

Abstract

During the Taimurid and Safawid periods the masters introduced a number of dyes for coloring paper for the purpose of calligraphy and paintings. Historical analysis was carried out to identify different dyes, shade of colors, and the application of techniques recommended in the paper-dyeing process based on Persian historical treatises.

Among the many dyes introduced by the masters, henna has been the most recommended, with a ratio of 1:10 henna and water. Scientific analysis was carried out in two stages to investigate the effect of henna on paper in a lower concentration than the advised ratio, followed by laboratory work to investigate the fungicidal properties of henna dye in the advised concentration. Our experiment showed that henna acts as a fungicide on *aspergillus flavus* only when the ratio of henna to water is higher than 1:10. The present study revealed the secret behind the henna concentration that has been stressed in historical recipes.

Résumé

Pendant les périodes safavide et timouride, les maîtres ont introduit un certain nombre de teintures pour colorer le papier destiné à la calligraphie et aux peintures. Des études historiques ont été menées pour identifier les différentes teintures, nuances de couleurs et l'application des techniques recommandées pour le procédé de teinture de papier dans les traités historiques persans.

Parmi les nombreuses teintures introduites par les maîtres, le henné était le plus recommandé, avec un ratio de 1/10 de henné et d'eau. Les analyses scientifiques ont été conduites en deux étapes, d'abord pour étudier l'effet du henné sur le papier, en concentration plus faible que celle recommandée, puis pour étudier, par une recherche en laboratoire, les propriétés fongicides de la teinture de henné dans les volumes recommandés. Nos expériences ont démontré que le henné agit comme un fongicide sur l'*aspergillus flavus* uniquement lorsque le ratio de henné et d'eau est supérieur à 1/10. L'étude a révélé le secret derrière la concentration de henné recommandée dans les recettes historiques.

Historical analysis of materials used in Iranian paper dyeing with special reference to the effect of henna dye on paper based on scientific analysis

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Introduction

The art of calligraphy and illumination which emerged in the great Islamic civilization is undoubtedly one of the most important achievements in the history of human civilization. Within the world of Islam, paper was produced for the first time in the eastern part of the Islamic world – or in Khorāsān – by Chinese captives during the last decades of the first part of the 2nd century AH (751 AD). This spread to other Islamic territories and, soon, paper became a significant item of export by the Islamic world. Persia, too, was considered one of the most important centers of paper-making and it also acted as a bridge in transferring the art of paper-making from the East to the West. This art had become so popular throughout Iran that there were some cities in which the entire population was engaged in paper-making (Māyel Heravi 1993). The increased need for inscribing books on paper prompted the paper makers of the mid-Hejira centuries and thereafter, those of the Timurid and Safavid eras, to pay more attention to the aesthetic aspects of paper-making, the outcome of which was the production of various kinds of artistically colored papers.

Methods of study

There are a number of dyes, revealed by masters in Iranian treatises, which have been overlooked by scientists and conservators. Studying these historical references not only opens a new chapter in the identification of materials used in manuscripts and miniature paintings throughout history, but it also helps to understand their role in preservation due to the science behind their use. Our analytical study on materials used in Iran in the paper dyeing process was broken down into two phases: historical analysis and scientific analysis.

Synopsis

Durante los periodos Timurid y Safavid, los maestros introdujeron un gran número de tintes para colorar papel con el fin de realizar caligrafía y pinturas. Se ha llevado a cabo un análisis histórico para identificar los diferentes tintes, la tonalidad de los colores y la aplicación de las técnicas recomendadas en el proceso de tintura del papel. Este estudio ha usado como fuentes los tratados históricos persas.

Entre los muchos tintes introducidos por los maestros, la henna ha sido el más recomendado en una proporción de 1:10 (henna / agua). El análisis científico se realizó en dos fases para investigar el efecto de la henna sobre el papel en una concentración inferior a la proporción recomendada; siguió un trabajo de laboratorio para investigar las propiedades fungicidas del tinte de henna con la concentración recomendada. Nuestro experimento demostró que la henna actúa como fungicida sobre el *aspergillus flavus* sólo cuando la proporción de henna y agua es mayor de 1:10. El estudio actual ha revelado el secreto que se escondía tras la concentración de henna recomendada en las recetas antiguas.

Historical analysis

A number of treatises relating to methods used in paper dyeing during the periods under study have survived and are accessible to us today. Historical evidence pertaining to the Timurid and Safavid eras and right up to the Qajar period reveals that the papers used for inscribing books during these periods were generally dyed. Experts in this field had recommended the dyeing of paper with two aspects in view: one was the aesthetic aspect and the other was from the viewpoint of the effect that the color of the paper rendered on the eyesight of the reader. According to them, viewing white paper rendered a harmful effect on the eyesight of the reader, while dyed paper exerted lesser effect on the reader's eyes. In his famous treatise entitled "*Golzār-e Safā*", (Seyrafi 1545), a renowned expert in this field during the Safavid period, included the following quatrain that touches upon this subject:

*Paper, once dyed is better,
For white surely harms the eyesight;
It is not favorable to hurt the eyes,
To refrain from penning on uncolored paper is wise.
First, dye the paper to beauty,
So that your hand and eyes remain fine;
Since I expect you to scale great heights in this art,
I have benefited you with this little part.*

In some old treatises, references have also been made to the harmful effects of some kinds of dyed paper on the eyesight. Furthermore, some recommendations can also be found regarding the inks used for calligraphy and the dyes employed for coloring paper. For instance, in "*Favāyid al-Khotuʿ*", (Mohammad Bokhāri 1590) has suggested the following points:

While writing with black ink it is not advisable to use certain colors as background. Papers which are dyed by color shades such as red, green, dark blue and white dazzle the eyes like looking at the sun. For writing purpose papers with temperate colors are good. For writing with colored inks, papers with dark background are advisable.

In another source, "*Rasm al-khat*", (Heravi 1504) advises that:

Eye dazzle to see red, green and white paper as they do while looking at the sun. For calligraphy temperate colors shall be used as they relax the eyes. The colored lines are good on dark papers. Write on red papers with white colors. On blue paper, writing with white is pleasant.

19 treatises of historical importance belonging to the period from the Taimurid, Safavid and Qajar eras have been studied for the purpose of this research work; details are given in Table 1 along with their source number for easy reference.

From among the most important of these treatises, mention can be made of works like the "*Adab al-mashq*", "*Savad al-khat*" and the "*Golzāri Safā*", the latter being written in the form of verse. Among the historical treatises under study, five have classified the colors used for dyeing paper in two categories: primary (*mofradat*) and secondary (*morakabat*). All these treatises have almost unanimously classified colors such as yellow (*zard*), red (*sorkh*), safflower red (*al*), blue (*kabud*), grayish green (*zangari*), natural (*khodrang*), straw (*kabi*) henna (*hanna'i*) and peach (*shafstali*) as primary colors while colors such as black (*udi*), green (*sabz*), pink (*golgun*), kind of green (*farise*), orange (*narenji*), iris (*susani*) and malachite (*marmari*) have been categorized as secondary colors.

As in the case of textiles, the dyeing of paper involves various stages depending upon the conditions and the properties of the dye. Some colors such as saffron and henna are extracted by simple soaking or boiling prior to use, while the extraction of other colors such as indigo involves a more complicated process. On the other hand, some dyes like turmeric and safflower are stabilized with the use of acidic substances. According to

Table 1. The list of the historical treatises on Persian paper dyeing process from 13th to 19th centuries

Source No.	Historical treatises (resalat)	Author	Exact dates
1	Resaleh dar Bayani Kaghaz, Morakab va Hali Alvan	Anonymous	9th AH (15th AD)
2	Seratal Sotour	Soltan Ali Mashhadi	9th and 10th AH (15th and 16th AD)
3	Golzari Safa	Ali Seyrafi	10th AH (16th AD)
4	Khat va Morakab	Hossein Aghili Rostamdari	10th AH (16th AD)
5	Adab al-Mashgh	Ahmad Majnoon Rafigh Heravi	10th AH (16th AD)
6	Savad al-Khat	Ahmad Majnoon Rafigh Heravi	10th AH (16th AD)
7	Rasm al-Khat	Ahmad Majnoon Rafigh Heravi	10th AH (16th AD)
8	Adab al-Mashgh	Baba Shah Isfahani	10th AH (16th AD)
9	Favayed al-Khotout	Mohamad Ibn Doust Mohamad Bokhary	10th AH (16th AD)
10	Resaleh dar Bayani tarigheh Sakhtani Morakab va kaghazi Alvan	Anonymous	10th AH (16th AD)
11	Bayazi Khoshbouie	Anonymous	11th AH (17th AD)
12	Resaleh Sahafi	Syed Yousouf Hosseini	12th AH (18th AD) ?
13	Resaleh Khoshnevisi	Abdullah Seyrafi	12th AH (18th AD)
14	Johari Simi	Simi Neishapuri	12th AH (18th AD)
15	Morakab Sazi va Jeld Sazi	Ali Hosseini	13th AH (19th AD)
16	Majmoue al-Sanayie	Anonymous	13th AH (19th AD)
17	Resaleh dar Marefati Kaghazi Alvan	Anonymous	13th AH (19th AD)
18	Resaleh dar Bayani Rang Kardani Kaghaz	Anonymous	13th AH (19th AD)
19	Resaleh dar Bayani Khat va Morakab va Kaghaz va Sakhtani Rangha	Anonymous	13th AH (19th AD)

historical evidence, acidic substances like pomegranate and lemon extracts, namely *kesbteh*, commonly referred to as stabilizers, were used for the process. Some other dyes extracted from pomegranate peel and sappanwood need mordants like sulphate to help them to permeate the yarn. A classification of the colors used in dyeing paper – on the basis of the historical treatises – is presented in Table 2. Tables 3 and 4 present classifications of the primary and secondary colors along with the respective dyes, mordants and acids used.

Although the basic dyes are limited in number, a large range of color shades is obtained by a series of intricate mixing and dosing. To obtain different shades of colors, simple techniques have been applied, such as soaking in a diluted solution and drying in the sun to obtain a light color, whereas with repeated soaking and drying in the shade, bright or dark colors have been obtained (Porter 1994). Besides primary and secondary colors, many other shades are described in detail in Persian historical treatises for dyeing purposes; bluish to greenish, greenish to purplish and yellowish to reddish shades. In the research that was conducted on the basis of the aforementioned historical treatises, the process for extracting more than 50 different types of dyes was identified, including primary and secondary

Table 2. The category of colors used in dyeing paper process (*gooneh kardan*) based on Persian treatises

Source No.	Primary colors (<i>Mofradat</i>)									Secondary colours (<i>Morakabat</i>)						
	Yellow (<i>zard</i>)	Red (<i>sorkh</i>)	Safflower red (<i>al</i>)	Blue (<i>kabud</i>)	Greyish green (<i>Zangari</i>)	Natural (<i>khodrang</i>)	Straw (<i>kahi</i>)	Henna (<i>hanna'i</i>)	Peach (<i>shaftalu</i>)	Black (<i>udi</i>)	Green (<i>sabz</i>)	Pink (<i>golgun</i>)	Kind of green (<i>farise</i>)	Orange (<i>narenji</i>)	Iris (<i>susani</i>)	Malachite (<i>marmari</i>)
1	x	x	x	x	x	x	x			x	x	x	x	x		
14	x	x	x	x	x	x	x			x	x	x	x	x		
17	x	x	x	x	x	x	x			x	x	x	x	x		
10	x	x	x	x	x	x		x			x	x		x		
18	x	x	x	x	x	x	x	x	x	x				x	x	x

Table 3. The list of dyes used as primary colours during Safawid to Qajar period based on Persian historical treatises

Primary colours	Ingredients	Mordants and acids
Yellow (<i>zard</i>)	<ul style="list-style-type: none"> • Saffron (<i>zafaran</i>) • Turmeric (<i>zardchoobeh</i>) 	- Alkaline ash (<i>ashkhar</i>), Lemon juice
Red (<i>sorkh</i>)	<ul style="list-style-type: none"> • Sapanwood (<i>baqqam</i>) • Purple amaranth (<i>bostan afrooz</i>) • Mulberry (<i>shahtoot</i>) • Mixture of <i>baqqam</i>, <i>bostan afrooz</i> and <i>shahtoot</i> • Lac (<i>laak</i>) • Safflower (<i>moasfar</i>) 	- Alum (<i>zaji turki</i>) - Alkaline ash (<i>ashkhar</i>) - Salt - Extract of lemon, orange or sour pomegranate (<i>keshteh</i>), old vinegar, wine,
Safflower red (<i>al</i>)	<ul style="list-style-type: none"> • Safflower (<i>moasfar</i>) 	- Alkaline ash (<i>ashkhar</i>) - Extract of lemon, orange or sour pomegranate or any sour fruit (<i>keshteh</i> or <i>qalya</i>), extract of unripened grape (<i>ghura</i>), sour grape or old vinegar
Blue (<i>kabud</i>)	<ul style="list-style-type: none"> • Seed of sunflower (<i>tokhmi alafi aftar gardesh</i>) • Indigo (<i>nil</i>) 	- Sal-ammoniac (<i>naushadour</i>)
Greyish green (<i>zangari</i>)	<ul style="list-style-type: none"> • Verdigris (copper + vinegar) • Verdigris (Sal-ammoniac + copper + grape vinegar) 	- Vinegar - Vinegar
Straw (<i>kahi</i>)	<ul style="list-style-type: none"> • Safflower (<i>moasfar</i>) 	
Natural (<i>khodrang</i>)	<ul style="list-style-type: none"> • Henna • Henna + Black ink (<i>medad</i>) and/or Saffron (<i>zafaran</i>) 	

Table 4. The list of dyes used as Primary colors during Safawid to Qajar period based on Persian historical treatises

Secondary colors	Ingredients	Mordants and Acids
Black (<i>udi</i>)	Red + Blue: <ul style="list-style-type: none"> • Red Lac (<i>lak</i>) + Blue (<i>Kabud</i>): Indigo (<i>nil</i>) + Seed of sunflower (<i>tokhmi alafi aftar gardesh</i>) 	- Sal-ammoniac (<i>naushadour</i>)
Green (<i>sabz</i>)	Blue + Yellow: <ul style="list-style-type: none"> • Blue (<i>kabud</i>): Indigo (<i>nil</i>) + Seed of sunflower (<i>tokhmi alafi aftar gardesh</i>) + Yellow: Safflower (<i>moasfar</i>) + Saffron (<i>zafaran</i>) 	- Sal-ammoniac (<i>naushadour</i>)
Kind of green (<i>farise</i>)	Gall + Blue: <ul style="list-style-type: none"> • Extract of Gall (<i>Mazu</i>) + Blue (<i>kabud</i>): Seed of sunflower (<i>tokhmi alafi aftar gardesh</i>) + Indigo (<i>nil</i>) 	- Sal-ammoniac (<i>naushadour</i>)
Orange (<i>narejni</i>)	Reddish Yellow + Yellow: <ul style="list-style-type: none"> • Safflower (<i>moasfar</i>) + Saffron (<i>zafaran</i>) • Red <i>al</i>: Safflower + Saffron (<i>zafaran</i>) • Turmeric (<i>zardchoobeh</i>) or Rind of Pomegranate (<i>naspal</i>) + Safflower (<i>moasfar</i>) 	- Alkaline ash (<i>ashkhar</i>) + Sour Fruit Extracts (<i>keshteh</i>) - Extract of Lemon
Pink (<i>golgun</i>)	Yellow + Red: <ul style="list-style-type: none"> • Saffron (<i>zafaran</i>) + Lac (<i>lak</i>) • Safflower (<i>moasfar</i>) 	- Extract of Lemon
Iris (<i>susani</i>)	Blue + yellow Indigo (<i>nil</i>) + Safflower (<i>moasfar</i>)	- Extract of Lemon
Malachite (<i>marmari</i>)	Blue + Yellow: <ul style="list-style-type: none"> • Indigo (<i>nil</i>) + (<i>guli balas</i>) • <i>guli zaban dar ghafa</i> 	- Alkaline Ash (<i>ashkhar</i>) + Extract of Lemon - Potash (<i>ghalyab</i>) + Alum (<i>zaj</i>)

colors and their various shades; this work deserves to be described in a separate article. From among these 52 shades of colors and on the basis of historical evidence, the researcher has been able to identify 16 organic dyes, 5 minerals and 10 mordants and acids that were used for coloring paper. Table 5 presents a list of the substances that were used for the purpose of dyeing paper.

Table 5. The list of the identified colors, dyes, pigments and additives used in Persian paper process during Safavid to Qajar period

Nos.	Dyes	Minerals	Mordants and acids
1	Henna (<i>hanna</i>)	Ceruse, lead or tin carbonate (<i>sefeedab</i>)	Vitriol (<i>zaj</i>)
2	Saffron (<i>zafaran</i>)	Gold (<i>tala</i>)	Sal-ammoniac (<i>naushadour</i>)
3	Turmeric (<i>zardchoobe</i>)	Blue vitriol, copper sulphate (<i>zaji kabud</i>)	Alkaline ash (<i>ashkhar</i>)
4	Sapanwood (<i>baqqam</i>)	Verdigris (<i>zangar</i>)	Extract of Lemon
5	Safflower (<i>moasfar, kajira</i>)	Orpiment (<i>zarnikh</i>)	Extract of orange
6	Rind of Pomegranate (<i>naspal</i>)		Extract of pomegranate
7	Indigo (<i>nil</i>)		Unripe grape (<i>ghura</i>)
8	Purple amaranth (<i>bostan afrooz</i>)		old vinegar
9	Mulberry (<i>shahtut</i>)		Potash (<i>ghalyab</i>)
10	Lac (<i>laak</i>)		Alum (<i>zaj</i>)
11	Seed of sunflower (<i>tokhmi alafi aftab gardesh</i>)		
12	Bhutea (<i>guli balas</i>)		
13	Delphinium orientia, larkspur, (<i>guli zaban dar ghafa</i>)		
14	Lily flower (<i>goli susan</i>)		
15	Black ink (<i>medad</i>)		
16	Gall (<i>mazu</i>)		

Henna the most advised dyes for coloring paper

One of the methods that has always attracted the attention of experts in this field was the dyeing of paper with natural dyes extracted from henna, which was used by artists throughout various periods of history.

Among different colored papers, henna is specifically recommended to achieve a natural color (*kehodrang*) in historical documents, either in pure form or mixed with saffron. For example, Baba Shas Isfahani (16th AD) in *Adab-almashq* mentioned in poetry form that, *There is no color comparing to henna; there is no need for experiment.*

Moreover Majnoon Rafigh Heravi (16th AD) in *Savad al-khat* advises that,

The best color for paper is henna in which calligraphy, gold and decorative lines on it become elegant. It is made by a mixture of henna dye, black ink and saffron.

As mentioned earlier, henna dye is fully recommended by different masters based on their experiences. Perhaps that is why this color is widely used in Persian paper manuscripts. Most of the historical treatises have stressed the point of the ratio of henna to water for obtaining the dye. The technique has been explained in the different historical sources under study. Among them *Resaleh dar bayani kaghaz morakab va hali alvan*, *Golzāri Safā*, *Resaleh dar bayani tariqeh sakhtani morakab va kaghazi alvan*, and *Resaleh dar bayani rang kardani kaghaz*, can be mentioned. In all these sources the methods that have been advised to make henna are mostly as follows:

Heat a few clean free unbeaten henna leaves for a while. Keep it for one full day and then filter it to get the extract of henna leaves. To get the best result, add 10 sers' water and one ser henna. If more water is added, the color becomes dull and dusty (malleh).

It has to be mentioned that in only one treatise, *Resaleh Sabafi*, a lower concentration has been advised which is one ser henna and half man water (1.5 kg). In treatise source no. 18 the ratio is not indicated and it is advised to obtain the desired color by personal experience.

Since all the sources above strongly advise dyeing paper with henna, the author initially studied the anti-fungal properties of henna and then conducted a scientific examination in the second phase of the study.

Scientific analysis

In scientific analysis, the effects of dye extracted from henna are specifically examined to explore the reason behind the use of henna for coloring paper as is repeatedly advised by masters in Persian historical treatises. For this purpose, the investigation was carried out in two stages.

First stage

In the first stage the chemical composition of henna was reviewed. The coloring matter of henna has been investigated by many authors. It has been reported that the leaves of henna contain 7% tannin, 6% fat, 1.2% essences, and 2–3% Lawson (2-hydroxy-1:4-naphthaquinone) responsible for the antimicrobial properties of henna. The antibacterial activity of an aqueous extract of henna leaves has been demonstrated by (Malekzadeh 1968) and (Malekzadeh and Shabestari 1989). It is also reported by (Soker 2000) that henna has fungicidal properties. Since the fungicidal property of henna was already studied and confirmed, at this first stage we were not concerned about the ratio of henna to water.

Materials: Three different types of handmade paper were selected from the conservation laboratory of the congress library in Tehran. Each paper was divided into three groups. In each group, the paper was divided into four pieces of 2.5 cm² in size for the sample experiment. 1, 2 and 3 grams powdered henna leaves were prepared from Yazd, the center part of Iran, for dyeing the paper samples

Methods: 1, 2 and 3 gram powdered henna leaves were soaked in 60 ml of distilled water in three separate containers and kept under artificial light for four hours. Each solution was filtered through a filter paper. The four paper samples collected from each group were soaked in solutions for 5, 15, 30 and 60 minutes. Each paper group was dyed simultaneously in the three different concentrations for the four different lengths of time to observe the effects of time and the concentration of dyes used in the coloring process against the growth of fungus on paper samples. A phosphate buffer (pH = 7.0) was prepared and the process was carried out in the Microbiological Laboratory of Gamma Irradiation Centre, Tehran.

Culturing: In all experiments, 10 ml of *aspergillus flavus* was prepared of approximately 1×10^{-7} spores per ml. The concentration of the suspension was estimated by the pour plate method. Potato dextrose agar was applied to each sample.

Fungus growth: The samples were incubated at 25 °C for four weeks and the diameter of the zone of inhibition was measured to the nearest mm by means of a celluloid millimeter ruler. A magnifying glass was used when needed (Figure 1).

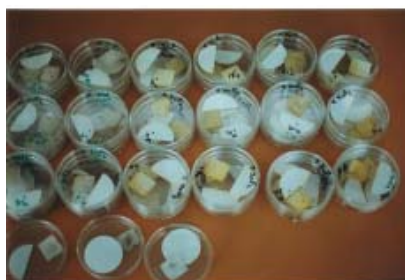


Figure 1. Fungus growth of paper group samples dyed with henna extract solutions

Second stage

In this stage we decided to carry out further laboratory work to investigate the effect of henna dye on the *aspergillus flavus* fungus in a different ratio and to study the relationship of the henna concentration to the growth of fungus.

Material: Pure henna leaf samples were prepared from Yazd provenance. Sabouraud dextrose agar was prepared from Merck. All samples were diluted in double distilled water and then sterilized in an autoclave at 121 °C for 15 min.

Methods: The laboratory work was conducted directly on the henna solution in concentrations of 2.5%, 5%, 7.5%, 10%, 12.5%, 15%, and 17.5%. These solutions were incubated for 2 hours at 75 °C. To get the complete extract, the solutions were kept at room temperature for 24 hours and then filtered. The extracted solution was used to culture the fungus (Figures 2 and 3).



Figure 2. Preparation of henna in different concentrations



Figure 3. Filtering henna extracts for preparing solutions

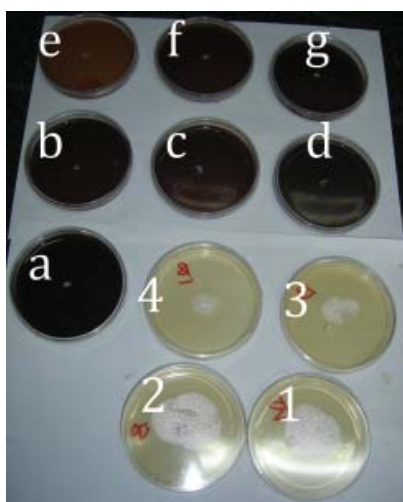


Figure 4. Culture of *aspergillus flavus* on untreated samples (plate 1 to plate 4) and treated samples 2.5% to 17.5% (plate a to plate g) in 5 days

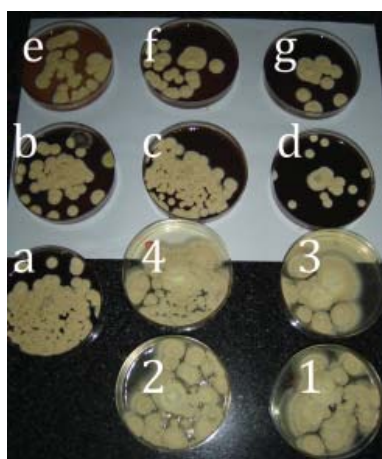


Figure 5. Culture of *aspergillus flavus* on untreated samples (plate 1 to plate 4) and treated samples 2.5% to 17.5% (plate a to plate g) in 8 days

Culturing: 1 gram Sabouraud dextrose agar brass was mixed in 15 mL (ml?) of henna solution extracts where the samples were all sterilized.

Fungus growth: To all henna samples 2.5% (plate a), 5% (plate b), 7.5% (plate c), 10% (plate d), 12.5% (plate e), 15% (plate f), and 17.5% (plate g) including untreated samples (plate 1 to plate 4) *aspergillus flavus* fungi were inoculated and the samples were studied every 12 hours.

Results of first stage

As a result of our experiment we were surprised to find that the samples which were dyed with henna showed that the growth of *aspergillus flavus* fungus depended on the concentration of henna. In a higher concentration of henna the growth of *aspergillus flavus* fungus decreases. This result led us to study further the chemistry of henna dye and carry out further laboratory work, described in the second stage.

Results of second stage

Our experiment on henna solutions in different concentrations (2.5–17.5%) showed that henna can minimize fungus growth and can also delay the process. Comparing the samples under study, the growth of fungus in henna solutions of different concentrations from 2.5% to 17.5% (plate a to plate g) [this is 7, not 8?] starts after five days, whereas the untreated samples (plates 1 to 4) show fungus growth after two days (Figure 4). As shown in Figure 5, by increasing the concentration of henna extract from 2.5% to 17.5%, the growth of *aspergillus flavus* fungus will decrease. As shown in Figure 6, the experiment shows clearly that this decrease starts from 10% concentration; at this point, the growth of fungus reduces to 60% (plate d). The experiment also shows that in the higher concentrations, this depreciation continues to reduce the fungus growth (plates e to g).

Discussion

To analyze these results we did further study on phenol and quinine compounds present in henna leaves. One of the properties in phenols and quinone compounds is their antimicrobial property. As the study has shown, they are also very effective on pathogenic fungi which produce a number of illnesses. Compared to pathogenic fungus, saprophyte fungi such as *aspergillus flavus* are more resistant to phenolic and quinone compounds. The study by (Soudi 1989) has shown that they can easily stand 10 $\mu\text{mol/ml}$ naphthaquinone. In henna dye it is shown that a concentration of 2% is effective on *M.gyoeum* (pathogenic fungus) and 1% is effective on *T. rubrum*, whereas it is only effective at 10% on *aspergillus flavus*. This might be due to the presence of carbon in quinone compounds that can be a source of food for *aspergillus flavus*.

Our experiment in the second stage also showed that the extract of henna dyes in higher than a 10% concentration can act as a fungicide against *aspergillus flavus*. The fungicidal property of henna can be maximized at the 17.5% concentration. At a higher concentration it turns into paste and is no longer suitable for dyeing. According to our investigation henna extract higher than 10% can reduce fungus growth by more than 60%.

Conclusion

Our study based on historical analysis shows that Iranians used to dye paper manuscripts for aesthetic and scientific reasons. In the present research 52 shades of color, 16 main dyes and 5 minerals were identified, based on historical treatises. In the first phase, paper samples were dyed in different ratios at lower concentrations than the ratio that the masters had advised. Surprisingly, we found that the samples which were dyed with henna have a

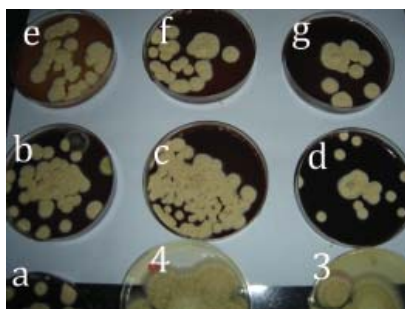


Figure 6. Depreciation of the fungus growth starting from 10% concentration is shown on the plate d

greater tendency to inhibit the growth of *aspergillus flavus* compared with undyed samples. In the second phase, an aqueous solution of henna dye was prepared using traditional techniques in different ratios and the fungicidal property of henna extract in different concentrations was examined. Our experiment showed that only henna dye higher than 10% can act as a fungicide against *aspergillus flavus* fungus. According to our investigation, henna extract in higher than a 10% concentration can reduce the growth by more than 60%.

Our experiment shows that the recipes suggested by the Iranian masters during the 15th and 16th centuries have scientific validity and that henna dye was used historically as a preventive measure for fungus growth on paper.

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Note

1 *Ser* is a traditional Iranian measure equivalent to 75 grams.

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