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# An overview of microbial pigment: A reliable source of biocolor

Dhiraj Patil[1], Murtaza Hajoori[2]
*[1PG]Student, Bhagwan Mahavir College of Basic and Applied Sciences, Bhagwan Mahavir university, Surat.*
*[2]Assistant professor, Bhagwan mahavir college of basic and applied sciences, Bhagwan mahavir university, Surat.*

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**ABSTRACT**
Biological pigments have a significant influence on the growing market for natural substances due to their favorable impacts on human health and the environment. Furthermore, because of their unique propertiesantimicrobial, antioxidant, and anti-cancerthey are valuable in a variety of applied fields, including the food sector, cosmetics, and pharmaceuticals. Microbes may be suitable candidates to achieve the stated goal. Microorganisms are the source of several colors, including prodigiosins, violacein, carotenoids, melanins, flavones, quinones, and, more precisely, monascins and violacein of red wine. The function of microorganisms as a possible source of natural colors and their uses are highlighted in this review study.

**Keywords:** Bacterial pigments, Microorganisms, Application, Bicolour, Food

## I. INTRODUCTION

Numerous artificial synthetic colorants, which are extensively utilized in the production of food, dyes, cosmetics, and pharmaceuticals, have a variety of harmful consequences. Plants and microbes are the two main sources of natural pigments [1]. Given that microbial pigments are frequently more stable and soluble than those derived from plant or animal sources, they are of industrial relevance [2]. Vegetable impacts on pH and temperature were used to isolate and identify several pigment-producing bacteria. Utilizing several biochemical tests, the specific strain was determined. Therefore, research on microbial pigment generation is one of the newest areas showing promise for a range of commercial applications.

The Food and Drug Administration (FDA) regulates pigments strictly because it views them as additives [3]. Food coloring lobbyal market demand was 2400 MT in 2000, rising to 3000 MT by 2005, 8000 MT by 2010, and is predicted to reach 15,000 MT by 2015 [4]. Due to consumer demand for natural food colors over synthetic ones, investment in the natural food color market has reached a billion dollars worldwide and is still rising [5]. Large-scale usage of artificial food coloring is made possible by its affordability and simplicity of mass manufacture. On the other hand, people's awareness of the harmful effects of these artificial food coloring is growing. There has been a noticeable movement in the market towards the creation and manufacturing of Biocolours. The emergence of nutraceutical food products in recent years has further accelerated this trend because natural colors are environmentally friendly and biocolors are recognized to have several positive qualities.

Pigments produced by bacteria, fungi, algae, etc. can be utilized to create biocolors. Microorganisms generate several compounds, including carotenoids, melanins, flavons, phenazines, and quinines [6]. The ability to produce pigment from bacteria has several benefits, such as quick and simple growth in inexpensive culture media, weather independence, and a wide range of color colors. Therefore, one of the newest areas of study to show great promise for a range of commercial applications is microbial pigment synthesis [7].

## II. HISTORY OF BIOCOLOUR

Colour plays a vital role within the nutrition. People have been incorporating colour into their food for millennia, regardless of the type. Chinese candies and wine were dyed as early as 400 BC by the Egyptians. Traditionally, food colouring ingredients like saffron, turmeric, and paprika were used. Around 1300 BC, butter was first made with yellow colour. To provide a bright golden hue to their food, the Romans employed saffron and other spices. In addition, vegetables such as parsley, carrots, pomegranates, grapes, mulberries, spinach, beets, and flowers were used as natural food colouring additives. The Bible mentions saffron, but henna was used well before

2500 BC. According to the article, natural biocolorants are used in food and are recognized to originate from Japan [7]**.**

The history of bio-colors, also known as natural dyes or pigments, dates back thousands of years to ancient civilizations where humans utilized various plant, animal, and mineral sources to color fabrics, artworks, and even their bodies. The use of natural dyes can be traced to civilizations such as ancient Egypt, Mesopotamia, China, India, and Mesoamerica, where artisans developed sophisticated techniques for extracting and applying colorants derived from natural sources[8].

1. **Ancient Civilizations**: Ancient Egyptians used natural dyes obtained from plants, minerals, and insects to color textiles, pottery, and even mummification materials. Indigo, extracted from the leaves of the Indigofera plant, was one of the most prized dyes in ancient Egypt and was used to dye textiles such as linen. In ancient Mesopotamia, Sumerians and Babylonians also used natural dyes derived from plants and minerals to color textiles and ceramics [7].
2. **Classical Antiquity**: In ancient Greece and Rome, natural dyes played a significant role in textile production and were used to create colorful garments worn by the elite. Tyrian purple, obtained from the glands of certain species of sea snails, was one of the most prestigious natural dyes in classical antiquity and was reserved for royalty and high-ranking officials due to its rarity and expense [7].
3. **Medieval and Renaissance Europe**: During the Middle Ages and the Renaissance, Europe saw the widespread use of natural dyes extracted from plants such as woad, weld, and madder. These dyes were used to color textiles, tapestries, and manuscripts, and their production became an important industry in regions such as France, England, and the Low Countries [7].
4. **Colonial Expansion and the Spice Trade**: The Age of Exploration in the 15th and 16th centuries led to the discovery of new sources of natural dyes in regions such as the Americas, Asia, and Africa. European colonial powers established trade routes to procure valuable dye materials such as cochineal (from insects), brazilwood (from trees), and annatto (from seeds) from the New World and other distant lands[7].
5. **Industrial Revolution and Synthetic Dyes**: The 19th century saw the development of synthetic dyes, beginning with the discovery of mauveine, the first synthetic dye, by English chemist William Henry Perkin in 1856. The invention of synthetic dyes revolutionized the textile industry, leading to the rapid decline of natural dyes due to their lower cost, greater color range, and improved colorfastness [7].
6. **Revival of Natural Dyes**: In the late 20th and early 21st centuries, there has been a resurgence of interest in natural dyes due to growing concerns about the environmental and health impacts of synthetic dyes. Artists, artisans, and designers have rediscovered traditional dyeing techniques and have begun to explore sustainable alternatives to synthetic dyes, leading to a revival of interest in natural dyeing processes [7].
7. **Contemporary Applications**: Today, natural dyes are used in various industries, including textiles, cosmetics, pharmaceuticals, and food. Consumers are increasingly seeking eco-friendly and sustainable products, driving demand for natural dyes derived from renewable biological sources. Bio-colors, derived from plants, fruits, vegetables, and microorganisms, are gaining popularity as safe, non-toxic alternatives to synthetic dyes in a wide range of applications [7].

### 2.1 Sources of Bio-Colors

Bio-colors can be sourced from a wide range of biological materials, including plants, fruits, vegetables, flowers, insects, and microorganisms. Each source offers unique color compounds that can be extracted and used as natural dyes. Common examples of bio-color sources include [18].

- Plant-based dyes: Plants such as indigo, turmeric, madder, and henna contain natural pigments that can be extracted and used for dyeing textiles and other materials.
- Fruit and vegetable dyes: Fruits and vegetables like berries, beets, onions, and spinach contain vibrant pigments that can be extracted and utilized as natural colorants in food, cosmetics, and textiles.
- Microbial dyes: Certain bacteria, fungi, and algae produce pigments with various colors, such as red, yellow, green, and blue. These microbial pigments can be cultivated and harvested for use in dyeing applications**.**

## III. MICROBIAL PIGMENTS AS FOOD COLOURS

There are countless varieties of microorganisms on this planet. Numerous microorganisms, including bacteria, fungus, and micro algae, are known to create a wide range of colours. Microbial pigment has grown to be a significant source of biocolors as color extraction from microbial sources has several advantages, including less expensive manufacturing, simpler extraction, greater yields through strain enhancement, reduced shortage of raw materials, and no seasonal changes Currently, canthaxanthin, astaxanthin, zeaxanthin, and other carotenoids are important microbial pigments. Conversely, additional pigments consist of Prodigiosin, Indigoidine, Violacein, and others. Therefore, if properly extracted, these pigments may be used to color a variety of culinary products, including cakes, pastries, and milk[9]

## IV. CLASSIFICATION OF MICROBIAL PIGMENTS

Microbial pigments can be categorized based on their source, color, and solubility. Depending on the source (fungi, biological sources, and algae),Bacteria are classified according to their solubility (fat-soluble, water-soluble), with yellow pigments (riboflavin, carotenoids) and red pigments (prodigiosin, carotenoids, and porphyrins)[7].

**4.1: Carotenoids**: A class of pigments known as carotenoids is found in many different types of bacteria, algae, fungus, and plants. They range greatly in color, from red to yellow [11].Carotenoids are abundant in the natural world. Carotenoids are mostly lipid-soluble substances that are widely available in foods like carrots, pumpkins, and tomatoes. In the food sector, carotenoids are a crucial type of coloring ingredient. Zeaxanthin, astaxanthin, canthaxanthin, and beta-carotene are examples of the carotenoids that are produced by a vast array of microorganisms. Therefore, if extracted, these chemicals can be employed as a food-grade bio-colorant[10].

**4.2: Zeaxanthin:** In general, zeaxanthin is a pigment found in bacteria. Bacteria including Staphylococcus aureus, Bacillus, Corynebacterium michigannise, Flavobacterium sp., and others contain it. Therefore, it is simple to utilize as a biocolorant after extraction. Zeaxanthin is sold as a food ingredient with the number E161h on the market [11]. This pigment imparts a yellow-orange color to wolfberries, maize, and paprika. When zeaxanthin degrades, picrocrocin and safranal are produced [12].

**4.3: Prodigiosin:** Prodigiosin is an alkaloid secondary metabolite that is mostly generated by bacteria and has a distinct tripyrrole chemical structure [13]. Other Gram-negative, gamma proteo bacteria like Vibrio psychroerythrus and Hahella chejuensis are also responsible for producing this pigment, but Serratia marcescens is the primary bacterial strain that makes it. Prodigiosin-containing spray-dried microcapsules were created despite the pigment's drawbacks, which include sensitivity, solubility, short stability to pH, and high temperature. Milk, yogurt, and carbonated beverages were successfully made with the particles [14].

**4.4: Indigoidine:** a member of the Azaquinones group of chemical compounds, is indoidine. Some bacteria-induced stains have created a blue tint. Artificial food coloring like FD&C Blue No. 1 and 2 can be substituted with it. is often used in cereals, yogurt, ice cream, baked products, snack foods, and confections.

**4.5: Astaxanthin:**As a keto carotenoid, astaxanthin is included under xanthophylls. This pigment dissolves in lipids. A reddish-orange pigment is called astaxanthin. Farm-grown salmon and chicken eggs have long been colored pink with the use of artificially produced astaxanthin. However, astaxanthin generated by microbes is currently sold and used to color meats, dairy goods, and drinks [15]. The primary carotenoid pigment of red yeast, Xanthophyllomyces dendrorhous, is astaxanthin, which is utilized as a food colouring [16].

**4.6: Anthocyanins:** Anthocyanins,which belong to the flavonoid group of polyphenols, are significant substances found in plants that serve as antimicrobials, pigments, and antioxidants [17]. Many food plants, including black rice, black soybeans, blueberries, and raspberries, contain anthocyanins. Up until now, the primary method of producing anthocyanins has been extracting and purifying plant tissues such as fruits, flowers, and other tissues.

**4.7: Canthaxanthin**: Canthaxanthin is a carotenoid pigment found in various organisms, including plants, fungi, and certain microorganisms. It is responsible for the pink-orange color observed in foods such as salmon, shrimp, and some fruits and vegetables. Canthaxanthin is often used as a food coloring agent (E161g) to add a yellow-orange hue to products like beverages, dairy products, and processed foods. Additionally, canthaxanthin has antioxidant properties and has been studied for its potential health benefits, such as protecting against UV-induced skin damage and reducing the risk of certain diseases [18].

**4.8: Granadaene**: Granadaene is a red pigment found in the pomegranate (Punica granatum) fruit. It belongs to a class of compounds called ellagitannins, which are polyphenolic compounds known for their antioxidant properties. Granadaene contributes to the vibrant red color of pomegranate arils and juice. Pomegranate consumption has been associated with various health benefits, including anti-inflammatory, anticancer, and cardioprotective effects, attributed in part to the presence of compounds like granadaene[18].

**4.9: Riboflavin**: Riboflavin, also known as vitamin B2, is a water-soluble vitamin involved in various metabolic processes in the body, including energy production and the metabolism of fats, proteins, and carbohydrates. Riboflavin is naturally present in foods such as dairy products, eggs, lean meats, green leafy vegetables, and whole grains. It serves as a precursor for the synthesis of coenzymes involved in redox reactions, making it essential for cellular function and growth. Riboflavin is also used as a food additive (E101) to fortify and colorize a wide range of processed foods and beverages[18].

**4.10: Melanin**: Melanin is a complex polymer pigment produced by melanocytes in the skin, hair, and eyes of animals, including humans. It plays a crucial role in determining skin color and protecting against UV radiation-induced damage. Melanin absorbs and scatters UV light, preventing excessive penetration into the skin and reducing the risk of sunburn and skin cancer. The amount and distribution of melanin in the skin are influenced by genetic factors, sun exposure, and hormonal regulation. Disorders affecting melanin production, such as albinism and vitiligo, can result in abnormal skin pigmentation and increased susceptibility to sun damage[18].

**4.11: Rubropunctatin**: Rubropunctatin is a red pigment produced by certain species of fungi, particularly those belonging to the genus Penicillium. It contributes to the reddish coloration observed in moldy foods and environmental substrates contaminated with these fungi. While rubropunctatin itself does not have significant commercial or nutritional importance, it serves as a marker for fungal contamination and can indicate potential spoilage or toxin production in food products. In addition, some researchers have explored the potential pharmacological properties of rubropunctatin and related fungal metabolites for their antimicrobial and anticancer activities [18].

## V. HEALTH BENEFITS OF USING BIO COLOURS EXTRACTED FROM MICRO-ORGANISMS

Their function is to shield against deadly photooxidation. Mutagenesis is inhibited. Using biocolor can strengthen defenses against infections. Additionally, they could prevent the growth of tumors. Biocolours are known for their numerous biological qualities that are beneficial to the body's health. Antioxidant polyphenols are found in most pigments. It has been demonstrated that microbial pigments such as naphthoquinone, carotenoids, anthocyanins, and violacein are strong antioxidants Numerous research have reported on the anti-cancer benefits of biocolors derived from microorganisms. These pigments have anti-cancer properties.

The aquatic cyanobacterium Scytonemin produces a green-yellow pigment that has an anti-proliferative impact by blocking the activity of the cell cycle regulating protein kinase [19].

| **Microorganisms** | **Molecules /Pigments** | **Colour/Appearance** |
|---|---|---|
| Agrobacterium aurantiacum | Astaxanthin | Pink-red |
| Cornybacterium insidiosum | Indigoidine | Blue |
| Bradyrhizobium species | Canthaxanthin | Dark-red |
| Penicillium oxalicum | Anthraquinone | Red |
| Serratia marcescens, Serratiarubidaea | Prodiogiosin | Red |

| | | |
|---|---|---|
| Xanthomonas oryzae | Xanthomonadin | Yellow |
| Chromobacterium violaceum | Violacein | Purple |
| Haloferax alexandrinus | Canthaxanthin | Dark-red |
| Janthinobacterium lividum | Violacein | Purple |
| Staphylococcus aureus | Staphyloxanythin, Zeaxanthin | Golden Yellow |
| Pseudomonas aeruginosa | Pyocyanin | Blue-Green |

**Table 1: Highlights of some pigment producing Microorganisms**[20].

## VI. ROLE IN THE FOOD INDUSTRY

**6.1. Research and Development:**Research to find new sources of bio-colours, improve extraction techniques, and evaluate the stability and safety of these colours in various food compositions might include [21].

**6.2. Quality Control:**Ensuring the quality and consistency of bio-colours is crucial. It's possible that it will work on creating quality control procedures to make sure bio-colours satisfy both client and regulatory requirements[21].

**6.3. Regulatory Compliance:**Staying abreast of regulations concerning food colorants is essential. Making sure bio-colours used in food items adhere to the regulations set by organizations like the European Food Safety Authority (EFSA) and the Food and Drug Administration (FDA) might be part of the job [21].

**6.4. Technical Support:**One aspect of the job description would be offering food producers technical assistance on how to successfully and economically incorporate bio-colours into their goods. This might involve offering guidance on formulation, stability, and application techniques[21].

**6.5. Marketing and Consumer Education:** Communicating the benefits of using bio-colors to food manufacturers and consumers is essential. Creating instructional materials and marketing campaigns to emphasize the organic and nutritious qualities of bio-colours might be part of the job description [21].

**6.6. Sustainability Initiatives:** Many consumers are increasingly concerned about the to ensure sustainable sourcing practices for bio-color ingredients[21].

## VII. ROLE'S BIO-COLORS PLAY IN PHARMACEUTICALS:

**7.1 : Identification and Branding:** Bio-colors are often used in pharmaceuticals to distinguish different medications, dosage forms (such as tablets, capsules, or liquids), or strengths. This helps healthcare professionals and patients easily identify and differentiate between various drugs [22].

**7.2: Patient Compliance:** By adding color to medications, it can make them more appealing and easier for patients to recognize and remember to take their medications as prescribed. This can be particularly important for patients who are taking multiple medications or who have cognitive impairments [22].

**7.3: Safety and Quality Assurance:** Bio-colors can be incorporated into pharmaceutical formulations to help detect any potential issues such as contamination or counterfeiting. For example, color-coding can help identify if a tablet has been tampered with or if there are any defects in the manufacturing process [23].

**7.4: Masking Unpleasant Taste or Odor:** Some medications have a bitter taste or unpleasant odor, which can be masked or improved by adding bio-colors. This can enhance patient acceptability and adherence to treatment regimens [24].

**7.5: Dosage Formulation:** Bio-colors can also be used to indicate different dosages within the same product [24].

## VIII. APPLICATION OF PIGMENTS IN TEXTILES

Bacterial pigments in methanol were used as the stock solution for yellow, green and red pigment respectively. Attempts were made to dye cotton, viscose and polyester fibers with the extracted microbial biocolourants. The dyeing of cotton/polyester and viscosafibers were carried out at 100℃ for 60 min. One set of experiment may be done with the addition of thiourea, alum, copper sulphate, ferrous sulphate and lime as a mordant for increasing binding capacity of pigments to fibers[25].

### 8.1 Washing fastness

The preheated soap solution (Tide, at 60 ºC) and water was taken in the ratio of 1: 50 (0.5 g/25 mL) in beaker. Dyed fibres were added in the solution for 30 minutes. Then the specimen was removed and rinsed in cold water[26].

### 8.2Rubbing fastness

The rub fastness of the dyed fibres was carried out by rubbing the fibres manually and checking for fading of colour.

### 8.3 Light fastness

The dyed fibers were exposed to sunlight for 24 h. The color fastness to light was evaluated by comparison of color change of the exposed portion to the unexposed original material. The same procedure can be repeated for the dyed textile material treated with any mordant. The rating for rubbing, light and washing fastness was determined to respect to staining on cotton, polyester showing rating between 1 to 5[26].

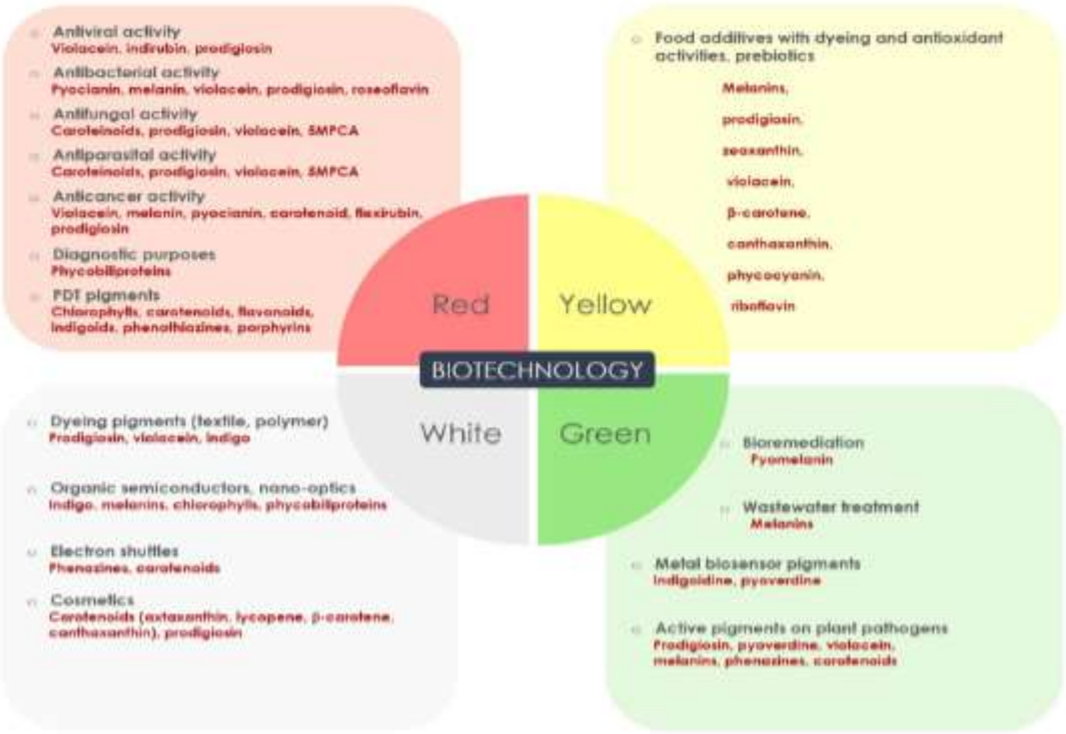

**Figure 3: Applications of bacterial pigments in biotechnology for medicinal (red), food (yellow), industrial (white), and environmental/agricultural (green)[28].**

### ➢ Methods of Pigment extraction from bacterial cells:

Centrifuging the bacterial cells at 8000×g for 5 minutes and discarding the supernatant is the first step in pigment extraction from the cells. To recover the cells, the pellets of cells are centrifuged at 8000×g for five minutes after being washed with deionized water. through another supernatant discharge. Next, 2 milliliters of 99.7% pure ethanol are thoroughly mixed with the retrieved cells. Once the cells are fully bleached, the combination of the cells and ethanol is subjected to ultrasonication. Centrifugation at 10,000×g for 5 minutes is used to remove the ethanol extract from the cells [27].

### ➢ Advantages Of Bacterial Pigments

Bacteria-produced pigments have been utilized for a long time in eastern nations, and in recent decades, they have been the subject of much investigation because to their prospective uses.

Bacterial pigments offer the following advantages and benefits:

a. Increasingly attractive to sceince because of broad ranging activities…
b. Broad strain selection and simple propagationHigh versatile and productive over other sources
c. Fermentation is inherently faster and more productive production
d. Quick and easy culture methods that enable continuous bioreactor functioning.
e. Low-cost substrate utilized in large quantities.

➢ **Disadvantages Of Bacterial Pigments**

There are numerous elements that affect the quality and efficacy of natural pigment products. Moreover, natural pigments have certain drawbacks despite the push toward environmentally friendly procedures. Removing colour from unprocessed materials requires time. Limited supply exists for natural pigments. Natural pigments give rise to brilliant colours in a range of tones at first, but when exposed to sunshine and domestic laundering, they tend to fade more quickly than synthetically dyed materials. Using natural pigment to dye might be difficult in terms of colour consistency. Unacceptably high levels of toxicity may be present in certain mordants. Certain of the more vivid colours need the use of more hazardous mordants like tin and chromium. The fact that some mordants—such as alum, lime, and turpentine are not harmful should be noted [28].

➢ **Current limitation and future perspective**

One of the interesting areas of study in the biological sciences is bacterial pigments and how they may be used to different businesses. Researchers are working to replace synthetic pigments with bacterial pigments derived from biological sources, but there are a number of difficulties in their way. These include high production costs, low stability, variations in color due to operational parameters, low annual production percentages, specifications regarding the types of bacteria that can produce specific colors, and technological flaws that cause delays in the industrial production of pigment from bacteria and the extraction of pure and concentrated forms. Finding the simplest way to extract bacterial pigments should be the focus of research in order to increase the industrial uses for these pigments. There's also an imperative.

## IX. CONCLUSION AND FUTURE PERSPECTIVE

Nature is rich in colour and also the source of colour. Microorganisms being one of the sources and the innumerable microbes are found. Food grade biocolours extracted from ,in coming years ,has the potential to commercially replace artificial colour with shifting of market. They will have positive effects on the ecosystem in addition to us humans. There will be less chemical contamination. In order to fully harness these bacteria for human benefit, further research and development are being conducted. The spectrum of biocolors available currently is limited, but research is being done to increase it in order to partially replace artificial colorants on the market and take use of the benefits of these pigments.

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