Influence of dyeing condition on cloth color by yellow dyes from mixed fungi grown in different culturing media

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Abstract. In the present study, cloth was dyed with yellow dyes obtained from a mixture of Aspergillus and Paecilomyces fungi grown in three different media, three mordant techniques, six mordant types and four dyeing repetitions. By matching with the Royal Horticultural Society (RHS) color chart, the range of colors developed on the dyed cloth was measured. The results showed that medium-1 and medium-3 produced stronger dyed cloth colors than medium-2; and different mordant types and techniques each also produced more varied cloth colors from both media than did medium-2. Meta- and pre-mordant using different mordant types applied to the dyes from all of the media each produced more varied cloth colors than did post-mordant; while mordant technique using Cu$^{+2}$ and Fe$^{+2}$ in the same media each produced more varied cloth colors than did other mordant types. The optimum condition for the cloth dyeing by the dyes in medium-1 and medium-3 added with pre-mordant Cu$^{+2}$ was 3 times, while with meta-mordant Cu$^{+2}$ was 3 and 5 times respectively. The optimum condition for the cloth dyeing by the dyes in medium-1 and medium-3 added with pre-mordant Fe$^{+2}$ each was 1 time, while with meta-mordant Fe$^{+2}$ was 1 and 5 times, respectively.

1. Introduction

Throughout the 19th century, natural dyes were the main dyes for textiles [1]. Since the discovery of synthetic dyes, these dyes have almost completely replaced natural dyes. The advantages of synthetic dyes are the variety of colors available, greater reproducibility, improved color quality, and economic benefits. However, it is familiar that some synthetic dyes are toxic for the environment and have a negative impact on the ecosystem. Research conducted by Velmurugan et al [2] reported that 10-35% of synthetic dyes were lost in wastewater during the textile dyeing process. Due to serious environmental pollution problems, textile industry faces extensive problems and some synthetic dyes have been banned. As a result, there is a demand for environmentally friendly and non-toxic dyes for industrial applications.

Some organisms such as algae, animals, bacteria, fungi, and plants are able to synthesize pigments, but the fungi are one of the potential organisms to produce large amounts of pigments very prominently [3]. Most pigment-producing fungi are from species of Aspergillus, Emericella, Fusarium, Monascus, Paecilomyces (Isaria), and Penicillium [4-5]. Most of the pigments produced by fungi are quinone, flavonoids, melanin and azaphilone, which belong to the aromatic polyketide chemical group [6] and have been widely reported for the use of drugs and dyes [7-8].

In the previous study, greyed-purple dyes produced by mixed Aspergillus and Paecilomyces successfully dyed cotton cloth [9-11]. They produced dyes at room temperature in mineral salt glucose
medium of Backer and Tatum [12]. However, the effect of other media on dyes production by the same mixed fungi was unknown.

Natural dyes, however, have a little coloring power within themselves. The problem can be overcome by applying a pretreatment to the textile with mordant type and technique as well as dyeing repetition. Mordant type helps in the binding of dyes to the fiber by forming a chemical bridge between the dyes and fiber [13]. Selection of mordant types and techniques is important in natural dyeing processes. Single dyes source added with different mordant types and techniques produce different colors and tones [10]. Dyeing repetition also intends to get a stronger color than the previous dyeing and a better color resistance to the material so that it is not so easy to fade [14]. Previous study by Suciatmih [11] observed the cloth dyeing by mixed Aspergillus and Paecilomyces fungi dyes repeated 3, 6, and 9 times. The dyeing repetition distance between the treatments was relatively long. In order to find cloths quickly saturate the dyes, it is necessary using dyeing repetition where distance between treatments shorter, such as, 1, 3, 5, and 7 times.

In this study, we aim to examine the dyes production of mixed Aspergillus and Paecilomyces fungi in different media and evaluate the combination effect of different mordant techniques, mordant types, and media; and the combination effect of different media, dyeing repetitions, mordant techniques, and mordant types on cloth color by the fungal dyes. The series of studies conducted to find the best combination of fungal culture medium, mordant type and technique as well as dyeing repetition produced a strong color on cloths.

2. Materials and methods

2.1. Materials

Aspergillus and Paecilomyces used in this study were isolated from soil collected at the Cibinong Science Center (CSC), Indonesia. The cloth used was cotton, purchased from Bogor Market, Bogor, Indonesia. Primisima brand cotton was chosen as the substrate due to its relatively widespread use in consumer products and natural dyes research. The study was conducted at the Environmental Microbiology Laboratory, Research Center for Biology, Indonesian Institute of Sciences (LIPI), Cibinong. The chemicals used were analytical grade from Merck, except potato dextrose broth (PDB) from Pronadisa, and technical grade CaCO$_3$ and KAl(SO$_4$)$_2$.12H$_2$O from local market in Bogor.

2.2. Methods

2.2.1. Inoculation process.

Inoculation of mixed Aspergillus and Paecilomyces fungi was performed as our previous method [9]. Briefly, each five mycelial prints of the mixed fungi were inoculated into three different media separately: 1) Medium-1 contained: 24 g PDB, 2 g yeast extract, 2 g peptone, 0.5 g MgSO$_4$.7H$_2$O, 0.46 g KH$_2$PO$_4$, 1 g K$_2$HPO$_4$; 2) Medium-2 contained: 24 g PDB, 5 g yeast extract, 3 g NaNO$_3$, 0.5 g KCl, 1 g K$_2$HPO$_4$, 1 g MgSO$_4$.7H$_2$O; and 3) Medium-3 contained: 24 g PDB. All of the media were incubated at room temperature in a static way for 4 weeks in the dark. After the incubation periods, the cultures were passed through five layers of cheesecloth, and the filtrate was centrifugated (Kubota 6500, Japan) at 8500 rpm for 20 min [15]. The optical density of the filtrate was determined spectrophotometrically (Shimadzu, Japan) at 530 nm for quantifying the dyes [15]. The dyes' yield was calculated as OD units (UA400).

2.2.2. Dyeing and Mordant Technique.

Different mordant techniques (pre-, meta-, and post-mordant) and types (CaCO$_3$, CuSO$_4$.5H$_2$O, FeSO$_4$.7H$_2$O, KAl(SO$_4$)$_2$.12H$_2$O, K$_2$Cr$_2$O$_7$, and MnSO$_4$.H$_2$O) applied to the dyes obtained from different media for the cloth dyeing was carried out. Mordant concentration used was 1%. Pre-mordant cotton samples (4 cm x 4 cm or 0.24 g) were soaked in a mordant solution first before dyeing with the dyes. Cotton samples introduced into the mordant bath with a ratio of 1: 30 w/v (material: mordant).
After 15 min of boiling at 90°C, the samples were lifted, squeezed and dried at room temperature. Pre-mordant samples were then placed into the dyes bath with a ratio of 1: 30 w/v (material: coloring). After dyeing, the samples were soaked in 1% detergent for 10 minutes with stirring, rinsed and dried at room temperature. Post-mordant samples were dyed in the dyes first before soaking in mordant. After dyeing, the samples were lifted, squeezed and placed into the mordant bath for 15 min at 90°C. The samples were then soaked in 1% detergent for 10 minutes with stirring, rinsed and dried at room temperature. Meta-mordant samples were concurrently soaked by the dyes and mordant in one bath. Non-mordant samples were dyed by the dyes without mordant. Each treatment was conducted in duplicates. Mordant type and technique applied to the dyes obtained from different media yielded more varied color on the cloth was used for the investigation of dyeing repetition.

2.2.3. Dyeing repetition.
Dyeing repetition was carried out with pre- and meta-mordant each using CuSO₄·5H₂O and FeSO₄·7H₂O by the dye obtained from medium-1 and medium-3. Cloth (4 cm x 4 cm or 0.24 g) was dipped in the dyes or the mordant with a ratio of 1: 30 w/v (material: coloring/mordant) at 90°C for 15 minutes, lifted, squeezed, and dried at room temperature. According to the treatment, the colored cloth was dyed again in the dyes at the same temperature and time. Furthermore, the colored cloth was lifted, squeezed, soaked in 1% detergent for 10 minutes with stirring, rinsed with water, and dried at room temperature. There were four treatments for dyeing repetition, namely 1) One time repeated dyeing; 2) Three times repeated dyeing; 3) Five times repeated dyeing; and 4) Seven times repeated dyeing. Each dyeing repetition was conducted in duplicates.

2.2.4. Color determination.
The color of the cloth after dyeing from each treatment was determined by matching the cloth color with the Royal Horticultural Society (RHS) color chart repeatedly [16].

3. Results and discussion

3.1. The effects of fungi culture media on the colors of cloth
A mixture of Aspergillus and Paecilomyces cultures in different media produced the same dyes color (figure 1). The color of the dyes produced in this study (yellow) was different from the color of the dyes (greyed-purple) when the same mixed fungi was cultured in the medium of mineral salt glucose [9-12]. It might be because the changes in some nutrients in the culture media used could influence fungal growth [15,17]. Similar results in previous study [18] reported that the same mixed fungi cultured in the Backer and Tatum medium [12] produced greyed-purple dyes, whereas when it cultured in the Cho et al [19] and Poorniammal et al [20] media each produced red dyes.

The optical density of the dyes in medium-1, medium-2, and medium-3 was 1.8382 UA/L, 1.5626 UA/L, and 1.7894 UA/L, respectively. The study indicated that medium-1 gave higher dyes production than medium-2 and medium-3. It could be because of the components, such as metal ions/or other micronutrients in medium-1, are suitable and facilitate enzymes to work effectively, increasing the growth of metabolites and dyes production [21]. There is a tendency that higher dyes absorbance result in a stronger dyed cloth color. The control’s cloth color dyed with the dyes in medium-1 (1.8382 UA/L) and medium-3 (1.7894 UA/L) was stronger (159A orange-white) than that in medium-2 (1.5626 UA/L) (159D orange-white) (tables 1, 2, and 3).
Dyes produced by the three different media when applied to cloth without mordant (control) each produce colors that are still white in color such as 159A orange-white (medium-1), 159D orange-white (medium-2), and 159A orange-white (medium-3) (tables 1, 2, and 3). All dyes have not yet put a good color (yellow) on the cloth unless they were added with mordant. Different mordant techniques combined with different mordant types applied to the dyes produced some colors that were stronger and more varied colors (grey-brown, greyed-orange, greyed-yellow, and yellow) than the color of the control cloth without mordant (orange-white). These results indicated that those dyes were included in non-substantive dyes or the dyes have a little coloring power within themselves so that they need mordant for fixation of a dye into the fiber [22]. This result is different from the result of Suciatmih [11] on the same mixed fungi cultured in the Backer and Tatum medium [12] that produced substantive dyes.

3.2. The effects of mordant type and technique on the colors of cloth
The dyeing process in this study used the dyes in three media in various treatments of mordant techniques (pre-, post-, and meta-) and mordant types (Al$^{3+}$, Cu$^{2+}$, Cr$^{3+}$, Cu$^{3+}$, Fe$^{3+}$, and Mn$^{2+}$). Based on 54 different treatments (3 media, 3 mordant techniques, and 6 mordant types), 14 different colors on the cloth were obtained (tables 1, 2, and 3). Widihastuti [23] informed that the content of the dyes when it comes to the metal elements contained in the mordant with various mordant techniques would produce certain colors according to the nature and elements of the dyes.

According to Uddin [24], mordant also play very important role in imparting color to the cotton cloth. He reported that depending on the metal ions of mordant, the complex formation not only strengthens dye fixation on the fiber, but also modifies the final cloth color. The control cloth dyed with only the dyes obtained from three different media each produced an orange-white color (tables 1, 2, and 3). The color was visually the same as the color of the cloths dyed with the dyes obtained from medium-1 using post- and meta-mordant Ca$^{2+}$, pre-mordant Cr$^{3+}$ and Mn$^{2+}$, and post-mordant Al$^{3+}$. The dyes from medium-2 with pre-, post-, and meta-mordant Ca$^{2+}$, Cr$^{3+}$, and Mn$^{2+}$, pre- and post-mordant Cu$^{2+}$, pre- and meta-mordant Fe$^{3+}$, and post- and meta-mordant Al$^{3+}$; and the dyes from medium-3 with post-mordant Cr$^{3+}$ and Al$^{3+}$, post- and meta-mordant Cu$^{2+}$, and pre-, post, and meta-mordant Mn$^{2+}$. Other cloth colors, such as grey-brown was produced from post-mordant Cr$^{3+}$ and Mn$^{2+}$, pre- and meta-mordant Fe$^{3+}$, and post-mordant Cu$^{2+}$ in medium-3. Greyed-orange color was generated from pre-, post, and meta-mordant Fe$^{3+}$ in medium-1; post-mordant Fe$^{3+}$ in medium-2; and post-mordant Fe$^{3+}$ in medium-3.
Greyed-yellow color was yielded from pre- and meta-mordant Al$^{3+}$, post- and meta-mordant Cu$^{2+}$, and meta-mordant Cr$^{3+}$ in medium-1; pre-mordant Al$^{3+}$ and meta-mordant Cu$^{2+}$ in medium-2; and pre- and meta-mordant Al$^{3+}$, Cr$^{3+}$, and Cu$^{2+}$ in medium-3. Yellow color was obtained from pre-mordant Ca$^{2+}$ and Cu$^{2+}$, and meta-mordant Mn$^{2+}$ in medium-1; and pre-mordant Ca$^{2+}$ in medium-3. These color differences are due to the reaction of different metal cations in the mordant namely Al$^{3+}$ in KA Al(SO$_4$)$_2$.12H$_2$O, Ca$^{2+}$ in CaCO$_3$, Cr$^{3+}$ in K$_2$Cr$_2$O$_7$, Cu$^{2+}$ in CuSO$_4$.5H$_2$O, Fe$^{2+}$ in FeSO$_4$.7H$_2$O, and Mn$^{2+}$ in MnSO$_4$.H$_2$O. Mongkholtanasit et al. [25] informed that the metal ions of mordant act as electron acceptors for electron donors to form coordination bonds with the dye molecules, making them insoluble in water, while Vankar et al. [26] reported that mordant is metal salts that produce an affinity between the cloth and dye.

**Table 1.** Cloth color in various mordant techniques and types by the dyes from mixed *Aspergillus* and *Paecilomyces* grown in medium-1.

| Mordant technique/type | Dyed cloth |
|------------------------|------------|
|                        | Ca | Cr | Cu | Fe | Mn | Al |
| Pre-mordant            | 12D Yellow | 159D Orange-white | 12D Yellow | 177D Greyed-orange | 159A Orange-white | 161B Greyed-yellow |
| Post-mordant           | 159A Orange-white | 199D Grey-brown | 161C Greyed-yellow | 177C Greyed-orange | 199D Grey-brown | 159A Orange-white |
| Meta-mordant           | 159A Orange-white | 162D Greyed-yellow | 162B Greyed-yellow | 177C Greyed-orange | 12D Yellow | 161C Greyed-yellow |
| Medium-1 control       | 159A Orange-white |
| Cloth control          | 155D White |

Similar results on the cotton cloth color dyed with fungal dyes treated with different mordant types and techniques has been reported. Grey-brown color will be produced when the cloth is dyed with *Aspergillus* dyes treated with pre-mordant Fe$^{3+}$ and *Penicillium* dyes using pre-mordant Fe$^{2+}$ [27]. Greyed-orange color from *Aspergillus* and *Penicillium* dyes each using pre- and post-mordant Al$^{3+}$ and Cu$^{2+}$ [4] and *Aspergillus* and *Penicillium* dyes each with pre-mordant Ca$^{2+}$ and Fe$^{2+}$ [27]. Greyed-yellow color from *A. fumigatus* dyes using pre-mordant Al$^{3+}$ and Cu$^{2+}$ [4] and *Aspergillus* dyes with pre-mordant Al$^{3+}$, Cu$^{2+}$, and Fe$^{2+}$ and *Penicillium* dyes with pre-mordant Ca$^{2+}$ and Cu$^{2+}$ [27]. Orange-white color from *Trichoderma harzianum* dyes using post-mordant Al$^{3+}$ and Cu$^{2+}$ [4] and *Penicillium* dyes with pre-mordant Fe$^{2+}$ [27]; and yellow color from *Penicillium* dyes using pre- and post-mordant Al$^{3+}$ and Cu$^{2+}$ [4].
Based on the variation in color intensity on dyed cotton cloth by the dyes obtained from medium-1 treated with pre-mordant using different mordant types, the sequence of color from highest to lowest were Fe\textsuperscript{2+} > Al\textsuperscript{3+} > Ca\textsuperscript{2+} or Cu\textsuperscript{2+} > Mn\textsuperscript{2+} > Cr\textsuperscript{3+}; with post-mordant were Fe\textsuperscript{2+} > Cu\textsuperscript{2+} or Mn\textsuperscript{2+} > Ca\textsuperscript{2+} or Al\textsuperscript{3+}; and with meta-mordant were Fe\textsuperscript{2+} > Cu\textsuperscript{2+} > Al\textsuperscript{3+} > Mn\textsuperscript{2+} > Cr\textsuperscript{3+} > Ca\textsuperscript{2+}. From medium-2 with pre-mordant were Al\textsuperscript{3+} > Ca\textsuperscript{2+} or Fe\textsuperscript{2+} > Cr\textsuperscript{3+} or Cu\textsuperscript{2+} or Mn\textsuperscript{2+}; with post-mordant were Fe\textsuperscript{2+} > Al\textsuperscript{3+} or Ca\textsuperscript{2+} or Cr\textsuperscript{3+} or Cu\textsuperscript{2+} or Mn\textsuperscript{2+}; and with meta-mordant were Cu\textsuperscript{2+} > Fe\textsuperscript{2+} > Al\textsuperscript{3+} or Ca\textsuperscript{2+} or Cr\textsuperscript{3+} or Mn\textsuperscript{2+}. From medium-3 with pre-mordant were Fe\textsuperscript{2+} > Ca\textsuperscript{2+} > Al\textsuperscript{3+} > Cr\textsuperscript{3+} or Cu\textsuperscript{2+} or Mn\textsuperscript{2+}; with post-mordant were Fe\textsuperscript{2+} > Al\textsuperscript{3+} or Ca\textsuperscript{2+} or Cr\textsuperscript{3+} or Mn\textsuperscript{2+} > Cu\textsuperscript{2+}; and with meta-mordant were Fe\textsuperscript{2+} > Al\textsuperscript{3+} or Cr\textsuperscript{3+} or Cu\textsuperscript{2+} > Ca\textsuperscript{2+} or Mn\textsuperscript{2+}. The study indicated that the sequence of color intensity on the cloth dyed (from highest to lowest) by the dyes obtained from different media treated with different mordant types and techniques were Fe\textsuperscript{2+} > Al\textsuperscript{3+} or Cu\textsuperscript{2+} > Ca\textsuperscript{2+} > Mn\textsuperscript{2+} > Cr\textsuperscript{3+}. There is a tendency of Fe\textsuperscript{2+} makes a strong coordination, therefore, enhances the interaction between the fiber and dye, resulting in a greater depth of shades [28]. Similar results showed that Fe\textsuperscript{2+} produced dark colors on silk and wool fabrics dyed with Eucalyptus leaf extract dyes [25] and cotton cloth dyed with fungal dyes [11; 27]. Al\textsuperscript{3+} and Cu\textsuperscript{2+} each ranks second after Fe\textsuperscript{2+} in producing color intensity on the cloth, but Cu\textsuperscript{2+} produces more varied colors on the cloth (199D greyed-brown, 161C & D greyed-yellow, 162B & D greyed-yellow, 159D orange-white, and 12D yellow) than Al\textsuperscript{3+} (161B, C & D greyed-yellow, 162D greyed-yellow, and 159A & D orange-white (tables 1, 2, and 3). Therefore, Cu\textsuperscript{2+} and Fe\textsuperscript{2+} were selected for further testing.

**Table 2.** Cloth color in various mordant techniques and types dyed with the dyes from mixed *Aspergillus* and *Paecilomyces* grown in medium-2.

| Mordant technique/type | Dyed cloth |
|------------------------|------------|
|                        | Ca        | Cr        | Cu        | Fe        | Mn        | Al        |
| Pre-mordant            |           |           |           |           |           |           |
| 159A                   | Orange-white | 159D    | Orange-white | 159D    | Orange-white | 159D    | 162D      | Greyed-yellow |
| Post-mordant           |           |           |           |           |           |           |
| 159D                   | Orange-white | 159D    | Orange-white | 159D    | Greyed-orange | 159D    | 159D      | Orange-white |
| Meta-mordant           |           |           |           |           |           |           |
| 159D                   | Orange-white | 159D    | Greyed-yellow | 159A    | Orange-white | 159D    | 159D      | Orange-white |
| Medium-2 control       |           |           |           |           |           |           |
| 159D                   | Orange-white | 159D    | Greyed-yellow | 159A    | Orange-white | 159D    | 159D      | Orange-white |
| Cloth control          |           |           |           |           |           |           |
| 155D                   | White     |           |           |           |           |           |

The cloth dyed by the dyes in medium-1 treated with different mordant techniques and types produced ten cloth colors, namely 199D grey-brown, 177C & D greyed-orange, 161B & C greyed-
yellow, 162B & D greyed-yellow, 159A & D orange-white, and 12D yellow (table 1), in medium-2, five diverse colors were obtained on the cloth (165D greyed-orange, 161D greyed-yellow, 162D greyed-yellow, and 159A & D orange-white (table 2), while in medium-3 composed of seven shades on the cloth (199C & D grey-brown, 164D greyed-orange, 161D greyed-yellow, 162D greyed-yellow, 159A orange-white, and 12D yellow (table 3). The results indicated that different mordant techniques and types treated to the dyes cultured in medium-1 (10 colors) produced more varied colors on the cloth than did medium-2 (5 colors) and medium-3 (7 colors). There is a tendency that higher dye absorbance resulted in more varied cloth colors. Medium-1 used to culture the mixed fungi gave the highest affinity dyes production of 1.8382 UA/L than did medium-2 (1.5626 UA/L) and medium-3 (1.7894 UA/L). Therefore, the dyes obtained from medium-1 and medium-3 were used in the next investigation.

The cloth dyed by the dyes in medium-1 treated with pre-mordant using different mordant types produced five colors (177D greyed-orange, 161B greyed-yellow, 159A & D orange-white, and 12D yellow). With post-mordant, four diverse colors were obtained on the cloth (199D grey-brown, 177C greyed-orange, 161C greyed-yellow, and 159A orange-white); while meta-mordant generated six different colors on the cloth (177C greyed-orange, 162B & D greyed-yellow, 161C greyed-yellow, 159A orange-white, and 12D yellow) (table 1). The dyes obtained in medium-2 treated with pre-mordant combined with different mordant types composed of three distinct cloth colors (162D greyed-yellow and 159A & D orange-white). Two different cloth colors generated with post-mordant (165D greyed-orange and 159D orange-white) were obtained, while meta-mordant yielded 161D greyed-yellow and 159A & D orange-white (table 2). The cloth dyed by the dyes in medium-3 treated with pre-mordant using different mordant types yielded five different colors on the cloth (199C grey-brown, 161D & 162D greyed-yellow, 159A orange-white, and 12D yellow). When with post-mordant, it produced three different cloth colors (199D grey-brown, 164D greyed-orange, and 159A orange-white), while with meta-mordant, 199C grey-brown, 162D greyed-yellow, and 159A orange-white was obtained (table 3). The study showed that meta-mordant using different mordant types applied to the dyes obtained from different media produced nine cloth colors (199C greyed-brown, 177C greyed-orange, 161C & D greyed-yellow, 162B & D greyed-yellow, 159A & D orange-white, and 12D yellow). With pre-mordant, eight diverse cloth colors were obtained (199C greyed-brown, 177D greyed-orange, 161D & B greyed-yellow, 162D greyed-yellow, 159A & D orange-white, and 12D yellow); while post-mordant yielded seven cloth colors ((199D greyed-brown, 164D greyed-brown, 165D greyed-brown, 177C greyed-brown, 161C greyed-yellow, and 159A & D orange-white). There is a tendency that meta-mordant result in more varied cloth colors than pre- and post-mordant techniques. It might be because of some sites of the textile materials occupied with the mordant and some directly with the dyes compounds [29] causes more varied cloth colors in meta-mordant. Similar result was reported by Kumaresan [30], which showed that meta-mordant gave better color strength than pre- and post-mordant using different mordant types by the dyes of *Achras sapota* and *Cordia sebestena*. Therefore, pre- and meta-mordant were used in the next testing.

**Table 3.** Cloth color in various mordant techniques and types dyed with the dyes from mixed *Aspergillus* and *Paecilomyces* grown in medium-3.

| Mordant technique/type | Dyed cloth | | | | | |
|------------------------|------------|------------|------------|------------|------------|------------|
| Pre-mordant            | Ca         | Cr         | Cu         | Fe         | Mn         | Al         |
| 12D Yellow             | 162D Greyed-yellow | 162D Greyed-yellow | 199C Grey-brown | 159A Orange-white | 161D Greyed-yellow |
3.3. The effects of dyeing repetition on the colors of cloth

Dyes producing fungi cultured in medium-1 and medium-3 treated with pre- and meta-mordant using Cu\textsuperscript{2+} and Fe\textsuperscript{2+}, were used for the investigation of dyeing repetition.

The cloth color dyed by the dyes in different media combined with different dyeing repetitions (1, 3, 5, and 7 times) using pre-mordant Fe\textsuperscript{2+} is presented in Table 4. The dyeing repetition 1, 3, 5, and 7 times with the dyes in medium-1 using pre-mordant Fe\textsuperscript{2+} each yielded the same color (177D greyed-orange) on the cloth. The dyeing repetition 1, 3, 5, and 7 times with the dyes in medium-3 using pre-mordant Fe\textsuperscript{2+} each also produced the same color (199C grey-brown) on the cloth. Theses indicated that the different dyeing repetitions using the dyes in both media added with pre-mordant Fe\textsuperscript{2+} each produced the same color. Meira [31] informed that when the dyeing process enters the maximum into the dyed material, it produced the color aging of textile materials. The more repetitions of dyeing, the more dyes were absorbed on the cloth. In this study, the cloth fibers began to saturate with the dyes in both media in the dyeing repetition 1 time, so that the next dyeing (3, 5 and 7 times) of cloth fibers, did not have ability or ineffective to absorb the dyes. In other words, the optimum condition for the cloth dyeing by the dyes in both media added with pre-mordant Fe\textsuperscript{2+} each was 1 time. The cloth dyeing repetition by the dyes in both media added with pre-mordant Fe\textsuperscript{2+} is not necessary, because a strong cloth color has been obtained or the cloth is saturated with the dyes at dyeing 1 time.

The cloth color dyed by the dyes in different media added with different dyeing repetitions using meta-mordant Fe\textsuperscript{2+} is shown in Table 5. The dyeing repetition 1, 3, 5, and 7 times with the dyes in medium-1 using meta-mordant Fe\textsuperscript{2+} each yielded the same color (177C greyed-orange) on the cloth. The dyeing repetition 1 and 3 times with the dyes in medium-3 using meta-mordant Fe\textsuperscript{2+} each yielded the same lighter color (199C grey-brown) on the cloth than the dyeing repetition 5 and 7 times (each 199B grey-brown). The results showed that the optimum condition for the cloth dyeing by the dyes from medium-1 added with meta-mordant Fe\textsuperscript{2+} was 1 time, while from medium-3 was 5 times. A different result was reported by Suciatmih [11], which showed that the optimum condition for the cloth dyeing by the dyes from the same mixed fungi cultured in the Backer and Tatum medium [12] with meta-mordant Fe\textsuperscript{2+} was 6 times. The cloth dyeing repetition by the dyes in medium-1 treated with meta-mordant Fe\textsuperscript{2+} is not necessary, because a strong cloth color has been obtained or the cloth is saturated with the dyes at dyeing 1 time. While the cloth dyeing repetition 5 times by the dyes in medium-3 treated with meta-mordant Fe\textsuperscript{2+} must be done in order to get a strong cloth color or the cloth is saturated with the dyes.
Table 4. Cloth color in various dyeing replications with pre-mordant Fe\textsuperscript{2+} by the dyes from mixed Aspergillus and Paecilomyces grown in different media.

| Medium/Dyeing replication | Dyed cloth                  |
|---------------------------|----------------------------|
| Medium-1                  |                            |
| 1                         | 177D Greyed-orange          |
| 3                         | 177D Greyed-orange          |
| 5                         | 177D Greyed-orange          |
| 7                         | 177D Greyed-orange          |
| Medium-3                  |                            |
| 1                         | 199C Grey-brown             |
| 3                         | 199C Grey-brown             |
| 5                         | 199C Grey-brown             |
| 7                         | 199C Grey-brown             |

Table 5. Cloth color in various dyeing replications with meta-mordant Fe\textsuperscript{2+} by the dyes from mixed Aspergillus and Paecilomyces grown in different media.

| Medium/Dyeing replication | Dyed cloth                  |
|---------------------------|----------------------------|
| Medium-1                  |                            |
| 1                         | 177C Greyed-orange          |
| 3                         | 177C Greyed-orange          |
| 5                         | 177C Greyed-orange          |
| 7                         | 177C Greyed-orange          |
| Medium-3                  |                            |
| 1                         | 199C Grey-brown             |
| 3                         | 199C Grey-brown             |
| 5                         | 199B Grey-brown             |
| 7                         | 199B Grey-brown             |

The cloth color dyed by the dyes in different media combined with different dyeing repetitions using pre-mordant Cu\textsuperscript{2+} is presented in table 6. The dyeing repetition 1 time with the dyes in medium-1 using pre-mordant Cu\textsuperscript{2+} yielded 161B greyed-yellow on the cloth, while the dyeing repetition 3, 5, and 7 each produced the same brighter colors (162B greyed-yellow). The dyeing repetition 1 time with the dyes in medium-3 using pre-mordant Cu\textsuperscript{2+} produced lighter colors on the cloth (162D greyed-yellow) than the dyeing repetition 3, 5, and 7 times (162B greyed-yellow). The studies indicated that the optimum condition for the cloth dyeing by the dyes in medium-1 and medium-3 added with pre-mordant Cu\textsuperscript{2+} each was 3 times. It is necessary to repeat the cloth dyeing 3 times by the dyes in both media added with pre-mordant Cu\textsuperscript{2+} in order to obtain a strong cloth color or the cloth saturated with the dyes.

The cloth color dyed by the dyes in different media treated with different dyeing repetitions using meta-mordant Cu\textsuperscript{2+} is shown in table 7. The dyeing repetition 1 time with the dyes in medium-1 using meta-mordant Cu\textsuperscript{2+} yielded lighter colors (162B greyed-yellow) on the cloth than the dyeing repetition 3, 5, and 7 each produced the same darker color (162A greyed-yellow). The dyeing repetition 1 and 3 times with the dyes in medium-3 using meta-mordant Cu\textsuperscript{2+} each produced lighter colors (162D greyed-yellow) on the cloth than the dyeing repetition 5 and 7 each produced the same darker color (161C greyed-yellow). The studies showed that the optimum condition for the cloth dyeing by the dyes in medium-1 and medium-3 added with meta-mordant Cu\textsuperscript{2+} was 3 and 5 times respectively. It is necessary to repeat the cloth dyeing 3 times by the dyes in medium-1 treated with meta-mordant Cu\textsuperscript{2+} in order to acquire a strong cloth color or the cloth saturated with the dyes, while in medium-3 it needs 5 times.
Table 6. Cloth color in various dyeing replications with pre-mordant Cu\(^{2+}\) by the dyes from mixed *Aspergillus* and *Paecilomyces* grown in different media.

| Medium/dyeing replication | Dyed cloth  |
|---------------------------|-------------|
| Medium-1                  | 161B Greyed-yellow | 162B Greyed-yellow | 162B Greyed-yellow | 162B Greyed-yellow |
| Medium-3                  | 162D Greyed-yellow | 161B Greyed-yellow | 161B Greyed-yellow | 161B Greyed-yellow |

Table 7. Cloth color in various dyeing replications with the meta-mordant Cu\(^{2+}\) by the dyes from mixed *Aspergillus* and *Paecilomyces* grown in different media.

| Medium/dyeing replication | Dyed cloth  |
|---------------------------|-------------|
| Medium-1                  | 162B Greyed-yellow | 162A Greyed-yellow | 162A Greyed-yellow | 162A Greyed-yellow |
| Medium-3                  | 162D Greyed-yellow | 162D Greyed-yellow | 161C Greyed-yellow | 161C Greyed-yellow |

4. Conclusion
Colors that appear on the cloth dyed by the dyes from mixed *Aspergillus* and *Paecilomyces* cultured in different media using different mordant types and techniques as well as dyeing repetitions, adding color variations to textile dyeing. To get 162A greyed-yellow color, cloths have to be dyed with the combination of the dyes obtained from medium-1, meta-mordant Cu\(^{2+}\) as well as dyeing repetition 3 times, while 161B greyed-yellow was obtained from the combination of the dyes obtained from medium-3, pre-mordant Cu\(^{2+}\) as well as dyeing repetition 3 times. To get 177C greyed-orange color, cloths have to be dyed with the combination of the dyes obtained from medium-1, meta-mordant Fe\(^{3+}\) as well as dyeing repetition 1 time, while 199B grey-brown color was produced from the combination of the dyes obtained from medium-3, meta-mordant Fe\(^{3+}\) as well as dyeing repetition 5 times.

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