person takes adequate number of fruits every day because fruits are directly connected with sound health. Fruits are main sources of antioxidants that prevent cancer and cardiovascular diseases. Scientific publications firmly suggest the taking of phenolic component rich food for the prevention of degenerative diseases, such as cardiovascular, cancer, Alzheimer, diabetes, and neurodegenerative diseases. Consequently, there is anintensivesearch for plant sources rich in phenolics for maintaining sound health. Antioxidants interrupts oxidation of lipids or other molecules by disturbing the commencing of oxidizing chain events in situations when redox reactions are essential for biological functions. Free radicals are considered sometimes important for normal physiological functions, but they are surely disastrous when produced in excess. Superoxide anion radicals(O_2^-), hydroxyl radicals (-OH) and non-free radical species such as hydrogen peroxide (H_2O_2) and singlet oxygen ($^1O_2$) are examples of ROS. Two sources of free radicals, exogenous which is non-enzymatic reaction, endogenous, which is formed from cellular and intracellular reactions. Many dangerous diseases like cancer, Parkinson's disease etc. are directly linked to ROS. Free radicals react with DNA bases and damages the DNA which ultimately leads to cancer. Although many compounds likeα-tocopherol, catalase, glutathione peroxidase, Superoxide dismutase act as an antioxidant defense system and reduce the cell damage caused by free radicals, these multiple defense systems also fail due to increased production of ROS or decreased level of cellular antioxidants.

So, it is very important to find external sources of antioxidants. To balance between ROS and antioxidant level, natural fruits are the most viable option. Oxidative stress is known as the imbalance between level of antioxidants and oxidants in the body. Fruits contain exogeneous antioxidants which reduce the level of ROS. These antioxidants lower the ROS and disrupt the beginning of reactive oxygen species. Electron donation or metal ion chelation is used by antioxidants to achieve this. Some prime antioxidants are Vitamins C and E, carotenoids, and phenolic chemicals, particularly flavonoids. Phenolic compounds are naturally more antioxidant than vitamin and carotenoid. Phenols disturb the ROS by their electron donation nature. The factor upon which the antioxidant performance of phenol depend is the environment where the phenol is working and the location of the hydroxyl groups on the phenol. Different fruits antioxidants have different performance level, so it would be advisable to consume a range of them.

Usually, locally available fruits are consumed by local peoples in their daily diet. To my knowledge, no data is available about their composition, and it hasmotivated me to carry out this research. The species are *Mangifera indica*, *Phyllanthus emblica*, *Ziziphus mauritiana*.

Materials and Methods

Chemicals and reagents

Methanol (99.99%), Distilled Water (DW), Trolox (Sigma Aldrich, Germany), 2,2-Diphenyl-1-Picrylhydrazyl (Sigma Aldrich, Germany).

Raw materials collection

Fruits that are produced locally were collected for this analysis. Three seasonal fruits were collected from three village of Banshkhali, Chattogram, Bangladesh. Banigram, Hazigaon, Chapachori were selected as sampling sites based on their distance from river Shangu and the Bay of Bengal. Chapachori is adjacent to the Bay of Bengal, whereasHazigaon is close to river Shangu and Banigram is in between these two.

The three different fruit, namely Indian gooseberry, Indian jujube and Mango were collected. Samples collected from Banigram were leveled as IG-1, IJ-1 and MI-1. Which was IG-2, IJ-2 and MI-2 for Hazigaon and IG-3, IJ-3 and MI-3 for Chapachori.

Gooseberry was collected in the month of September. For I. Jujube it was the month of January and For Mango it was the month of March. These fruits were all collected in their unripe stage.

Preparation of fruit extracts

One of the most crucial steps in the DPPH assay is the preparation of fruit extract. This method involves several steps.

Washing of samples

First, to eliminate dirt, freshly gathered fruits that were disease-free were washed three to four times with tap water. To prevent contamination, they were then rinsed with double-distilled water (DI water).

Sample size reduction

The drying process was then facilitated by cutting fruit sam Antioxidant activity and total phenolic content of some indigenous fruits of Bangladesh ples into tiny, thin pieces. To prevent any contamination that might affect the results, a sterilized knife was utilized. Comparing the slices of Indian gooseberry and Indian jujube, Mango was substantially larger. Before being cleaned and chopped, samples were kept in the freezer.

Drying and grinding

The samples, which had previously been cut into little pieces, are oven dried at 60 °C for however long it takes to entirely remove moisture. Indian gooseberry and Mango took around 24 hours and Indian jujube took around 36 hours to dry completely. After the drying process, the samples were grinded in a clean blender to convert them into very fine powder and stored in the sample bottle.

Mixing with methanol and filtration

Extracts were prepared from this powder by mixing it with methanol. For extraction preparation [1], were followed with a slight modification.5g of finely grinded sample was mixed with 25mL methanol in a 50mL beaker.

The solution was then mixed in a magnetic stirrer for 1 hour at 40°C and 650rpm. This temperature has been maintained very strictly so that methanol doesn't evaporate during this process. After that, the mixture was filtered under suction through filter paper. The filtrate was reserved, and fresh 25mL methanol was added to the residue. The extraction and filtration procedure was repeated thrice. The filtrates were combined and stored.

This method was used for the preparation of all three samples. Filtrate showed different color and volume was different for each sample.

Evaporation in rotary evaporator

Combined filtrates were evaporated in a vacuum rotary evaporator under reduced pressure at 40°C for 4-5 hours. As the solution becomes thick, the rateof evaporation decreases. Afterwards, the concentrates were dried and weighed to determine total extractable compounds. Finally, the extracts were stored in a refrigerator at 4°C until used [1].

Weighing of extracts and storage

The extracts are collected, weighted, and frozen at 4°C after evaporating under pressure in the rotary evaporator.

Antioxidant potential evaluation

The 2,2-Diphenyl-1-Picrylhydrazyl (DPPH) radical test is commonly used to measure the antioxidant capacity of a substance. In this work, the DPPH test was utilized to assess the antioxidant capacity of the samples [2].

Preparation of DPPH solution

To prepare 1mM DPPH solution, 0.004gm DPPH was dissolved in 100mL methanol. The solution was kept in a dark room [3]. DPPH reactions are very sensitive to reaction media e.g., water, pH, DO, light exposure, etc. The absorbance of DPPH at 517nm is decreased by light [4].

Preparation of Trolox standard solution

0.001gm of Trolox was dissolved in 10mL methanol to prepare a 100 g/mL stock solution. To prepare 10g /ml to 60g /ml solution, 50L, 100 L, 150 L, 200 L, 250 L, 300 L stock solution was taken and add 450 L, 400 L, 350 L, 300 L, 250 L, 200 L methanol were respectively. 1.5 mL methanol and 1.5mL DPPH solution were used as a blank/control solution.1.5 mL methanol and 1.5mL DPPH solution were used as a blank/control solution [5].

Preparation of fruit extract solution

Different concentrations of extract solution were prepared for different fruit samples. For Indian gooseberry, 50 µg/mL stock solution was prepared by dissolving 0.0025 µg Indian gooseberry powder in 50mL methanol solution. Allowing it to dissolve properly with occasional shaking. Then it was diluted to 1 µg/mL, 2 µg/mL, 3 µg/mL, 4 µg/mL, 5 µg/mL, 6 µg/mL, 7 µg/mL, 8 µg/mL and 9 µg/mL 10 µg/mL.

For Indian jujube, 500 µg/mL stock solution was prepared by dissolving 0.005 µg Indian jujube powder in 50mL methanol solution. Allowing it to dissolve properly with occasional shaking. Then it was diluted to 100 µg/mL, 150 µg/mL, 200 µg/mL, 250 µg/mL, 300 µg/mL, 350 µg/mL, 400 µg/mL and 450 µg/mL.

For Mango, 250 µg/mL stock solution was prepared by dissolving 0.005 µg Mango powder in 50mL methanol solution. Allowing it to dissolve properly with occasional shaking. Then it was diluted to 50 µg/mL, 75 µg/mL, 100 µg/mL, 125 µg/mL, 150 µg/mL, 175 µg/mL, 200 µg/mL and 225 µg/mL.

Antioxidant activity

DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) radical scavenging activity

The free radical scavenging activity was measured by the 2-2-diphenyl-1-picryl-hydrazyl (DPPH) following the method. 1.5mL of DPPH solution was mixed with 1.5mL of each concentration (Trolox or Fruit extracts) solution and the mixture was then vortexed [6]. Here, Trolox was used as standard. After vertexing, the solution mixture was kept in a dark room for 30minutes. A blank solution containing all reagent (without Trolox or fruit extracts) solution was also taken.

The absorbance of the solution was measured at 517nm against a blank (methanol) using UV-Vis spectrophotometer. The percentage of inhibition capacity was calculated from the following formula:

$$\text{Percentage of Scavenging} = \frac{A_0 - A_1}{A_0} \times 100$$

Where,

A_0→Absorbance of the control

A_1→Absorbance of the Trolox\/fruit extract solution

Percentage of scavenging was plotted against concentration and from this curve value of IC_{50} was calculated [7].

Result and Discussion

Evaluation of antioxidant activity

At various concentrations, the antioxidant activity of fruit extracts produced using Indian gooseberry, Indian jujube and Mango was detected. The radical scavenging activity of fruit extracts was measured spectrophotometric ally by the color change of DPPH from purple to pale yellow [7]**.** Initially, the color of the solution was purple. The DPPH solution became pale yellow during incubation.

The characteristic is caused by the presence of functional groups on the surface of fruit extracts (Figure 20). The DPPH radical is a long-lived, deep purple, organic nitrogen radical. Antioxidants convert the radical equivalent of the purple chromogen to hydrazine [8].

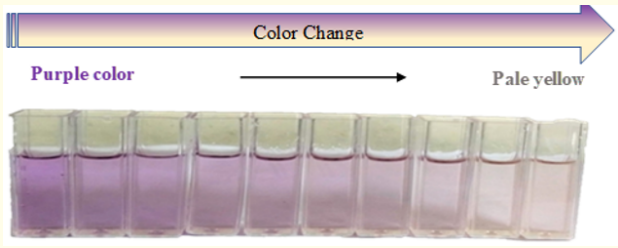

Figure 1: Changes of color of DPPH solution after 30 minutes incubation.

Calibration curve for trolox antioxidant

Trolox is an antioxidant that is a water-soluble vitamin E homologue. When administered to a biological system, it decreases oxidative stress. To determine the antioxidant activity of any substance, a standard may be utilized [3]**.**

The IC_{50} value of Trolox was determined to be 12.07 µg/mL. To compute the concentration of different fruit samples, that induced 50% DPPH radical scavenging ability (IC_{50}), the findings were fitted to the equation.

Data tables and findings for Indian Gooseberry

Data tables and findings for three Indian Gooseberry samples leveled as IG-1, IG-2 and IG-3 are shown below.

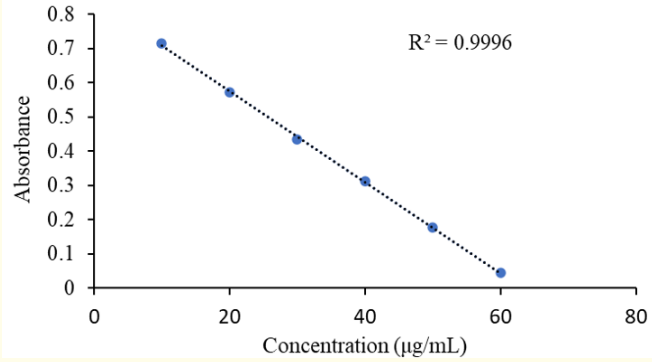

Figure 2: Standard curve for Trolox.

Table 1 displays the absorbance of IG-1 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity [9]**.**

Sample Concentration	Absorbance				Inhibition Concentration (%)
	Day-1	Day-2	Day-3	Average	
Control	0.501	0.524	0.519	0.515	
1	0.429	0.441	0.431	0.434	15.73
2	0.381	0.376	0.374	0.377	26.80
3	0.312	0.321	0.318	0.317	38.45
4	0.249	0.252	0.255	0.252	51.07
5	0.211	0.211	0.213	0.212	58.83
6	0.170	0.168	0.171	0.169	67.18
7	0.134	0.133	0.135	0.133	74.17
8	0.095	0.096	0.095	0.095	81.55
9	0.078	0.076	0.076	0.077	85.05
10	0.058	0.056	0.059	0.058	88.74

Table 1: % of Inhibition of IG-1 extracts in different concentration.

The findings were fitted to the equation y = 8.2302x + 13.491, R^2 = 0.9698. The IC_{50} value of IG-1 extracts was determined to be 4.44 µg/mL (Figure 3). Therefore, at a concentration of 4.44 µg/mL, IG-1 extracts exhibited 50% scavenging activity.

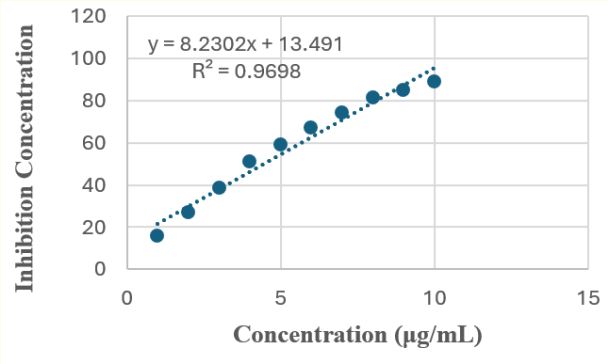

Figure 3: IC_{50} determination curve of IG-1 extracts.

Table 2 displays the absorbance of IG-2 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y = 8.1362x + 16.975, R^2 = 0.9563. The IC_{50} value of IG-2 extracts was determined to be 4.06µg/mL (Figure 4). Therefore, at a concentration of 4.06µg/mL, IG-2 extracts exhibited 50% scavenging activity.

Sample Concentration (µg/mL)	Absorbance				Inhibition Concentration (%)
	Day -1	Day -2	Day – 3	Average	
Control	0.532	0.557	0.564	0.551	
1	0.461	0.466	0.465	0.464	15.79
2	0.378	0.375	0.369	0.374	32.12
3	0.311	0.310	0.314	0.312	43.38
4	0.260	0.258	0.258	0.258	53.18
5	0.207	0.205	0.208	0.207	62.42
6	0.162	0.162	0.164	0.162	70.60
7	0.122	0.120	0.122	0.121	78.04
8	0.084	0.089	0.085	0.086	84.40
9	0.070	0.072	0.072	0.071	87.11
10	0.054	0.055	0.052	0.054	90.20

Table 2: % of Inhibition of IG-2 extracts in different concentration.

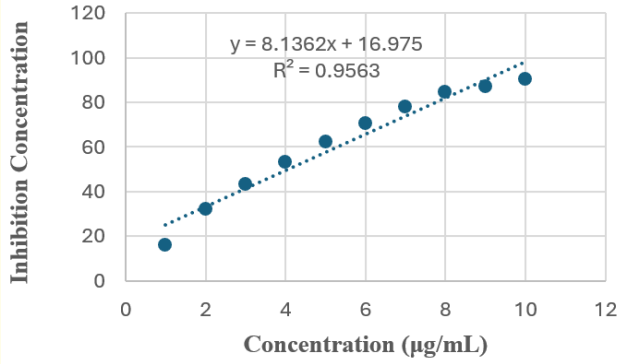

Figure 4: IC_{50} determination curve of IG-2 extracts.

Table 3 displays the absorbance of IG-3 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y = 8.0745x + 0.1033, R^2 = 0.9525. The IC_{50} value of IG-3 extracts was determined to be 6.18 µg/mL (Figure 5). Therefore, at a concentration of 6.18 µg/mL, IG-3 extracts exhibited 50% scavenging activity.

Sample Concentration (µg/mL)	Absorbance				Inhibition Concentration (%)
	Day-1	Day-2	Day-3	Average	
Control	0.520	0.528	0.526	0.525	
1	0.494	0.501	0.497	0.497	5.33
2	0.470	0.472	0.469	0.470	10.48
3	0.425	0.423	0.422	0.423	19.43
4	0.325	0.320	0.322	0.322	38.67
5	0.279	0.278	0.279	0.279	46.85
6	0.234	0.235	0.235	0.235	55.24
7	0.208	0.207	0.209	0.208	60.38
8	0.182	0.181	0.180	0.181	65.52
9	0.160	0.159	0.159	0.159	69.71
10	0.139	0.138	0.140	0.139	73.52

Table 3: % of Inhibition of IG-3 extracts in different concentration.

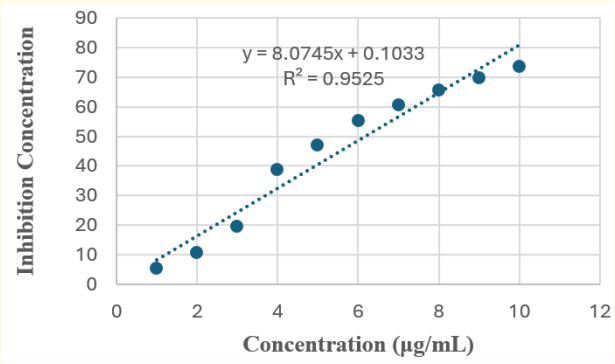

Figure 5: IC_{50} determination curve of IG-3 extracts.

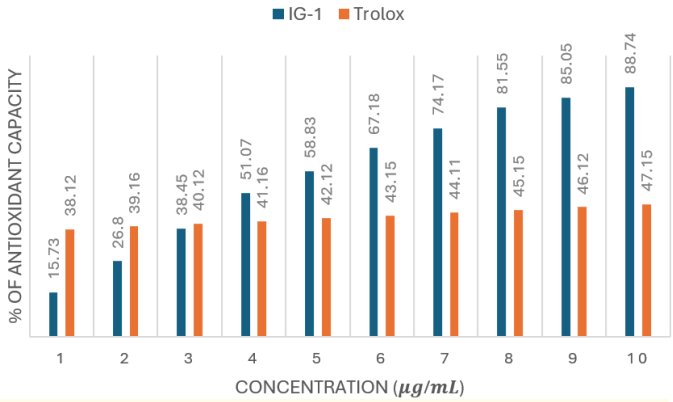

Figure 6: DPPH assay of IG-1 and Trolox.

Scavenging capacity of IG-1 extracts

Figure 6 depicts a comparison of the DPPH-scavenging ability of IG-1 extracts and Trolox at various concentrations. The results demonstrated that IG-1 extracts possess efficient radical scavenging capability when compared to the standard (Trolox). For Trolox 47.15% is the highest radical scavenging activity at 10µg/mL where for IG-1 its 88.74%.

Scavenging Capacity of IG-2 extracts

Figure 7 depicts a comparison of the DPPH-scavenging ability of IG-2 extracts and Trolox at various concentrations. The results demonstrated that IG-2 extracts possess efficient radical scavenging capability when compared to the standard (Trolox). For Tro-

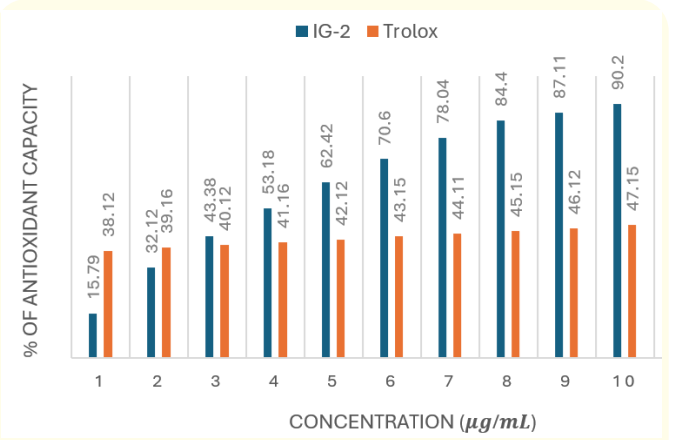

Figure 7: DPPH assay of IG-2 and Trolox.

lox 47.15% is the highest radical scavenging activity at 10 µg/mL where for IG-2 its 90.2%.

Scavenging Capacity of IG-3 extracts

Figure 8 depicts a comparison of the DPPH-scavenging ability of IG-3 extracts and Trolox at various concentrations. The results demonstrated that IG-3 extracts possess efficient radical scavenging capability when compared to the standard (Trolox). For Trolox 47.15% is the highest radical scavenging activity at 10 µg/mL where for IG-1 its 73.52%.

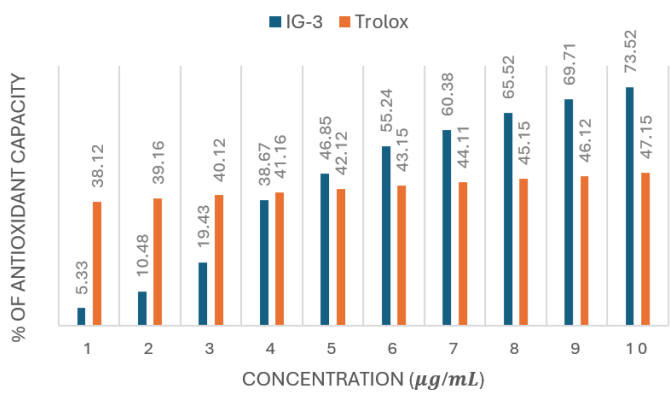

Figure 8: DPPH assay of IG-3 and Trolox.

Data tables and findings for indian jujube

Data tables and findings for three Indian Jujube samples leveled as IJ-1, IJ-2 and IJ-3 are shown below.

Table 4 displays the absorbance of IJ-1 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y = 0.1026x + 20.774, R^2 = 0.9597. The IC_{50} value of IJ-1 extracts was determined to be 284.85 µg/mL (Figure 9). Therefore, at a concentration of 284.85 µg/mL, IJ-1 extracts exhibited 50% scavenging activity.

Table 5 displays the absorbance of IJ-2 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y =0.1085x + 11.952, R^2 = 0.9842. The IC_{50} value of IJ-2 extracts was determined to be

Sample Concentration (µg/mL)	Absorbance				Inhibition Concentration (%)
	Day-1	Day-2	Day-3	Average	
Control	0.509	0.505	0.506	0.507	
100	0.377	0.380	0.375	0.377	25.64
150	0.322	0.325	0.324	0.324	36.09
200	0.282	0.283	0.284	0.283	44.18
250	0.258	0.259	0.260	0.259	48.92
300	0.239	0.236	0.240	0.238	53.06
350	0.210	0.208	0.207	0.208	58.97
400	0.189	0.188	0.189	0.188	62.92
450	0.175	0.174	0.176	0.175	65.48
500	0.157	0.160	0.161	0.159	68.64

Table 4: % of Inhibition of IJ-1 extracts in different concentration.

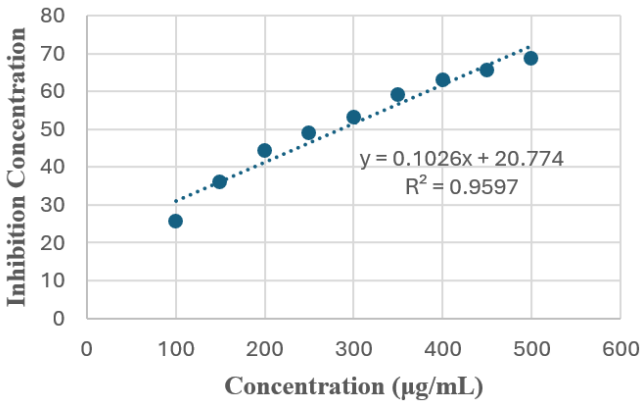

Figure 9: IC_{50} determination curve of IJ-1 extracts.

350.67 µg/mL (Figure 10). Therefore, at a concentration of 350.67 µg/mL, IJ-2 extracts exhibited 50% scavenging activity.

Table 6 displays the absorbance of IJ-3 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y =0.1038x + 16.721, R^2 = 0.975. The IC_{50} value of IJ-3 extracts was determined to be 320.61 µg/mL (Figure 11). Therefore, at a concentration of 320.61 µg/mL, IJ-3 extracts exhibited 50% scavenging activity.

Sample Concentration (µg/mL)	Absorbance				Inhibition Concentration (%)
	Day - 1	Day - 2	Day - 3	Average	
Control	0.512	0.510	0.514	0.512	
100	0.401	0.405	0.407	0.404	21.09
150	0.380	0.375	0.376	0.377	26.37
200	0.335	0.336	0.338	0.336	34.38
250	0.298	0.299	0.301	0.299	41.60
300	0.281	0.282	0.280	0.281	45.12
350	0.242	0.242	0.241	0.242	52.73
400	0.228	0.226	0.224	0.224	55.66
450	0.212	0.212	0.213	0.212	58.59
500	0.181	0.180	0.180	0.180	64.84

Table 5: % of Inhibition of IJ-2 extracts in different concentrations.

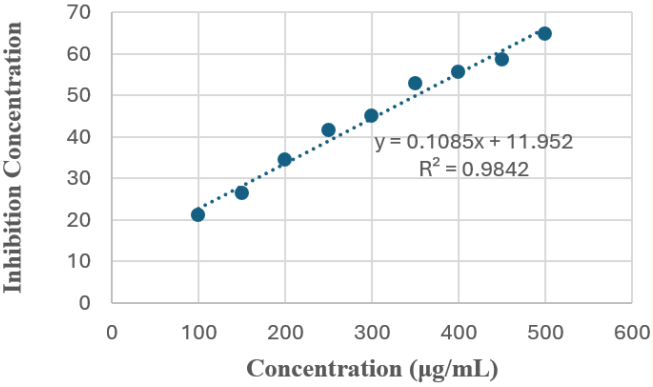

Figure 10: IC_{50} determination curve of IJ-2 extracts.

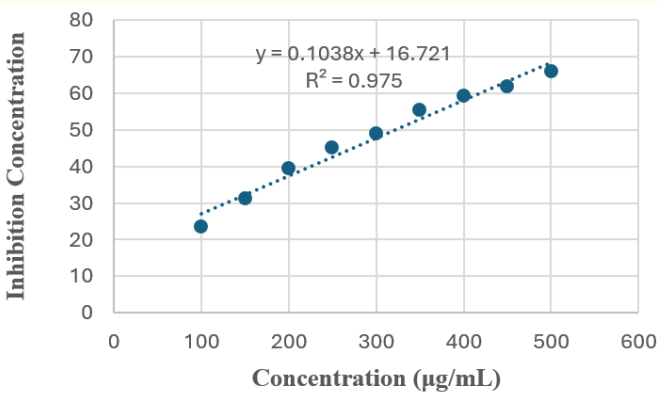

Figure 11: IC_{50} determination curve of IJ-3 extracts.

Sample Concentration (µg/mL)	Absorbance				Inhibition Concentration (%)
	Day - 1	Day - 2	Day - 3	Average	
Control	0.502	0.502	0.503	0.502	
100	0.385	0.384	0.383	0.384	23.51
150	0.342	0.345	0.348	0.345	31.27
200	0.302	0.307	0.304	0.304	39.44
250	0.275	0.278	0.277	0.276	45.02
300	0.256	0.255	0.257	0.256	49.00
350	0.223	0.225	0.224	0.224	55.38
400	0.205	0.204	0.203	0.204	59.36
450	0.190	0.195	0.191	0.192	61.75
500	0.171	0.170	0.173	0.171	65.93

Table 6: % of Inhibition of IJ-3 extracts in different concentrations.

Scavenging Capacity of IJ-1 extracts

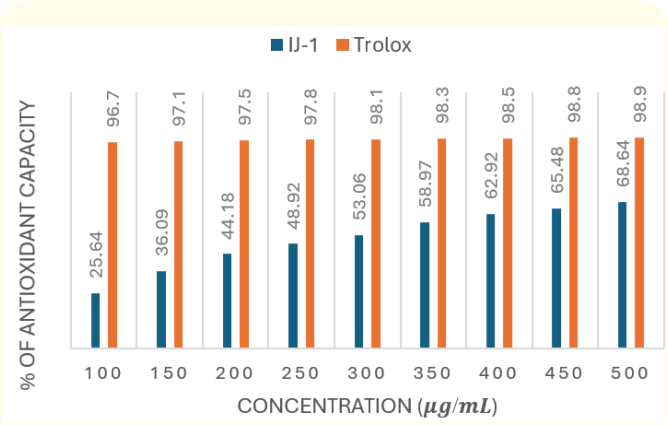

Figure 12: DPPH assay of IJ-1 and Trolox.

Figure 12 depicts a comparison of the DPPH-scavenging ability of IJ-1 extracts and Trolox at various concentrations. The results demonstrated that IJ-1 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 98.9% is the highest radical scavenging activity of Trolox at 500 µg/mL where for IJ-1 its 68.4%.

Scavenging Capacity of IJ-2 extracts

Figure 13 depicts a comparison of the DPPH-scavenging ability of IJ-2 extracts and Trolox at various concentrations. The results demonstrated that IJ-2 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 98.9% is the highest radical scavenging activity of Trolox at 500 µg/mL where for IJ-2 its 68.84%.

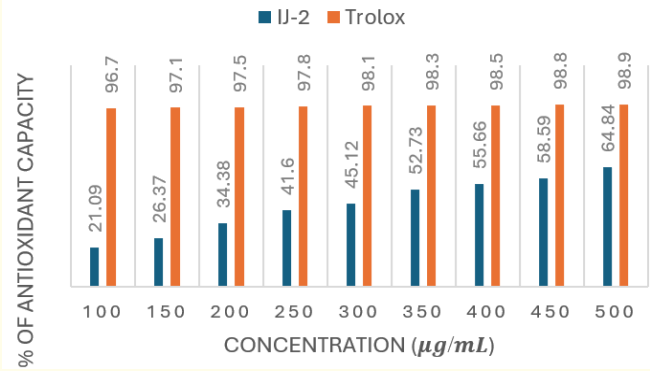

Figure 13: DPPH assay of IJ-2 and Trolox.

Scavenging Capacity of IJ-3 extracts

Figure 14 depicts a comparison of the DPPH-scavenging ability of IJ-3 extracts and Trolox at various concentrations. The results demonstrated that IJ-3 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 98.9% is the highest radical scavenging activity of Trolox at 500 µg/mL where for IJ-3 its 65.93%.

Data tables and findings for *Mangifera Indica*

Data tables and findings for three *Mangifera Indica* samples leveled as MI-1, MI-2 and MI-3 are shown below.

Table 7 displays the absorbance of MI-1 at various reaction mixture concentrations. The absorbance gradually dropped as the

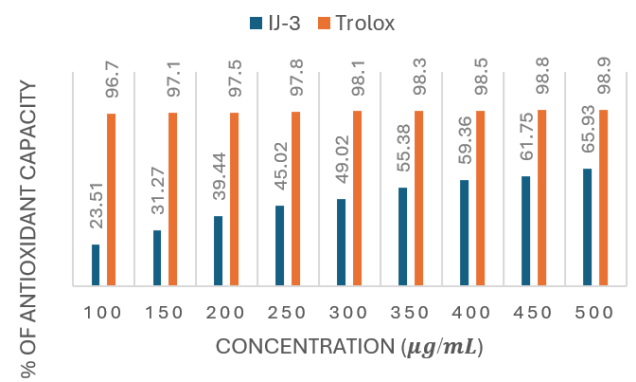

Figure 14: DPPH assay of IJ-3 and Trolox.

concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y =0.2725x + 8.9642, R^2 = 0.9698. The IC_{50} value of MI-1 extracts was determined to be 150.6 µg/mL (Figure 15). Therefore, at a concentration of 150.6 µg/mL, MI-1 extracts exhibited 50% scavenging activity.

Table 8 displays the absorbance of MI-2 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that

Sample Concentration 0	Absorbance				Inhibition Concentration (%)
	Day - 1	Day - 2	Day - 3	Average	
Control	0.501	0.502	0.503	0.502	
50	0.415	0.412	0.417	0.415	17.33
75	0.358	0.355	0.359	0.357	28.88
100	0.312	0.313	0.310	0.312	37.84
125	0.276	0.276	0.275	0.275	45.22
150	0.236	0.234	0.235	0.235	53.18
175	0.205	0.201	0.201	0.202	59.76
200	0.175	0.174	0.175	0.175	65.14
225	0.155	0.154	0.152	0.154	69.32
250	0.140	0.141	0.141	0.141	71.91

Table 7: % of Inhibition of MI-1 extracts in different concentration.

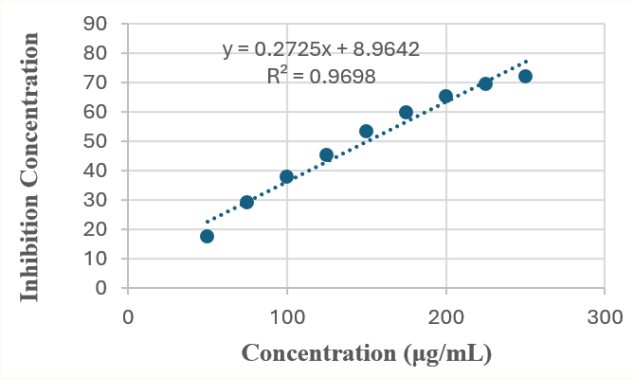

Figure 15: IC_{50} determination curve of MI-1 extracts.

the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

The findings were fitted to the equation y =0.2414x + 4.1783, R^2 = 0.9827. The IC_{50} value of MI-2 extracts was determined to be 189.82µg/mL (Figure 16). Therefore, at a concentration of 189.82 µg/mL, MI-2 extracts exhibited 50% scavenging activity.

Table 9 displays the absorbance of MI-3 at various reaction mixture concentrations. The absorbance gradually dropped as the concentration of the reaction mixture grew. It was observed that the lower absorbance of the reaction mixture indicated a greater proportion of scavenging activity.

Sample Concentration ()	Absorbance				Inhibition Concentration (%)
	Day - 1	Day - 2	Day - 3	Average	
Control	0.520	0.521	0.523	0.521	
50	0.450	0.451	0.452	0.451	13.44
75	0.407	0.407	0.405	0.406	22.07
100	0.378	0.375	0.375	0.376	27.83
125	0.338	0.335	0.334	0.336	35.51
150	0.296	0.295	0.295	0.295	43.78
175	0.267	0.268	0.269	0.268	48.56
200	0.241	0.240	0.241	0.241	53.74
225	0.225	0.224	0.221	0.223	57.19
250	0.201	0.201	0.202	0.201	61.42

Table 8: % of Inhibition of MI-2 extracts in different concentration.

Sample Concentration ()	Absorbance				Inhibition Concentration (%)
	Day-1	Day-2	Day-3	Average	
Control	0.492	0.495	0.497	0.495	
50	0.420	0.422	0.423	0.422	14.75
75	0.376	0.378	0.379	0.378	23.64
100	0.342	0.340	0.341	0.341	31.11
125	0.298	0.299	0.300	0.299	39.60
150	0.271	0.270	0.271	0.271	45.25
175	0.242	0.245	0.247	0.245	50.50
200	0.220	0.221	0.220	0.220	55.55
225	0.197	0.197	0.198	0.197	60.20
250	0.173	0.174	0.173	0.173	65.05

Table 9: % of Inhibition of MI-3 extracts in different concentrations.

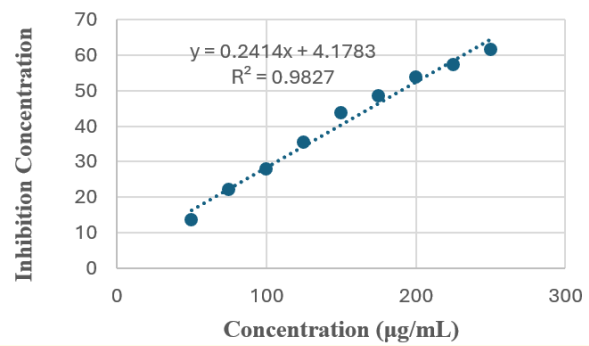

Figure 16: IC_{50} determination curve of MI-2 extracts.

The findings were fitted to the equation y =0.2471x + 5.784, R^2 = 0.9842. The IC_{50} value of MI-3 extracts was determined to be 178.94 µg/mL (Figure 17). Therefore, at a concentration of 174.94 µg/mL, MI-3 extracts exhibited 50% scavenging activity.

Scavenging Capacity of MI-1 extracts

Figure 18 depicts a comparison of the DPPH-scavenging ability of MI-1 extracts and Trolox at various concentrations. The results demonstrated that MI-1 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 97.8%

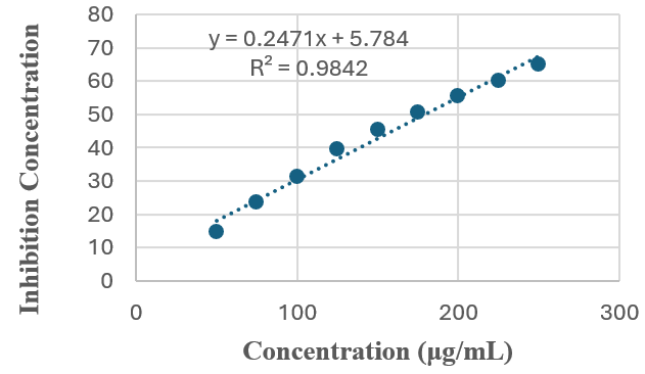

Figure 17: IC_{50} determination curve of MI-3 extracts.

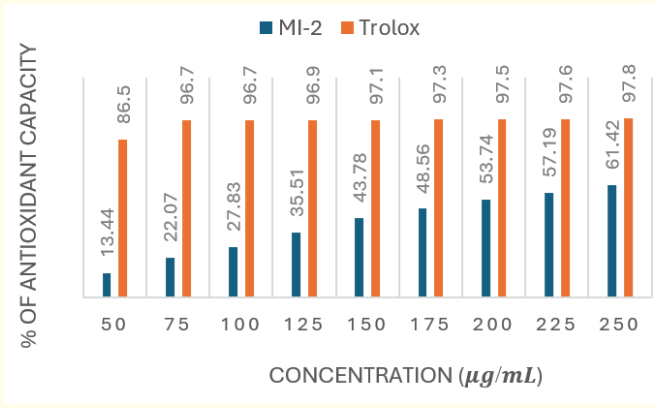

Figure 19: DPPH assay of MI-2 and Trolox.

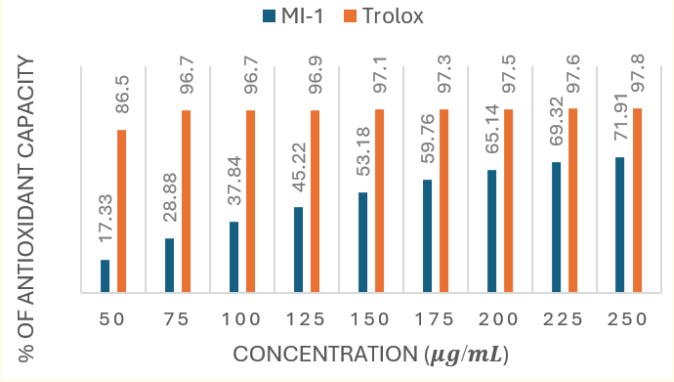

Figure 18: DPPH assay of MI-1 and Trolox.

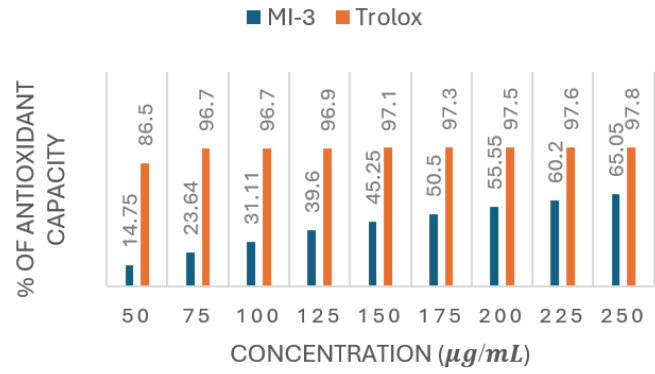

Figure 20: DPPH assay of MI-3 and Trolox.

is the highest radical scavenging activity of Trolox at 250µg/mL where for MI-1 its 71.91%.

Scavenging Capacity of MI-2 extracts

Figure 19 depicts a comparison of the DPPH-scavenging ability of MI-2 extracts and Trolox at various concentrations. The results demonstrated that MI-2 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 97.8% is the highest radical scavenging activity of Trolox at 250 µg/mL where for MI-2 it is 61.42%.

Scavenging Capacity of MI-3 extracts

Figure 20 depicts a comparison of the DPPH-scavenging ability of MI-3 extracts and Trolox at various concentrations. The results demonstrated that MI-3 extracts possess less efficient radical scavenging capability when compared to the standard (Trolox). 97.8% is the highest radical scavenging activity of Trolox at 250µg/mL where for MI-3 it is 65.05%.

Comparison among fruit samples

Despite the fact that all of the fruit samples had significant antioxidant potential, Indian gooseberry has the greatest antioxidant capacity which shows IC_{50} at a concentration of only 4.06 µg/mL and Indian jujube has the lowest which shows IC_{50} at a concentration of 284.85 µg/mL.

The Hazigaon sample, IG-2, has the highest antioxidant capacity of the three Indian gooseberry samples. The IC_{50} value of IG-2 extracts was determined to be 4.06 µg/mL which is much lower that was found in the Sylhet region [10]. TheChapachori sample; IG-3 has the lowest IC_{50} value among the three Indian gooseberry samples which is 6.18 µg/mL.

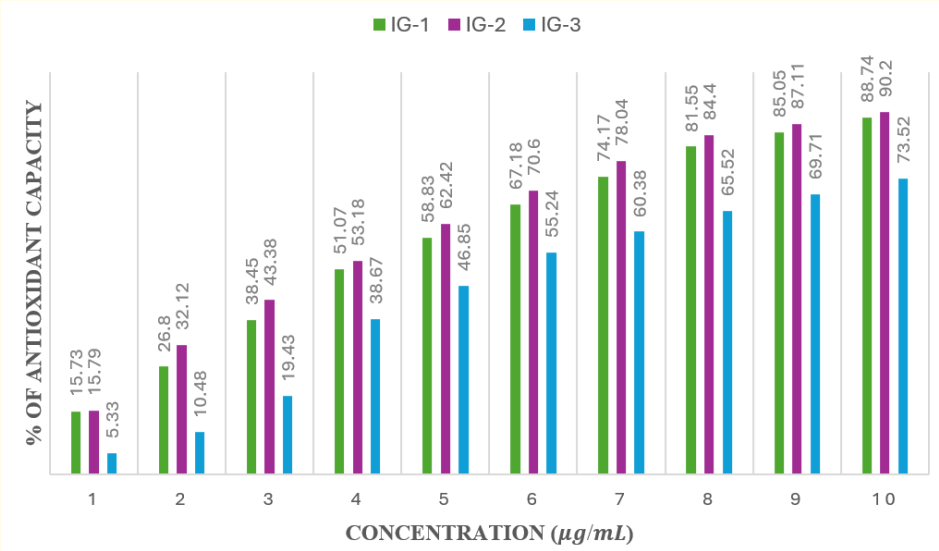

Figure 21: Comparison among the Indian gooseberry samples.

The Banigram sample, IJ-1, has the highest antioxidant capacity of the three Indian jujube samples. The IC_{50} value of IJ-1 extracts was determined to be 284.85 µg/mL which is much lower that was found in the India [11]. The Hazigaon sample, IJ-2, has the lowest IC_{50} value among the three Indian jujube samples which is 350.61 µg/mL.

The Banigram sample, MI-1, has the highest antioxidant capacity of the three Mango samples. The IC_{50} value of MI-1 extracts was determined to be 150.6 µg/mL which is much lower that was found in the Pakistan (Sultana., *et al*. 2012). The Hazigaon sample, MI-2 has the lowest IC_{50} value among the three Mango samples which is 189.82 µg/mL.

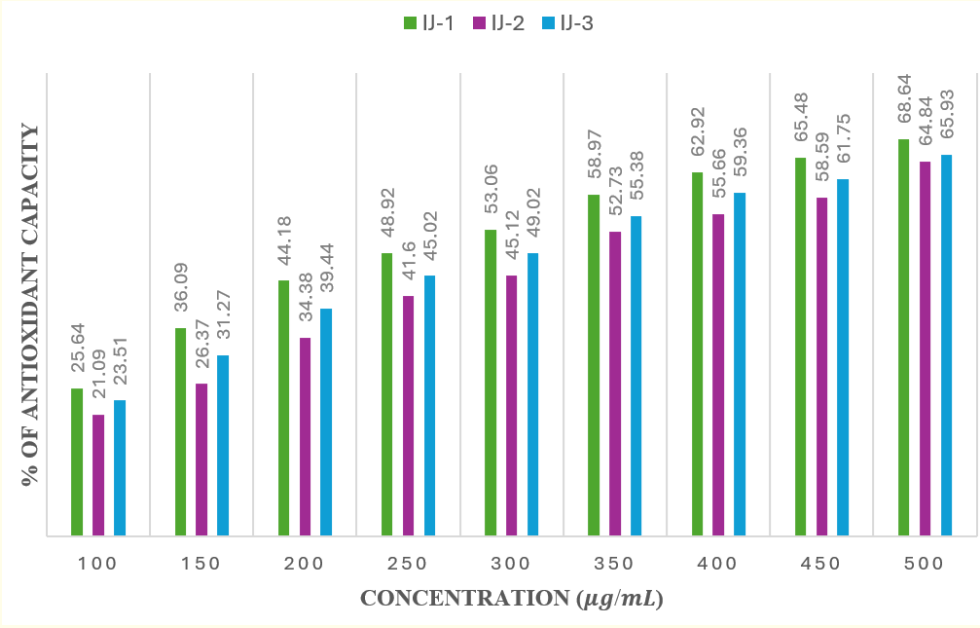

Figure 22: Comparison among the Indian jujube samples.

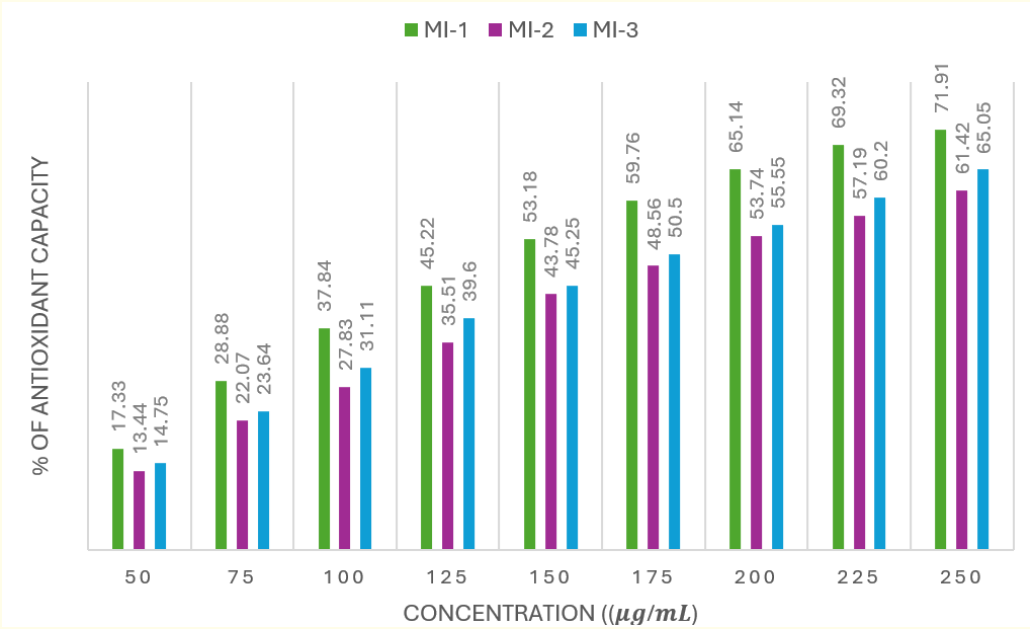

Figure 23: Comparison among the Mango samples.

Conclusion

From this study it is clearly evident that the fruit species are great source of antioxidants, thus readily acts as ROS scavenger. If we consider the scenario in Bangladesh, even average fruits consumption contributes a lot to the demand of antioxidant in the body. However, due to the diversity and complexity of natural mixtures of phenolic compounds in the citrus fruit extracts, it is quite difficult to characterize every compound and compare their antioxidant activities. Each fruit generally contains various phenolic compounds and each of these compounds possesses differing amounts of antioxidant activity [10]. The result of present study shows that Indian gooseberry has the highest antioxidants activity. Although Indian jujube has the lowest antioxidant activity among the three samples, the activity is still high enough to maintain health and disease-free life. As ensuring a healthy life is a must for leading a happy life, the local people should consume these fruits to uptake proper amount of antioxidant in their diet and maintain a sound health.

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