APPLICATION OF NATURAL-BASED ANTI-FUNGAL FORMULATIONS FOR BAMBOO AND WOODEN HOUSEHOLD PRODUCTS

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ABSTRACT

This paper is concerned with the ability of natural products such as vinegar, tea, okra, lemon juice, grapefruit peel essential oil, and baking soda to prevent mold growth on bamboo and wood materials. The results showed that grapefruit peel essential oil had the highest biological activity compared to the other products in preventing mold growth on wood materials. Additionally, a process for preventing mold growth on bamboo and wood materials was developed and the parameters were optimized using the Box-Behnken surface response correlation method. The optimal process conditions were determined by analyzing the response surface of the three-dimensional surface plot and solving the regression model equation using Design Expert software. The optimal conditions, including the ratio of product to coverage area 5/1, concentration 53%, time in 238 minutes and temperature 40^oC, have been tested and showed that the time for mold appearance was 254 days.

Keywords: Bamboo and wood materials; anti-mold; household appliances; bio-based products.

1. Introduction

Using household items from bamboo and wood materials and developing, using bamboo wood has become a new industry. However, mold often appears on bamboo and wood substrates and bamboo and wood processing materials during storage, processing, transportation and use, leading to surface pigment contamination that affects the quality of bamboo and wood. causing damage to businesses and consumers. However, due to many subjective and objective reasons, research on anti-mold forest product preservatives as well as types of anti-mold forest product preservatives is still limited.

Until 1998, the Forest Product Preservation Research Department, Vietnam Academy of Forest Sciences officially registered 13 types of forest product preservatives, including XM5 and PPB, which are preservatives capable of high fungus prevention. Because of its effect on the color of forest products after impregnation, XM5 is rarely used as an anti-fungal drug for forest products used for handicrafts and household furniture. As for PBB, in 2002, it was banned from use according to the decision on environmental protection and safety for human health issued by the Plant Protection Department, Ministry of Agriculture and Rural Development due to the presence of pentachlorophenolate sodium, an ingredient that has not been allowed to be used in Vietnam since 2002.

To replace this deficiency and meet the needs of actual production, it is necessary to conduct research to create a new alternative anti-mold drug, with drug ingredients that are not on the list of banned and restricted chemicals used in Vietnam.

Some preparations with antifungal activity of natural origin such as vinegar, baking soda, soapberry, tea, lemon juice, and grapefruit peel essential oil are widely used in life. Inhibition and anti-mold properties on bamboo and wood materials.

This article studies and evaluates the anti-mold ability of some biological products on bamboo and wood objects. Conditions affecting the anti-mold activity of the product such as product/coated area ratio; The inoculant coating time and inoculant concentration have been studied to find the optimal conditions for the anti-mold process on wood and bamboo when using the inoculant. With the purpose of applying products of natural origin to anti-mold and antibacterial for bamboo and wood products in practice. The process aims to bring a product that is safe for humans, especially children, and environmentally friendly, minimizing the use of toxic products in the production of consumer products friendly with human.

2. Experimental set-up

2.1. Materials and chemicals

The acacia wood samples have been pre-treated and are suitable for use as household products. Pure analytical chemicals, deionized distilled water, 1% phenolphthalein, ethanol, methanol, sodium hydroxide, sodium chloride, potassium hydroxide, sulfuric acid, sodium sulfate.

2.2. Research methods

- Determine the anti-mold activity and effectiveness of the product against mold

Evaluate effectiveness against mold. We proceed as follows: Samples after being processed, impregnated, allowed to dry naturally and placed in boxes in order of each concentration level and control sample. After a period of one month, the sample boxes are evaluated. Evaluation criteria: the effectiveness of the preservative product on mushrooms is evaluated by scoring based on three comparison criteria between the sample impregnated with the product and the control sample: Discolored area, soft rot area and firmness, sample mass loss.

- Testing method for anti-mold substances in controlling mold and fungal stains on wood (GB/T 18261-2013).

To avoid a too long research period due to different weather and environmental temperature mold conditions, when experimenting, we inoculated Aspergillus mold strains on products coated with the product (previously conducted). Conducting a control experiment with a mold culture sample and letting it mold naturally, the time to naturally mold is 210 days).

- Level 2 orthogonal experimental planning method. Box-Behnken model with factors: ratio of inoculant/coverage area, time and inoculant concentration.

The anti-mold experimental process on bamboo and wood materials was conducted at the Food Technology Laboratory - Vinh University Center for Experiments and Practice.

2.2. Methods of experimental arrangement

- Determine the effects of different preparations

The experiment was conducted with preparations at 50% concentration including: vinegar, baking soda, lemon juice, grapefruit peel essential oil, soapberry, and tea.

Wood from Acacia trees has been chosen to conduct experiments with the same side wooden blocks (in thickness, length and width). The best preparation (E) will be chosen for the next experiment.

- Determine the effect of product/coating area ratio

The experiment was conducted with a ratio range (product volume/area ml/cm^2): 1/1 (100 ml/100 cm^2); 2/1 (200 ml/100 cm^2); 3/1 (300 ml/100 cm^2); 4/1 (400 ml/100 cm^2); 5/1 (500 ml/100 cm^2); 6/1 (600 ml/100 cm^2); 7/1 (700 ml/100 cm^2); 8/1 (800 ml/100 cm^2). The product/area ratio factor (A) will be determined for the next experiment.

- Determine the influence of inoculant coating time

The product was coated with the following times: 60, 120, 180, 240, 300 and 360 minutes. The area of the sample is 100 cm^2, inoculant used (E), inoculant/area ratio (A). From there, the appropriate coating time for product (B) can be determined for subsequent experiments.

- Determine the effect of product concentration

The following concentration parameters have been applied: 40%; 50%; 60%; 70%; 80%; 90% and pure. Sample area is 100 cm^2, preparation used (E), ratio of preparation/area (A), coating time (B). From there, the appropriate concentration (C) can be determined for subsequent experiments.

- Determine the effect of product temperature

To determine the effect of temperature on the anti-mold ability of bamboo and wood products, experiments were conducted with concentration parameters of

$30^oC, 40^oC, 50^oC, 60^oC$ and 70^oC. Sample area is 100 cm^2, preparation used (E), ratio of preparation/area (A), coating time (B, concentration (C). After coating the product, we will determine the best anti-mold ability for bamboo and wood products to choose the appropriate temperature (D) for the next experiments.

- Experimental setup to optimize the application process of anti-mold products using the response surface method with the Box-Behnken model

In this experiment, four factors including product/area ratio (A), product coating time (B), product concentration (C) and temperature (D) were selected to perform process optimization. The objective function is the number of days before the appearance of mold.

3. Result and discussion

3.1. Anti-mold activity of the preparations

The wooden bars are soaked in the pure product for 60 minutes before being removed to dry. Experiments were repeated 3 times. The time for mold appearance corresponding to each product is shown in Table 1. The results showed that, with the same concentration of the preparation, the anti-mold ability of the essential oil extracted from grapefruit peel gave the longest time for mold to appear. Therefore, the essential oil extracted from grapefruit peel was chosen for the next experiments.

Table 1: *Mold appearance time corresponding to each preparation*

Preparations	Lemon juice	Vinegar	Grapefruit peel essential oil	Extract from locust	Tea	Baking soda
Concentration	50%	50%	50%	50%	50%	50%
Time of mold appearance	42 days	35 days	56 days	39 days	42 days	36 days

3.2. Effect of product/coating area ratio

The following scale ranges (volume of product/area ml/cm^2) had been applied: 1/1 (100ml/100 cm^2); 2/1 (200 ml/100 cm^2); 3/1 (300 ml/100 cm^2); 4/1 (400 ml/100 cm^2); 5/1 (500 ml/100 cm^2); 6/1 (600 ml/100 cm^2); 7/1 (700 ml/100 cm^2); 8/1 (800 ml/100 cm^2). The results are presented in Table 2.

Table 2: *Mold appearance time corresponding to product/coating area ratio*

Product/coating area ratio	**1/1**	**2/1**	**3/1**	**4/1**	**5/1**	**6/1**	**7/1**	**8/1**
Time of mold appearance	42 days	56 days	58 days	60 days	63 days	65 days	65 days	65 days

The results show that with a ratio of 6/1 or higher, the appearance of mold on the surface takes the longest time. Therefore, the ratio 6/1 was chosen for further experiments.

3.3. Effect of coating time

The following coating times were applied: From 60 minutes to 360 minutes. The experimental results are shown in Table 3.

Table 3: *Mold appearance time corresponding to coating time*

Coating time	60 minutes	120 minutes	180 minutes	240 minutes	300 minutes	360 minutes	60 minutes	120 minutes
Time of mold appearance	65 days	73 days	96 days	>120 days	120 days	120 days	65 days	73 days

Table 3 shows that a sample soaking (coating) time of 240 minutes gives the most optimal results, reflected in the longest mold appearance time, exceeding 120 days. Therefore, a time period of 240 min was applied for subsequent experiments.

3.4. Effect of the concentration

The experiment was conducted with concentration ranges from 40% to pure, the product was diluted in the solvent of absolute alcohol. The results are presented in Table 4.

Table 4: *Mold appearance time corresponding to the concentration*

Concentration (%)	**40**	**50**	**60**	**70**	**80**	**90**	**Pure**
Time of mold appearance	128 days	152 days	>190 days	181 days	180 days	188 days	185 days

From the results table, it can be seen that with a product concentration of 60%, the mold appearance time is more than 190 days, which is the most optimal time. Therefore, the product concentration of 60% was selected for the next experiments.

3.5. Effect of temperature

The temperature range of the product from 300C to 700C has been applied and obtained the results in the following table:

Table 5: *Mold appearance time corresponding to the temperature of grapefruit peel essential oil preparation*

Temperature (^{0}C)	**30**	**40**	**50**	**60**	**70**
Time of mold appearance	190 days	>252 days	201 days	151 days	125 days

With the experimental results obtained in Table 5, the appropriate temperature of the product when soaking the specimen is 40^{0}C for subsequent experiments.

3.6. Experimental results of multi-factor influence

From the results of the influence of each single factor on the anti-mold ability of essential oils extracted from grapefruit peel, experiments to determine the influence of multiple factors have been implemented and from there an optimal treatment process has been developed.

Table 6: *Factors affecting the anti-mold ability of grapefruit peel essential oil preparation*

Influential factors	**Level**			**Variation Range**
	-1	0	+1	
Volume of product/area X1 (ml/cm2)	5/1	6/1	7/1	1/1
Time X2 (minute)	180	240	300	60
Concentration X3 (%)	50	60	70	10
Temperature X4 (°C)	30	40	50	10

The experiment was arranged according to Box-Behnken quadratic planning, with k = 4 and included 29 experiments, including 5 experiments at the center, shown in Table 7 below.

Table 7: *Experimental setup to optimize the application process of anti-mold products for bamboo and wood products*

No	A: volume of product/area (ml/cm^2)	B: time (minutes)	C: concentration (%)	D: temperature (^0C)	Time of mold appearance (days)
1	-1.000	0.000	0.000	0.000	246
2	0.000	-1.000	0.000	0.000	248
3	-1.000	1.000	0.000	0.000	235
4	1.000	1.000	0.000	0.000	236
5	0.000	0.000	-1.000	0.000	251
6	0.000	0.000	1.000	0.000	250
7	0.000	0.000	-1.000	1.000	244
8	0.000	0.000	1.000	1.000	242
9	-1.000	0.000	0.000	-1.000	230
10	1.000	1.000	-1.000	-1.000	203
11	-1.000	0.000	0.000	1.000	239
12	1.000	0.000	0.000	1.000	233
13	1.000	-1.000	-1.000	1.000	205
14	1.000	1.000	-1.000	1.000	210
15	-1.000	-1.000	1.000	-1.000	209
16	0.000	1.000	1.000	0.000	235
17	-1.000	-1.000	0.000	0.000	243
18	1.000	0.000	-1.000	1.000	228
19	-1.000	-1.000	1.000	0.000	227
20	1.000	0.000	1.000	0.000	241
21	1.000	-1.000	0.000	-1.000	214
22	1.000	1.000	-1.000	-1.000	211
23	0.000	-1.000	0.000	1.000	227
24	-1.000	0.000	1.000	1.000	233
25	0.000	0.000	0.000	0.000	252
26	0.000	0.000	0.000	0.000	254
27	0.000	0.000	0.000	0.000	253
28	0.000	0.000	0.000	0.000	253
29	0.000	0.000	0.000	0.000	251

Evaluation results are recorded daily according to sensory standards of mold growth factors, the beginning of mold appearance with tiny spots that can be counted with the naked eye.

Figure 1: *Some experimental images*

The fit level of the model was analyzed, thereby evaluating the meaningfulness of the model through ANOVA analysis (Table 8) and correlation indices (Table 9). The significance of the regression coefficients was evaluated by the F standard, with p values <0.05 indicating significant regression coefficients. The "Model-F-value" obtained is 336.37, proving that the model is completely statistically significant with 97% confidence (p<0.0001). For all factors including product/coverage ratio, time, concentration, temperature and each pair of these factors, p < 0.05 for each pair of factors was significant. The F standard for "incompatibility" of the model is 2.25 (p=0.1927), which proves that the model is completely compatible with experiment. The results of ANOVA analysis show the value R^2 is 0,9732 (R-Squared) in Table 9, which proves that the experimental value is close to the predicted value of the model.

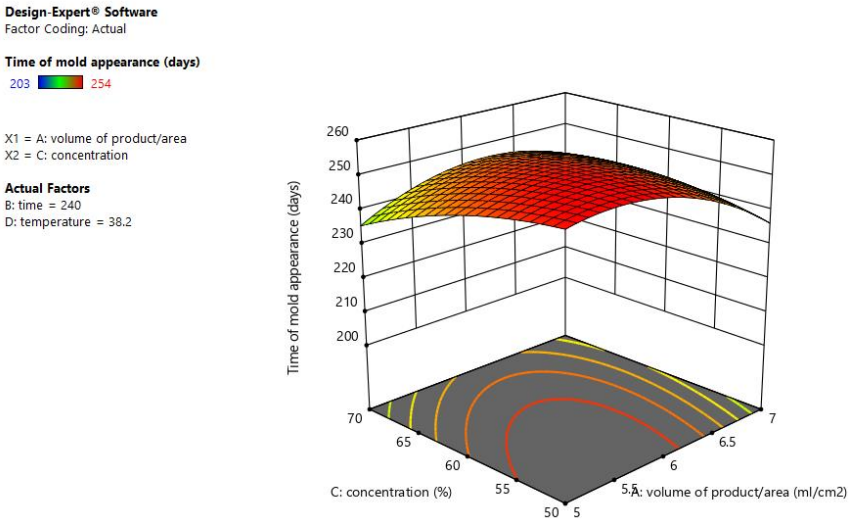
Table 8: *Results of ANOVA analysis to optimize the synthesis of factors*

Factors	Sum of squares	df	Mean square	F-value	p-value	
Model	6831.22	14	487.94	36.37	< 0.0001	significant
A- volume of product/area	98.64	1	98.64	7.35	0.0169	
B-time	12.95	1	12.95	0.9656	0.3425	
C-concentration	36.27	1	36.27	2.70	0.1224	
D-temperature	159.59	1	159.59	11.90	0.0039	
AB	49.18	1	49.18	3.67	0.0762	
AC	79.05	1	79.05	5.89	0.0293	
AD	4.38	1	4.38	0.3266	0.5767	
BC	13.92	1	13.92	1.04	0.3257	
BD	41.68	1	41.68	3.11	0.0998	
CD	21.77	1	21.77	1.62	0.2234	
A^2	278.51	1	278.51	20.76	0.0004	
B^2	633.85	1	633.85	47.25	< 0.0001	
C^2	40.64	1	40.64	3.03	0.1037	
D^2	730.73	1	730.73	54.47	< 0.0001	

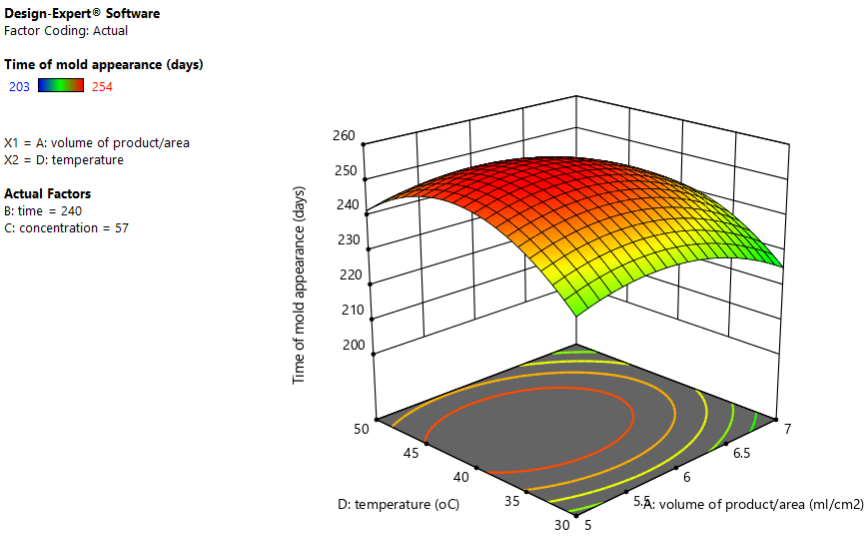
Factors	Sum of squares	df	Mean square	F-value	p-value	
Residual	187.81	14	13.42			
Lack of fit	150.61	9	16.73	2.25	0.1927	Not significant
Pure error	37.20	5	7.44			
Cor total	7019.03	28				

Table 9: *Analytical results of the model's agreement with experiments*

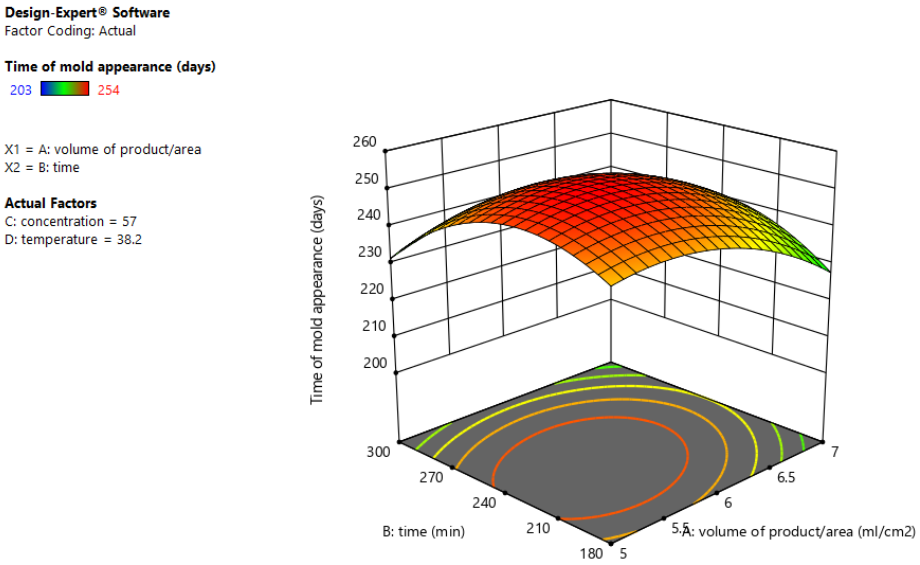
Std. Dev.	3.66	R^2	0.9732
Mean	234.59	Adjusted R^2	0.9465
C.V. %	1.56	Predicted R^2	0.7850
		Adeq Precision	17.7878



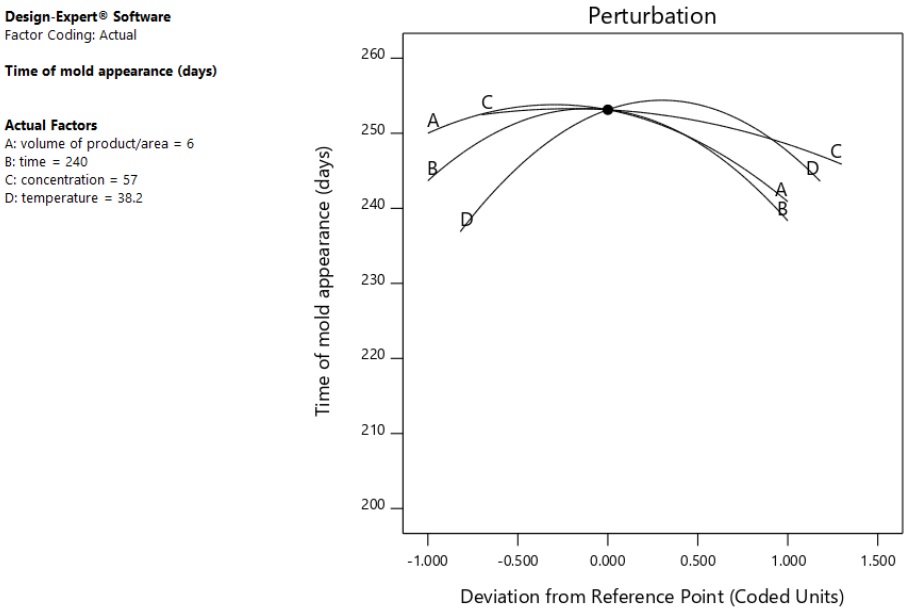
(a) Volume of product/area and concentration



(b) Volume of product/area and temperature



(c) Volume of product/area and time



(d) The simultaneous impact of three factors

Figure 2: *Influence of factors on mold appearance time on bamboo and wooden materials*

From the meaningful analytical values above, the expected function value is expressed according to the following specific equation:

$$Y = -79.26722 + 45.93217A + 0.739585B - 1.953C + 8.81368D + 0.059922AB + 0.546222AC - 0.097135AD + 0.004328BC + 0.005972BD + 0.032112CD - 7.69151A^2 - 0.003371B^2 - 0.139478C^2$$

In which, Y is the mold appearance time; A, B, C are the product/coating area ratio, time, concentration and temperature, respectively.

Expectation function model

The use of grapefruit peel essential oil preparations has been researched to determine the longest mold growth time. Optimization results using Design-Expert 10.1 software provide a number of optimal options as in Figure 3.

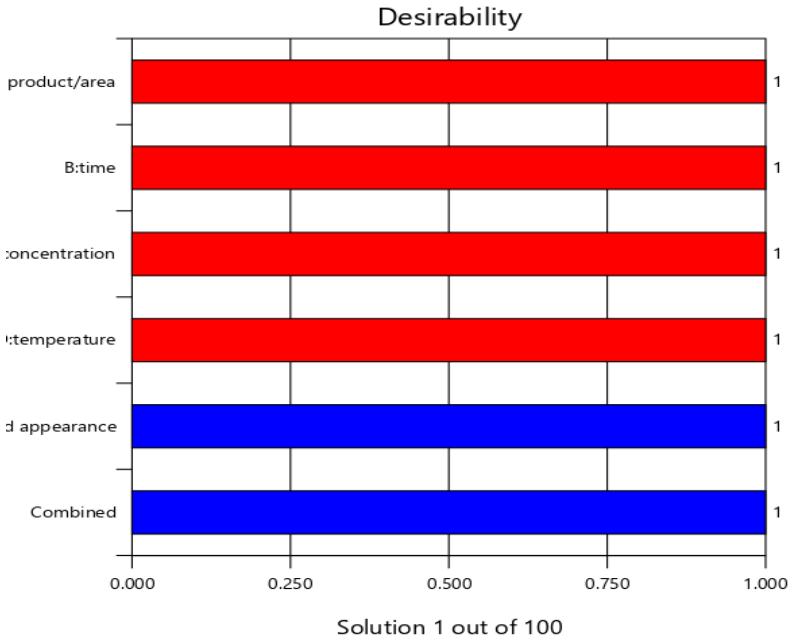


Figure 3: *Degree of meeting expectations of anti-mold treatment by grapefruit peel essential oil*

From the experimental results of the linear regression equation above, the optimization results are obtained as follows: Product/coverage ratio 5.992/1 (ml/cm^2), time 244.963 minutes, concentration 61.67 % and temperature 43.287°C. Applying these optimization parameters, the time from which the mold appeared was recorded as 254.33 days.

To verify the experimental results obtained above, the anti-mold ability of the product according to the optimal results has been experimentally verified, shown in Table 10.

Table 10: *Experimental results to verify the anti-mold ability of the product according to optimal results*

No	Volume of product/area (ml/cm^2)	Time (minutes)	Concentration (%)	Temperature (°C)	Time of mold appearance
1	6/1	245	62	43	254
2	6/1	245	62	43	254
3	6/1	245	62	43	255
Average					254.33±0.33

4. Conclusions

From the research results on the anti-mold ability of products of natural origin for bamboo and wood products, an essential oil extracted from grapefruit peel had been selected. The appropriate ratio of product/anti-mold coating area is 600 ml/ 100 cm^2. The coating time for the anti-mold product in accordance with the experimental product is 245 minutes. The concentration of the anti-mold preparation is consistent with the experimental product of 62% grapefruit peel essential oil in alcohol. The appropriate preparation temperature is 43°C. Optimizing the process of using biological products (essential oils extracted from grapefruit peels) to prevent mold, suitable for some products made from wood, is 254 days.

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TÓM TẮT

ỨNG DỤNG CÔNG THỨC KHÁNG NẤM TỪ THIÊN NHIÊN CHO SẢN PHẨM TRE, GỖ GIA DỤNG

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Bài viết trình bày kết quả nghiên cứu về khả năng chống nấm mốc trên vật liệu tre, gỗ của các chế phẩm có nguồn gốc tự nhiên: giấm, chè, bồ kết, nước cốt chanh, tinh dầu vỏ bưởi, thuốc muối. Kết quả cho thấy tinh dầu vỏ bưởi có hoạt tính sinh học cao nhất so với các chế phẩm còn lại trong việc chống nấm mốc trên vật liệu tre, gỗ. Đồng thời, xây dựng được quy trình chống nấm mốc trên vật liệu tre, gỗ, tối ưu hóa các thông số trong quy trình chống mốc bằng phương pháp đáp ứng bề mặt Box-Behnken. Các điều kiện quá trình tối ưu được xác định bằng cách phân tích bề mặt đáp ứng của biểu đồ bề mặt ba chiều và giải phương trình mô hình hồi quy bằng phần mềm Design Expert. Các điều kiện tối ưu bao gồm: tỷ lệ chế phẩm/diện tích phủ 6/1, nồng độ 62%, thời gian 245 phút, nhiệt độ 43^oC đã được thực nghiệm cho thấy thời gian xuất hiện mốc là 254 ngày.

Từ khóa: Vật liệu tre gỗ; chống nấm mốc; đồ gia dụng; chế phẩm sinh học.