

Research Paper

Investigation of Active Chemical Bulk Present in Solution of Beetroot Juice and Rose water Spectrophotometrically

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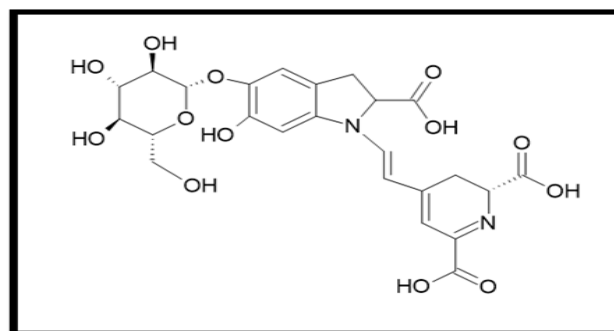
Abstract— Betanin is pharmacologically active component of beetroot which is responsible for its antioxidant activity. Spectrometry refers to the quantitative estimation of the reflection or transmission properties of a chemical substance. In this research work, the device is used to quantify the optical density of the solution. The amount of pigment present in the solution influences the reading of optical density of the solution so that a solution with more pigments will be associated with higher reading of optical density and lower reading of transmittance. In spectrophotometric techniques, the atom or molecule absorbs a convinced quantum of energy which causes to achieve higher energy level in the atom or molecule. Present Paper is the study of effect of concentration of betanin on their pharmacological application via calculating transmittance and absorbance analysis of pure Beetroot, rose water and in combination helps to the researchers to explore more about clinical significance of betanin pigment of beet root mixed with rosewater.

Keywords— Betanin, Pharmacological application, Beetroot, Rose water, Spectrometry, Electromagnetic Radiation.

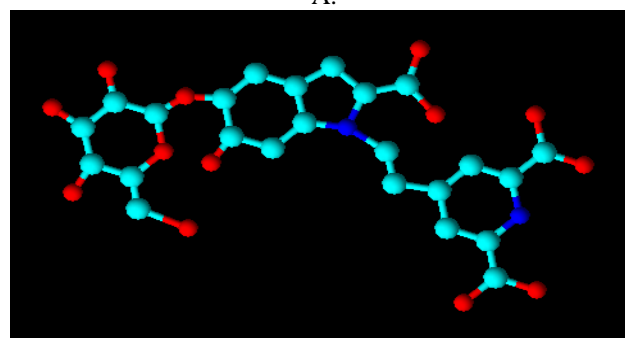
1. Introduction

The beetroot is associated with dietary nitrate which have importance for managing cardiovascular health, type-2 diabetes, atherosclerosis and dementia [1]. Betanin is directly associated with scavenging DPPH activity, prevention of DNA and reduction of low-density lipoprotein. It is well accepted that their pharmacologically active phytonutrient is betalain which consists betanin and isobetanin. The major component and actively participate in biochemical process in pharmacology of beetroot juice is betanin [2].

Now a days use of herbal cosmetics is in trend. Herbal cosmetics have negligible side effects on human health and well accepted by society. Herbal lipsticks, face packs, serums etc. utilizes the use of combination of beetroot juice and rose water in varied concentrations. Dilution of beetroot juice may alter the efficacy of the extract. Variety of pharmaceutical experimental methods are available to justify the quantization of beetroot juice like HPLC, GC-MS, Flash chromatography etc. which includes high cost of analysis [3]. The chemical structure of betanin is mentioned in fig.1 A, their 3-D structure mentioned in fig. 1 B indicates the binding sites for their pharmacological activity.S



A.



B.

Figure 1 A. Molecular chemical structure of betanin.

Figure 1 B. 3-D Structure of betanin

Spectroscopy techniques can explore us, the type of atom or molecule present in solution as well as How much quantity of an atom or molecule is present, the structure and bonding of a molecule. Spectroscopy techniques utilise the fact that atoms/molecules absorb and emit electromagnetic radiation of certain energies. when atoms or molecules absorb electromagnetic radiation, they undergo in some changes [4]. Different parts of the electromagnetic spectrum affect different parts of the atom/molecule. Atoms- movement of electrons to higher level. Molecules- movement of electrons to higher level, movement of molecules to a higher vibrational, rotational and nuclear spin energy levels. It is to be noted that all forms of spectroscopy use part of the electromagnetic spectrum. Ultraviolet light is very short-wave radiation with high energy, while radio waves have long wavelengths and low energy. With the help of this technique present studies of beetroot and rosewater concentration variation absorbance have been determined and result so obtained has been shown in the form of optical density in the table given.

There are two main techniques for taking observation of Absorbance are mainly-

1.1 Colorimetry

The concentration of coloured substances in solution is measured using colorimetry. To calculate the concentration of the solution, the absorption is compared to a calibration graph, which is a graph of the various absorptions of standard solutions with known concentrations [5].

1.2 UV-Visible spectroscopy

This approach is comparable to colorimetric in that it evaluates light absorption employing a light beam. Because it employs a monochromatic to choose particular wavelengths to better detect the existence of particular ions, UV-Visible is more productive.

The sample's wavelength of absorption is determined by a light detector, and this value is contrasted with the wavelength absorption of reference samples. Metal ions in clear fluids can be found using UV-Visible technology. Detecting chemicals in blood and urine and calculating the amount of coloured plastic dye are a few applications. Although the primary application of UV-Spec is to determine the concentration of specific elements in a solution, it can also be useful for qualitative analysis [6]. A material appears coloured when it absorbs light. The apparent colour is the complement of the srivetted colour because this is what remains to reach our eyes.

The scheme diagram for spectrophotometer is shown in fig. 2, which elaborates distinguish and similarities between colorimeter and UV spectrophotometer.

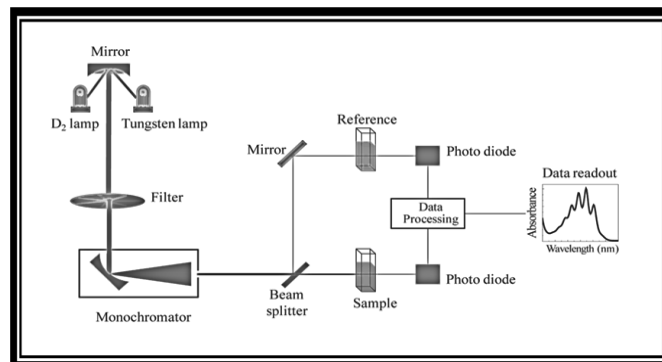


Figure 2. Scheme diagram for spectrophotometer

2. Related Work

From last few spans of time beetroot juice gained its natural medicinal value in the field of their application in food and pharmaceutical industries [1,2]. This extract has variety of pharmacologically active ingredients which gives clinical consequences such as Cardiovascular disorder, atherosclerosis, dementia and type-2 diabetes [7,8]. It has been well accepted by researchers that betalain family components i.e. betanin and iso-betanin are having antioxidant character of beetroot extract [9]. It has been studied that betanin abundance in beetroot juice is approximate $300\text{--}600\text{ mg.kg}^{-1}$ which exhibits the inhibition of lipid peroxidation. Betanin also suppresses the effect of cyclo-oxygenase-II remarkably which supports the fact that it is also having anti-inflammatory effect via selective COX-II inhibition mechanism [10]. Beetroot in combination with rose water used in series of cosmetics likely lipstick, face pack and face serum etc. their chemical constituent and activity can be identified by standard procedures mentioned in pharmacopoeia such as HPLC, GC-MS FRAP assay, DPPH assay etc. these methods utilize high cost [8].

The aim of study is to develop a simple, reliable and cheap method to find active chemical bulk present in beetroot juice or their combinations with rose water for their safer use in pharmaceutical industries.

3. Experimental Method

Stock solution of the purified pigment obtained from Beetroot juice is prepared by transferring 10 mg. of the finely powdered dry material, accurately but rapidly weighed, to a small pestle mortar in which it is meticulously ground with approx. 10 drops of water; 10 ml. of. The solution is then made-up to 100 ml. with water; it is stable for at least 1 hour. The sets of Rose water and Beetroot solutions were made in increasing proportions i.e. 9:1 to 1:9 [5].

This represents a concentration of rose water with beetroot solution in increasing and decreasing order. An approximate light transmission curve was obtained by observation of the standard solution with the colour filters of a Colorimeter. The maximum absorption was obtained at the effective wavelength $570\text{--}700\text{ cm}^{-1}$ and the filter 7 was accordingly chosen for subsequent measurements. It is not ideal and can be

employed with ease only with fairly dilute solutions. Satisfactory reproducibility is, however, obtained with concentrations between 0.001 and 0.01 mg. per ml. of each of the coloured pigment (Rose water and Beetroot) [6].

5. Results and Discussion

Table 1 showing the optical density (Absorbance) and transmittance of different aliquots prepared from beetroot extract and rose water. Results showing the direct effect of concentration of components of beetroot on absorbance value i.e. found in increasing order with increased concentration of beetroot juice in aliquot.

It has been found that on increasing dilution optical density found in decreased manner due to decrease in active chemical constituent of beetroot juice.

Table 1. Optical density and transmittance of different beetroot vs rose water aliquots

OD of Beetroot <i>l</i>) λ_{max} -7 570-700cm ⁻¹	Beetroot: Rose Water OD	Transmittance %
0.27	1:9	94
0.37	2:8	69
0.38	3:7	44
0.60	4:6	28
0.59	5:5	26
0.71	6:4	20
0.75	7:3	18
0.78	8:2	17
0.79	9:1	17

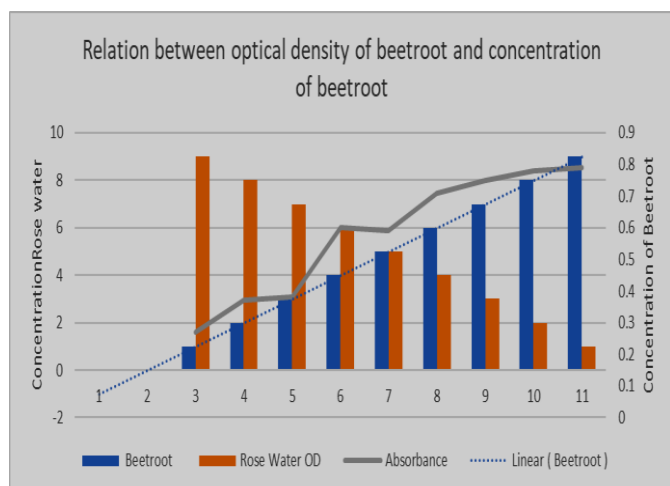


Figure 3. Relation between concentration of betanin in different aliquots with quantity of beetroot and rose water

Fig. 3 indicating the trend obtained in absorbance on increasing concentration of beetroot. It has been observed that after the aliquot ratio 6:4 (beetroot: rose-water) linear trend was found, signifying that further dilution can lose the efficacy of betanin present in beetroot juice. To achieve the maximum pharmacological benefit as antioxidant agent dilution up to 3:2 (beetroot: rose-water) is acceptable. This

fact was supported by fig.4 which is indicating the decreasing trend in transmittance upon increasing beetroot concentration in extract. It has been observed that on increasing the rose water concentration in combination with beetroot juice in pharmaceutical formulations decrease the active chemical bulk which is directly associated with the biological functioning of beetroot juice.

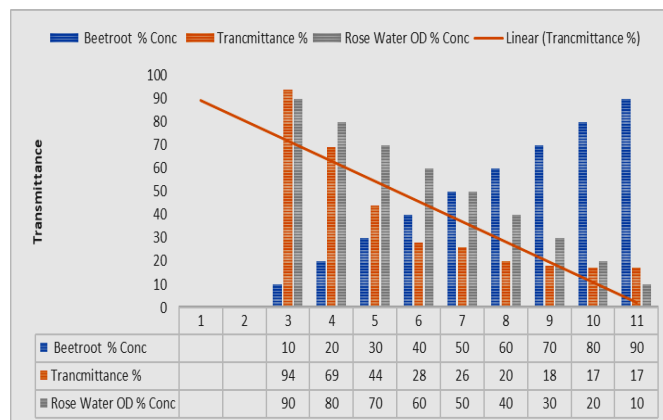


Figure 4. Relation between concentration of betanin in different aliquots with quantity of beetroot and rose water via transmittance.

The stability of the standard solution with respect to reaction and time was studied by means of observations on solutions obtained by dilution of 1 mL of the stock solution to 20 mL with appropriate buffer solutions, the observations in Table are expressed as percentages of this value.

6. Conclusion and Future Scope

It has been concluded that at highest concentration of beetroot, transmittance found at its least value which proves the minimum quantity of Pigment viz. Betanin. It has been indistinct that concentration of active chemical constituent decreases on increasing dilution of beetroot juice or extracts. This extract will have minimum clinical significance. The maximum benefit was found with formulations having greater active chemical bulk. In order to enhance the quality and quantity of extract, beetroot and rose water in the ratio of 3:2 is essential to produce appropriate pharmacological benefits. Beetroot juice can be used as antioxidant agent in combination with rose water in food and pharmaceutical industries by taking the prescribed limit i.e. 3:2. On the other hand, the spectrophotometric method is proved to pay attention towards it, due to its high preciseness, accuracy, feasibility and low cost. This method can be utilized in the field of pharma industry to obtained active chemical constituents present in pharmaceutical compounds.

Data Availability

Not Applicable

Conflict of Interest

The authors declare that they have no competing interests.

Funding Source

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Authors' Contributions

Neha Joshi wrote the primary manuscript. Vinay Marmat crosschecked the results. Both were associated with the planning phase of the study and verified the results and cross-checked the references. Both the authors approved the final version of the manuscript.

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Neha Joshi earned her B.Sc., M.Sc., in pharmaceutical chemistry and submitted Ph.D. in chemistry from DAVV indore in 2008, 2015, and 2023, respectively. she is currently working as Asst. Professor in Department of Chemical Science from Christian Eminent College, Indore since 2019. She has got Researcher of the year award in 2023 in Shikshak samman samaroh organized by Aarav wale-fare Society, Indore. She has published more than 10 research papers in reputed international journals including Thomson Reuters (SCI, Springer nature & Web of Science) and conferences and it's also available online. Her main research work focuses on Medicinal and drug chemistry. She has 6+ years of teaching experience and 6 years of research experience.



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