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Extraction of colours from plants as natural food colorants

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Abstract

Natural color additives are applied to many food, drug, and cosmetic products. with up to 85% of customer shopping for selections doubtlessly influenced through color, are compounds that import color to a variety of products in the food industry. Firstly, the aim of this study was the extraction of natural color from food such as Hibiscus, Red cabbage, Beet root and curcumin. Secondly, this work is focused to study different colors in basic and acidic conditions. The results showed that beetroot and curcumin are stable acidic media but unstable in basic media but anthocyanins are stable in acidic and basic media. The effect of colors on food was carried out.

Key Words: Natural food colors, Hibiscus, Red cabbage, Beetroot, Curcumin, anthocyanin, betanin.

1. Introduction:

Over the past century, the coloring agent that is used in food products which were divided into synthetic and natural coloring agent. Synthetic colorants are already being substituted for natural colorants in many foods. Synthetic colorants are more affordable, brighter, long-lasting, and improve the appearance of food. hundreds of synthetic food colorants have been created from naphthalene, a petroleum-derived chemical. (Dafallah *et al.,* 2015, 21).

Not only do artificial colors cause your Kraft Macaroni and Cheese to shine in the dark and your Light Red Raspberry yogurt to blush. An increasing number of scientific research indicate that they are disturbing children's attention and generating behavioural issues. On the other hand, natural dyes have been used as colorants in food since prehistoric times. These colorants are extracted from plants, fruit and floral with very little chemical processing. Natural colorants are good for human health as they are non-toxic, anti-inflammatory, antiarthritic, antimutagenic , antimicrobial, antioxidants, and anticancer and have medical and pharmaceutical applications (Carocho *et al.*, 2015, 284) & (Rodriguez-Amaya, 2016, 20). In addition to, they are friendly, sustainably reuse, recycle, and repurpose agricultural byproducts and biodegradable for environment.

So, natural colorants have long been thought to be a safer option than potentially dangerous synthetic dyes, particularly in the food industry (Ghosh *et al.,* 2022, 1).The natural coloring compounds that extracted from plants demand in present industry of food. Depending on the food elements used, such as fruits, vegetables, and herbs, the concept of "colored foodstuffs" is also expanding. Despite the various stability issues and limited use, natural colors have a lot of promise to replace a lot of synthetic colorants because of their practical and dietary advantages (Ghosh *et al.*, 2022, 1). (Miranda *et al.*, 2021, 335).

Due to the importance of natural colorants, the present article focused on extraction of natural colors from plants that have less side effects on human and its applications figure (1).

2. The Theoretical Framework

Natural food coloring can be categorized according to their chemical structure as flavonoid derivatives (anthocyanins), nitrogen–heterocyclic derivatives (betalains), polyphenol derivatives (curcumin), or based on their origin (vegetal, animal, bacterial, fungal, etc.) and color (red, yellow, purple, blue, green, etc.) (Viera *et al.*, 2019, 1).From the previous paragraph, introduce theoretical background for natural color agents according to their chemical structures.

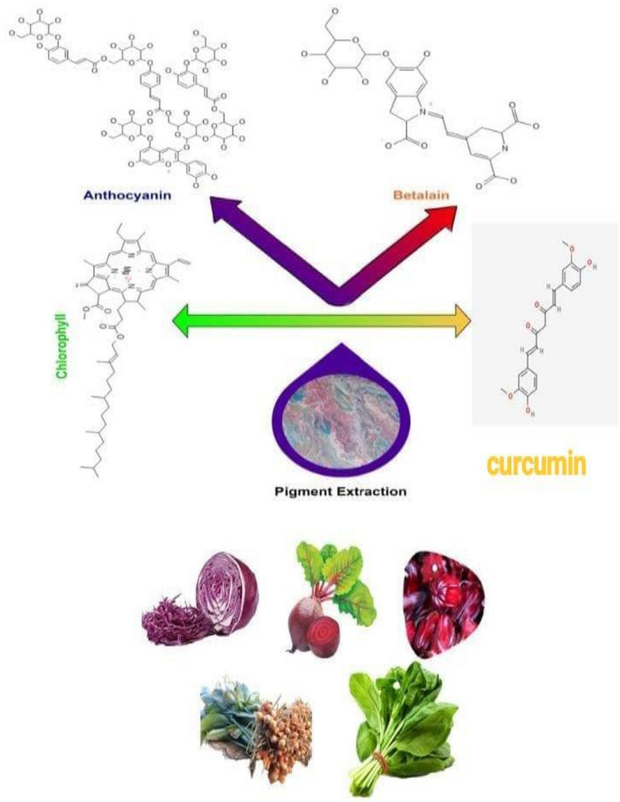

Figure (1): color extraction from food

Anthocyanin

Anthocyanin is a red pigment in acidic environments and a blue pigment in alkaline environments. Despite having a positive charge at the oxygen atom of the C-ring of the fundamental flavonoid structure, anthocyanin is regarded as one of the flavonoids. The flavylium (2-phenylchromenylium) ion is another name for it. (Laleh *et al.*, 2006, 90). anthocyanins' conjugated bonds are responsible for blue, red, or purple pigments in plants, particularly their fruits, flowers, and tubers figure (2).

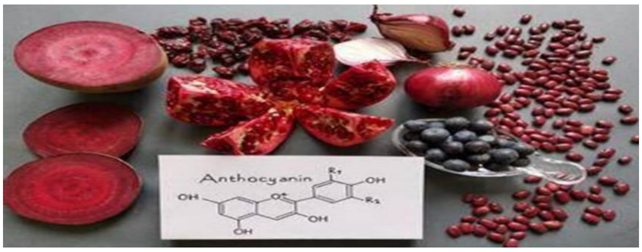

Figure (2): sources of anthocyanin and its structure.

The primary drawback of anthocyanins is their incredibly low stability, which is readily affected by a wide range of factors including co-pigments, enzymes, temperature, light, pH, relative humidity, sugars, vitamin C, oxygen levels, and sulfur dioxide or sulfites.

Anthocyanins are Present in various food such as flowers of Hibiscus rosa-sinensis L., purple cabbage, and Strawberry were chosen as sources of color due to their Richness in anthocyanin content. (Amogne *et al.*, 2020, 9).

Hibiscus is a genus of blooming plants in the Malvaceae family of mallows. Many kinds of secondary metabolites, such as flavonoids, anthocyanins, terpenoids, steroids, polysaccharides, alkaloids, amino acids, lipids, sesquiterpene, quinones, and naphthalene groups, have been shown to be present in numerous Hibiscus species that have been studied, according to the literature. Antibacterial, anti-inflammatory, antihypertensive, antifertility, hypoglycaemic, antifungal, and antioxidative properties have been demonstrated for a few of these substances.
(Kholkute *et al.*, 1977, 127) (Parmar & Ghosh, 1978, 277) (Gangrade *et al.,* 1979, 147) (Jain *et al.*, 1997, 91) (Faraji & Tarkhani, 1999, 231) (Lin *et al.*, 2003,42) (Sachdewa & Khemani, 2003, 61).

Hibiscus species (Malvaceae) have been used as a folk cure for skin diseases, antifertility, antiseptic, and carminative since ancient times. Certain chemicals that have been identified from the species, including polysaccharides, phenolic acids, and flavonoids, are thought to be in charge of these processes (Chopra *et al.*, 1950, 133), (Anonymous, 1959, 55). The first anthocyanin from the calyx of Hs to be extracted was "hiviscin", sometimes known as "hibiscin", later termed delphinidin-3-sambubioside and assigned the structure of cyanidin-3-glucoside (Yamamoto & Osima, 1932.) It was then called as delphinidin-pentoside-glucoside (Yamamoto & Osima, 1936).
Three distinct anthocyanins were found as cHs pigments: delphinidin-3-sambubioside (hibiscin), delphinidin-3-glucoside, and cyanidin-3-glucoside (chrysanthenin). The last study also identified cyanidin-3-sambubioside (gossypi-cyanin) Figure (3) (Du and Francis, 1973, 810) (Shibata *et al.*, 1969, 341).

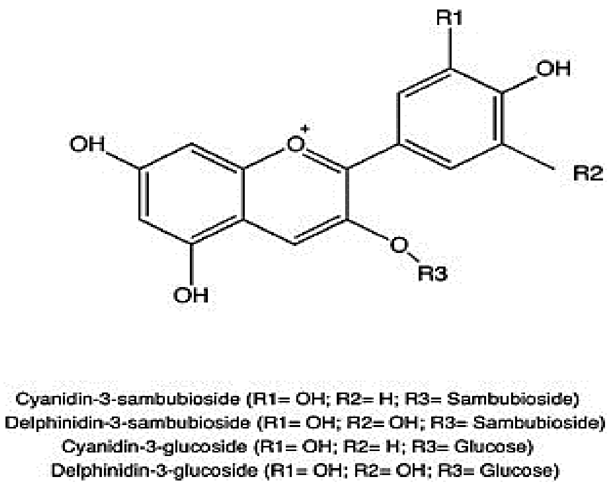

Figure (3):Anthocyanin derivatives in hibiscus.

Hibiscus sabdariffa (cHs) calyces, either fresh or dried, are used to make wine, jam, jellied confecti ons, ice cream, chocolates, puddings, cakes, and h erbal drinks. They can also be used to prepare ferm ented drinks and hot and cold beverages.(Bako *et* *al.*, 2009, 39) (Bolade *et al.*, 2009, 126) (Ismail *et al.*, 2008, 1) (Okoro, 2007, 158) also, it used to treat nerve and heart conditions, lower body temperature (Leung, 1996), Febrifugal , and hypotensive properties (Morton, 1987), diabetes disease (Mayasari *et al.*, 2018, 373), weight reduction (Chang *et al.*, 2014, 734), antianemic (Peter *et al.*, 2017, 288), kidney protection (Da-Costa *et al.*, 2014, 424).

In respect to, red cabbage also includes a number of bioactive substances, including polyphenols, particularly anthocyanins (Zayed *et al.,* 2022, 1). The primary anthocyanin pigments found in red cabbage are cyanidin 3-sophoroside-5-glucoside and its acylated forms with malonic acid, ferulic acid, sinapic acid, and p-coumaric acid12 (Anahi & Caciano, 2015, 944). Red cabbage contains thiamin, riboflavin, folate, and vitamins A, C, K, and B.Iron, nickel, manganese, magnesium, and calcium (Draghici G.A.,*et al.*, 2013, 52).

Betalains

Betalains (betalamic acid) are a kind of natural water-soluble dyes and possesses at least one heterocyclic nitrogen atom and do no longer belong to alkaloids in a strict sense, due to the truth they are acidic in nature due to the presence of several carboxyl groups. (Phytochemicals in Plant Cell Cultures, 1988, 449). it placed in sources such as beetroot, cactus, and cocklebur.

Betalains are hydrophilic and are collected in the vacuoles of the cells, more often than not in the epidermal and subepidermal tissues of plant lifestyles which synthesize these pigments. (Wink M., 1997, 141).Betalain pigments predominately contain betaxanthins (yellow-orange pigments) and betacyanins (red-violet pigments) which existing in flora consist of betanin, isobetanin, probetanin, and neobetanin as shown in figure (4) (Herrero F. *et al.,* 2016, 937) (Wootton-Beard PC. *et al.*, 2011, 329).

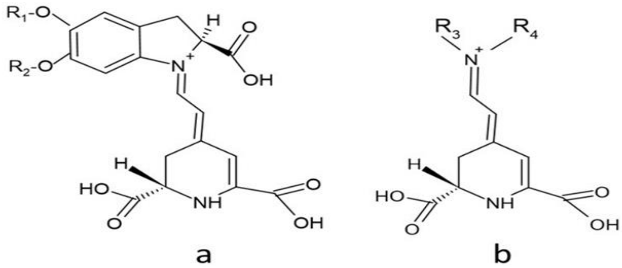

Figure (4): Structure of and betacyanins (a) and betaxanthins (S form) (b). R1 and R2: hydrogen or sugar moieties; R3: amine or amino acid group; R4: usually hydrogen.

There are many elements impact on betalanin balance such as OH, temperature, presence of oxygen. betalains (also betanin) such as antioxidative, anti-inflammation, anticancer, blood pressure and lipid lowering, moreover antidiabetic and anti-obesity consequences (Hadipour E. *et al.*, 2020, 1847) (Strack D. *et al.*, 2003, 247) (Pedreno M. *et al.*, 2000, 49).

Betalains and anthocyanins are high-quality in phrases of chemical structure, then once more they have comparable coloration and herbal performance. The most integral distinction in the chemical structure of these two instructions is that betalains comprise nitrogen, then once more anthocyanins do not (Gürses *et al.*, 2024, 49).

Betalains demonstrate broad range temperature and pH stability which is usually not possible with anthocyanins and hence makes betalain as a better choice for cooking (Stintzing FC. *et al.*, 2004, 19) (Chauhan SP. *et al.*, 2013, 1).

Beetroot (B. vulgaris) is one of the richest sources of betanin and used for acceptable red color. Unlike the synthetic dyes, these beetroot based natural dyes are ecofriendly and pose no environmental problems. The initial interest in the red beetroot was due to the presence of nitrate, which controls high blood pressure (Adv Nutr, 2017, 830).

In addition to nitrate, red beetroot has many other compounds such as ascorbic acid, carotenoids, phenolic acids and flavonoids (Chhikara N. et al., 2019, 192), also high levels of betalain pigments.

Betalain, natural food colorant is associated with the antioxidant, antiviral and antimicrobial activities. Therefore, beetroot dye is associated with nutrient value along with non-toxic nature suitable for coloring application where health aspect is a prime criterion. (Nayak *et al.*, 2006, 173). Red beetroot is used in foods industry as coloring agent in fruit yogurt, ice cream, jams, sauce, soup and cosmetic care products (Esatbeyoglu, et al., 2015, 36) Betanin is used in soups as properly as tomato and bacon products. (Dean D. Metcalfe *et al.*, 416).

Polyphenol derivatives (curcumin)

Curcuminoids are natural polyphenol compounds derived from turmeric, which is a member of the ginger family (Zingiberaceae). Among them, curcumin, with bright yellow color, is the principal composition. Curcumin is the principal curcuminoid produced from the popular Indian spice turmeric (Curcuma longa L.), a member of the ginger family (Akaberi M. *et al.*, 2021,15) (Sharifi-Rad J *et al.*, 2020, 11). These days, curcumin is derived nearly solely from turmeric plants via traditional farming, which is followed by the compound's extraction and purification from the powdered turmeric rhizome. Curcumin levels in fresh rhizome range from 2 to 5%, depending on the growth season (Himesh S *et al.*, 2011, 180).

Chemically, the molecule contains two phenolic rings joined by a seven-carbon linker, which is the reason for the antioxidant properties of curcumin. Curcumin is known by its IUPAC designation, 1,7-bis(4-hydroxy-3-methoxy phenyl)-1,6-heptadiene-3,5-dione (1E-6E), in which a β-diketone molecule is symmetrically connected to two aryl rings that contain ortho-methoxy phenolic OH– groups. Curcumin's β-diketone chain experiences intramolecular hydrogen atom transfer, which results in the equilibrium synthesis of enol and keto tautomeric conformations. Furthermore, these keto-enol tautomers occur in a variety of cis and trans forms, and the temperature, solvent polarity, pH, and aromatic ring substitution all affect the relative concentrations of these forms as shown in figure (5) (Cornago P. *et al.*, 2008, 8089) (Bertolasi V.*et al.*, 2008, 694). Also rings contain methoxy groups are connected to them, which are responsible for the compound's solubility and influence its overall reactivity(El-Saadony MT.*et al.*, 2023, 1040259).

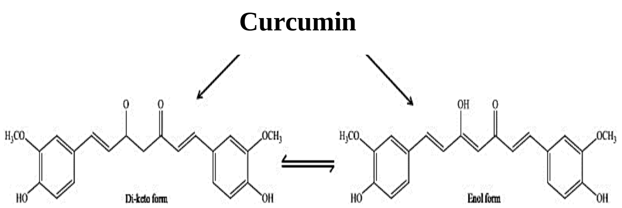

Figure (5): structure of curcumin.

Curcumin inhibits the growth of pathogens (Zare M. *et al.*, 2019, 309) enables people to track food deterioration through color changes (Roy S. *et al.*, 2022, 375) and gives a variety of meals with bright yellow color (Chen *et al.*, 2020, 100) (Cvek M. *et al.*, 2022, 12654). Furthermore, a numerous of research investigations have documented the antioxidant (Jakubczyk K. *et al.*, 2020, 1092), anti-inflammatory (Peng Y. *et al.*, 2021, 4503), anticancer (Tomeh MA. *et al.*, 2019, 1033), (Allegra *et al.*, 2017) neuroprotective (Huang L. *et al.*, 2018, 129), and antimicrobial (Adamczak A. *et al.*, 2020, 153) properties of curcumin.

Curcumin is a common yellow dye used in baked goods, dairy products, mustard, drinks, ice cream, and salad dressings (Novais C. *et al.*, 2022, 2789) (Serpa Guerra AM. *et al.*, 2020, 1842). As a result, curcumin provides a safer and more useful alternative to synthetic yellow colors. Since curcumin changes color according to pH (Etxabide A. *et al.*, 2021, 107), it has been used as a pH biosensor.

3.Methods of Research and the tools used

Materials:

Hibiscus flower, red cabbage, beetroots, and curcumin as color extract.

Sodium hydroxide, sodium carbonate, ammonium hydroxide, hydrochloric acid and acetic acid solutions.

Yougurt, cheese, flour, starch, whipping cream, macaroni as food.

Tools:

Burettes, beakers, balance, hot plates with stirring, test tubes, conical and filter papers.

Method:

Color extraction from Hibiscus.

Hibiscus rosa sinensis flowers were collected, grained and extracted by using distilled water at room temperature and heating. Prepared 5 test tubes and put extract color to examine different solutions as acetic acid, HCl, NaOH, Na_2CO_3, and NH_4OH.

Extract the colour of red cabbage.

Red cabbage was cut into small pieces and placed them into the pot. Water was added until the cabbage was covered with it. The cabbage was boil for 20 to 30 minutes until the liquid has a dark purplish colour. Decant the fluid into a beaker through a strainer to remove the cabbage. Some test solutions was made which are either acidic or basic.

Extract colour from beets.

Beetroots was collected, peeled, then cut into small piece and spread out the beets in a tray and put it inside the oven until it gets dry totally. The final step is to grinding beets using the blender. Prepared 5 test tubes and put extract color to examine different solutions as acetic acid, HCl, NaOH, Na_2CO_3, and NH_4OH.

Extract colour from curcumin.

Curcumin was extracted by using distilled water at room temperature and heating. Prepared 5 test tubes and put extract color to examine different solutions as acetic acid, HCl, NaOH, Na_2CO_3, and NH_4OH.

4. Results of Research

Table (1): Studying the effect of basic and acidic media on extracted color on room temperature

Source of Extracted color	Neutral (water)	Acids (Strong) (HCl)	Acids (Weak) (CH3COOH)	Bases (Strong) (NaOH)	Base (Weak) (NH4OH)	Bases (Weak) (Na2CO3)
Beetroot	red	red	red	brown	Blood red	violet
Hibiscus	red	red	red	Dark green	Red	green
Turmeric	yellow	yellow	yellow	Orange red	Orange red	Orange red turn to orange
Red cabbage	No color	red	pink	Green turn to yellow	Blue	Pale green

Figure (6): Beetroot colour in acidic and basic media at room temperature

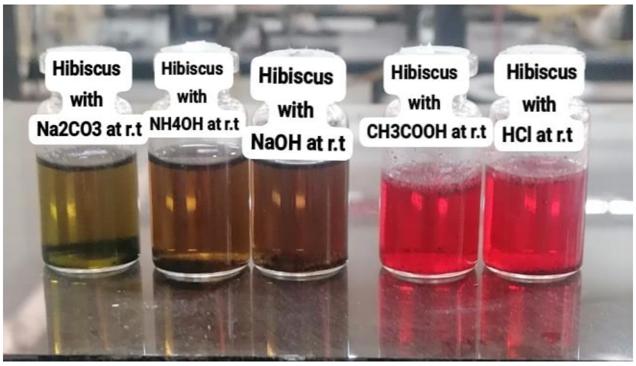

Figure (7): Hibiscus color in acidic and basic media at room temperature

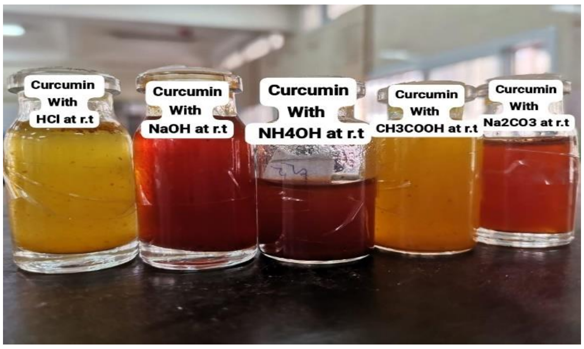

Figure (8): curcumin color in acidic and basic media at room temperature

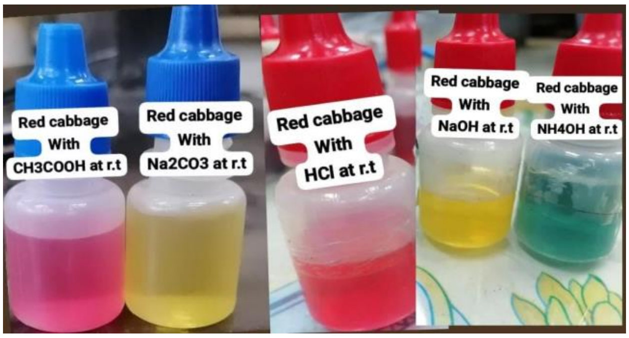

Figure (9): Red cabbage color in acidic and basic media at room temperature

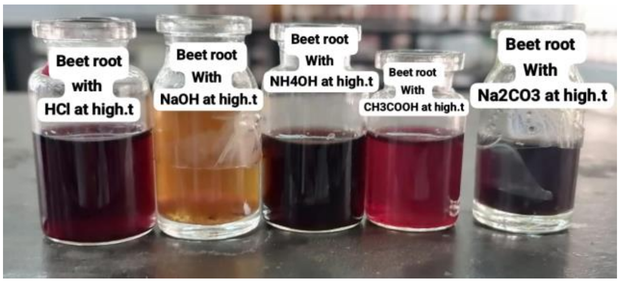

Figure (10): Beetroot color in acidic and basic media at high temperature

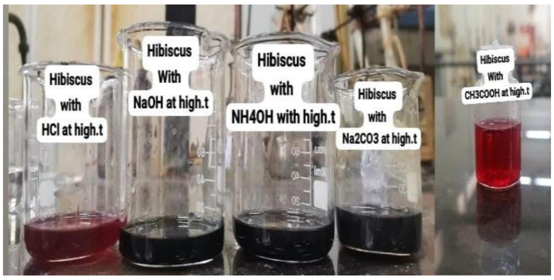

Figure (11): Hibiscus color in acidic and basic media at high temperature

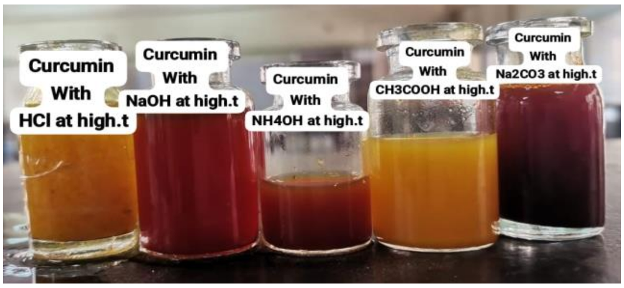

Figure (12): curcumin color in acidic and basic media at high temperature

Figure (13): Red cabbage color in acidic and basic media at high temperature

Table (2) Studying of the effect of basic and acidic media on extracted color on heating.

Source of Extracted color	(H_2O)	(HCl)	(CH_3COOH)	(NaOH)	(NH4OH)	(Na_2CO_3)
Beetroot	red	Dark red	red	brown	Violet	Brownish red
Hibiscus	red	red	Pale red	Dark green	Violet	Dark green
Turmeric	yellow	yellow	yellow	Orange red	Orange Red	Reddish brown
Red cabbage	purple	red	Pink	Pale green turn to yellow	Green	Dark green

Table (3)

	macaroni	**starch**	**flour**	**Whipping cream**
Turmeric				
Hibiscus				
Beetroot				
Red cabbage				

5. Interpretation of Results

In our method of color extraction, the solvent of extraction is water due to it is safe in food and friendly to environment in green chemistry despite its low yield. Anthocyanins color depends on type of medium (Turturică *et al.*, 2015, 9). In neutral medium, stabilized quinonoid anions, which are created by subsequent deprotonation of the quinonoidal species is responsible for purple color of anthocyanins (Khoo *et al.*, 2012, 133). While the medium become acidic, anthocyanins look red (Bąkowska-Barczak, 2005, 107) due to the formation of the flavylium cation (Coutinho *et al.* , 2015, 467). Acidic conditions enhance the flavylium cation stability and intensity of the anthocyanin pigment's red color (Coutinho *et al.*, 2015, 467). In respect to basic medium, deprotonation of the flavylium ion takes place where the quinoidal bases is formed which is responsible for blue colour (Coutinho *et al.*, 2015, 467) as shown in figure (14). So, this explain our results in hibiscus and red cabbage in acetic acid (weak acid) and hydrochloric acid (strong acid) gave pink and red colors, respectivtly as shown in table(1) and figure (7,9) but in basic medium such as ammonium hydroxide (weak base) and sodium hydroxide (strong base), gave green and blue colors as shown in table(1) and figure (7,9).

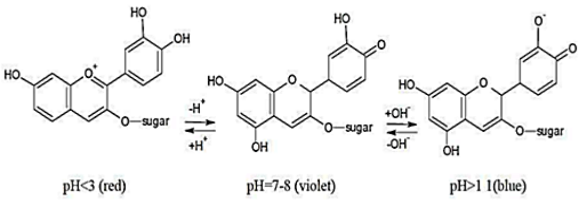

Figure (14): Anthocyanin in acidic and basic media.

(Brouillard *et al.*, 1994, 365) (Jackman *et al.*, 1992, 182)

Betanin is steady in mildly acidic options whereas the shade of the solution,(Von Elbe *et al.*, 1974, 334) along with betalanin, turns from pink to red and the depth of absorption barely will increase due to presence of carboxylic groups. When the solution reaches alkaline levels, betanin degrades via hydrolysis, the coloration of answer turns to yellow-brown, and the depth of absorption is sharply reduced,as shown in figure (15) (Hendry G. *et al.*, 1992),(Saguy I. *et al.*, 1978, 43).

The chemical degradation of colorant is affected by the duration and temperature of the heat treatment as well as different media and water activity of the product (Nemzer BZ. *et al.*, 2011, 42)

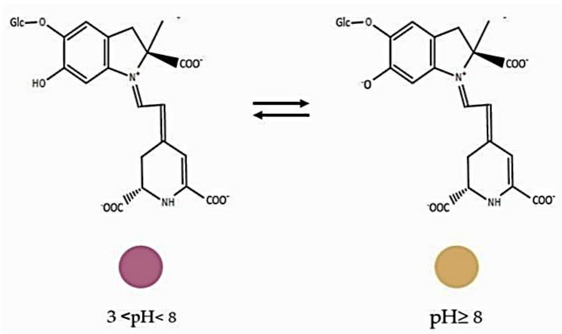

Figure (15): Betanin in acidic and basic media.

The last paragraph explained our data on beetroot as source of betanin which isn't affected by acidic medium such as acetic acid and hydrochloric acid, the color of solution still red as shown in table (1) and figure (6) but in basic medium (ammonium hydroxide, sodium carbonate and sodium hydroxide) the color of solution became between violet and brown due to proton replacement of carboxylic acid was occurred as shown in table (1) and figure (6).

Also, the effect of increasing tempearture on betanin in an acidic solution wasn't observed as shown in table (2) and figure (10) but in basic medium, the intensity of color increased due to degradation of red pigments.

In respect to Curcumin is a polyphenol extracted from the roots of turmeric and affords a sturdy yellow–orange colour (Balbinot-Alfaro *et al.*, 2019, 235) (Bhargava *et al.*, 2020, 385). In acidic medium, it offers yellow coloration due to have a tendency to crystallize out of aqueous acidic options and in basic medium, curcumin shade adjustments to red–orange due to the fact of instability and chemical degradation as proven as shown in the following figure (16).

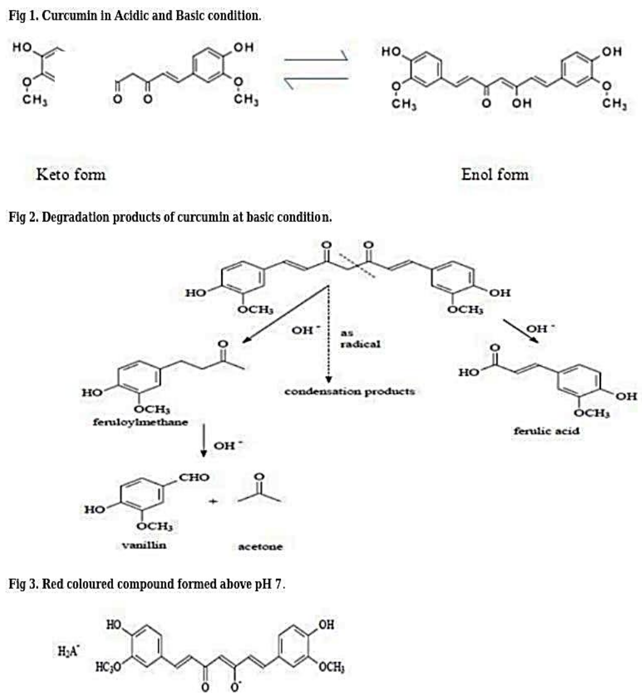

Figure (16): degradation of curcumin in basic medium

This previous information explains our data in curcumin which was without effects on color (yellow color) in acetic acid and hydrochloric acid as acidic media but in basic media (ammonium hydroxide, sodium carbonate and sodium hydroxide), the color of curcumin became orange-red, as shown in table (1) and figure (8).

As results from these data, hibiscus, red cabbage, beetroots and curcumin used as indicators in analytical analysis. Also,the health benefits of on human that takes our main attention to apply these natural colors on food such as yogurt, cheese, rice, starch and flour as shown in table (3)

6. Conclusion

From our experiments, we concluded that hibiscus and red cabbage colors due to anthocyanin in acidic media become red but in basic media turned into blue color. On the other hand, curcumin and beetroots are stable in acidic media while in basic media, chemical degradation was occurred. Also, it has been found that these natural colorants affect food as yogurt, cheese, rice, starch and flour.

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