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Development of Local Microorganism from Organic Waste as an Alternative Product for EM4

Fuji Astuti Febria*,and Siski Rahayu

Department of Biology, Faculty of Mathematics and Natural Sciences Andalas University
Padang 25163, West Sumatra
*Corresponding Author : fujiastutifebria@sci.unand.ac.id

Abstract— Research of Development Local Microorganism from Organic Waste by Adding Banana Peels as an Alternative Product for EM4 was conducted in the Laboratory Riset of Microbiology, Biology Department, Andalas University from February 2019 until July 2019. This study aims to find microbial groups and to determine the total amount of microbes in Local Microorganisms and to determine the quality of compost that produced by Local Microorganisms and EM4 as a control. This research design by survey method to sampling and experimental method to produced Local Microorganisms. The inoculation of bacteria colony was using Potato Dextro Agar (PDA) whereas inoculation of yeast colony using Malt Extract Agar (MEA). The result of this research showed that bacterial colonies on M4 2.8x 10^4 cfu/mL, fungi colonies 3.5 x 10^3 cfu/mL and yeast colonies 1.7 x 10^4 cfu/mL. The quality of compost produced has different color and texture however has the same in the aroma parameter.

Keywords – Bacteri, compost, fungi, local microorgasim, yeast.

I. INTRODUCTION

The growing and rapidly growing population in the world have resulted in increasingly large and dense residential areas. One of the problems caused by the increase in the high population is the increase for waste. The greater the number of the human population with various activities carried out, the greater the garbage that will be produced. Dhanya et al., (2020) explained, the garbage that is not managed properly, besides disturbing the view (aesthetics) of the environment, garbage can also cause several diseases, such as vomiting, diarrhea, and respiratory tract infection. Trash can also pollute water, pollute the soil, and cause flooding. Toni et al., (2020) explains that one of the biggest contributors to waste is traditional markets. The market waste composition is more dominant than organic waste that is biodegradable waste. Because of a large amount of waste in the traditional market, it often found that garbage generated by activities in the market, this should be a concern of the market sellers, market managers and the community, where the daily landfill will disrupt health, cleanliness and pollute the environment. According to Pavi et al., (2017) market vegetable waste is discarded material in an effort to improve the appearance of merchandise in the form of vegetables to be marketed. Vegetable waste usually consists of ingredients that have a considerable amount of water, making it easy to rot quickly. Vegetable waste that did not managed properly could result in reduced quality of sanitation, environmental pollution, and the emergence of certain diseases.

Processing of solid waste in the form of vegetables needs to be done, one way to treat solid waste is by making compost. Compost is organic fertilizer, the use of organic fertilizer is very widely used because it has 3 advantages, namely: benefits for the environment, soil and for plants. Compost is very helpful in solving environmental problems, especially waste. The raw material for composting is garbage, so the problem of house waste and municipal waste could be overcome. For soil, compost can add nutrients and can improve soil structure and texture and store water. Thus, the better the quality of the soil and supported by sufficient

nutrients, the plants will produce optimal production (Rich et al., 2017). According to Chen et al., (2019), decomposition in the open environment, organic fertilizer can be formed by itself. The decay process occurs naturally but not in a fast time, but gradually. The stages that go through are natural processes, grass, leaves and animal feces, and other debris that are decaying due to factors of radiation and weather. The decay process occurs for around 5 weeks to 2 months. Composting in an aerobic system can make approximately two-thirds of carbon (C) evaporate (into CO) and the remaining one-third of the part reacts with nitrogen (N) in living cells. During the aerobic composting process, it does not cause a foul odor. During the decomposition process, it will be exothermic so that it causes heat due to the release of energy. The increase in temperature in the accumulation of organic material benefits thermophilic microorganisms. However, if the temperature exceeds 65-70℃, the activity of microorganisms will decrease due to the death of organisms due to high heat.

Local microorganisms are organic fertilizers that rely on local microorganisms. Local Microorganism also often called Liquid Organic Fertilizers. Local Microorganisms can be another alternative as an effort to free plants from adverse effects is the chemical residue that been used by the community to fertilize the soil (Mirwandono et al., 2018). Local Microorganism Solution made from natural ingredients, as a living medium and the development of microorganisms that are useful for accelerating the destruction of organic matter. Local Microorganism can also be called a bio activator that consists of several collections of microorganisms by utilizing the potential of local natural resources. Local Microorganism can function as a breaker of organic matter and as a liquid fertilizer through a fermentation process. The main factors causing the widespread use of chemical fertilizers are easy to find or obtain, fast response and complete nutrients (Mohammadizadeh et al., 2020). Effective Microorganism 4 (EM4) is a mixture of microorganism inoculants (Lactobacillus, actinomycetes, yeast, photosynthetic bacteria, and cellulose decomposers) that can accelerate the maturity of organic fertilizers in composting or decomposing organic matter. The fermentation of organic matter by EM4 microbes takes place in semi-aerobic and anaerobic conditions at a temperature of 40-50℃ (Mirwandono et al., 2018).

II. Research Methodology

Provision of Organic Waste, Vegetables and Fruits

The materials that used in this study are market and domestic organic waste (vegetables, tomatoes, bananas, sugar cane, bones, Leguminosa plants, and associated washing water) obtained from homes, landfills, and traders in Pasar Raya Kota Padang or in Pasar

Making Local Microorganisms

Making Local Microorganisms refers to expert consultation carried out through four stages, namely:

Stage I making Local Microorganisms (M1)

The first stage in making Local Microorganisms is a blender of organic waste and added with banana skin, brown sugar, coffee pulp with a ratio of 2: 0.25: 1: 1 and added as much as 5L of rice water. Then incubated for 7 days then filtered and produced drunk Local Microorganisms 1 (M1).

Stage II making Local Microorganisms (M2)

For the second stage, the formation of Local Microorganisms is prepared a mixture of organic waste coupled with banana peel, eggshell with a ratio of 2: 2: 1. Then put into a container that has contained the results of the previous stage Local Microorganisms filter (M1). Then incubated for 7 days, after that the fermentation product was filtered so that Local Microorganisms stage 2 (M2) was obtained.

Stage III making Local Microorganisms (M3)

For the third stage, the formation of Local Microorganisms is prepared a mixture of vegetables and fruits, nuts, and banana peels with a ratio of 1: 1: 1. Then put into a container that has contained the results of the previous Local Microorganisms filter (M2). Then it was incubated for 7 days, after which the fermented product was filtered to obtain Local Microorganisms stage 3 (M3).

Stage IV making Local Microorganisms (M4)

For the fourth stage, the formation of Local Microorganisms is prepared with a mixture of banana and vegetable skins and fruits with a ratio of 1: 1. Then it put into a container that contains the results of the Local Microorganisms stage 3 filter (M3). Then incubated for 7 days, after that the fermentation product was filtered so that Local Microorganisms stage 4 (M4) was obtained.

pH Measurement of Solution for Local Microorganisms

PH measurement during anaerobic process carried out in the stages of forming a solution of local microorganisms by taking a small sample solution and measuring it with a pH meter paper.

Measurement of Temperature of Product for Local Microorganisms

The temperature measurement of a local microorganism solution aims to determine the temperature of each stage of formation of a local microorganism solution using a thermometer that measured when filtering samples of local microorganism solutions.

Detection of Microbial Groups and Total Microbes in Local Microorganisms Products

Microbial isolation is carried out after each screening at each stage of making Local Microorganisms. The isolation carried out refers to Cappuccino and Welsh (2017), by taking 1 mL of the sample then doing multilevel dilutions, namely, 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, and 10^{-6} next planted by the pour plate method into the medium NA, PDA, and MEA. The microbial group observed and calculated the number of colonies growing.

Making Composting

Making composting refers to Mohammadizadeh et al., (2020) by arranging intermittent organic waste. In this study used market organic waste and agro with a ratio of 1: 1. Market and agro organic waste is chopped first, then put in a bucket. Then splashed with Local Microorganisms (M4) evenly and tightly closed. Then EM4 in a different bucket with a ratio of 5: 1. Then left for 14 days. Next is the observation of pH, temperature and compost characteristics (texture, color and scent).

III. Result And Discussion

Total microbial colonies of Local Microorganisms

In this study, organic waste (vegetables and fruits) was added with banana peels. Organic waste is one of the potential raw materials to produce local microorganisms. During the process of making Local Microorganisms, the presence of bacterial, fungal and yeast colonies is very important. The fermentation process of making Local Microorganisms is principally the same as EM4, which carried out for 4 weeks fermentation. The presence and number of colonies of each microbial colony in the fermentation process of making Local Microorganisms shown in Table 1, as follows:

Table 1. Number of microbial presence in the process of making Local Microorganisms

	Number Of Colony (*cfu/ml*)		
	Bacteri	Fungi	Yeast
M 1	9.5×10^3	5.4×10^4	1.3×10^4
M 2	8.4×10^3	7.8×10^4	1.32×10^5
M 3	1.5×10^5	9.3×10^4	1.6×10^5
M 4	1.8×10^5	9.6×10^4	1.9×10^5

Description:
M1: Phase 1 fermentation process in making Local Microorganisms
M2: Phase 2 fermentation process in making Local Microorganisms
M3: Phase 3 fermentation process in making Local Microorganisms
M4: Phase 4 fermentation process in making Local Microorganisms

Based on Table 1, it shows that the total microbial groups (bacteria, fungi, and yeast) prove the existence of microorganisms sourced from organic waste and banana peels. The highest total number of bacterial colonies in Local Microorganisms stage 4 fermentation with a total bacterial presence of 1.8 X 10^5 *cfu / mL* and the lowest number of bacterial colonies in stage 1 Local Microorganisms fermentation with a total bacterial presence of 9.5 X 10^3 *cfu / mL*. In the fungi, the highest number of colonies in stage 4 Local Microorganisms fermentation with a total of 9.6 x 10^4*cfu / mL* and the lowest number in stage 1 Local Microorganisms fermentation was a total of 5.4 x 10^4 *cfu / mL*. Whereas the highest total yeast in stage 4 Local Microorganisms fermentation with a total of 1.9 x 10^5 *cfu / mL* and the lowest number of yeasts in Local Microorganisms fermentation stage 1 1,3 x 10^4 *cfu / mL*.

The results of the study showed that the total number of bacterial colonies of Local Microorganisms solution increased in fermentation results, the longer the increasing fermentation of bacterial growth can be seen in Table 1. Many factors affect the bacteria growing in fermentation, namely substrate, temperature, and pH. A substrate as a source of cabbage is a raw material for fermentation that contains nutrients needed by microorganisms to grow. The main source of making Local Microorganisms solutions is carbohydrates, glucose and the source of microorganisms themselves. The source of carbohydrates in this study is a banana skin, glucose from brown sugar and sources of microorganisms derived from organic waste. Fungi grow in acidic conditions with a pH range of 2-8.

In contrast to the total number of bacterial colonies that experienced a rapid increase in fermentation results, the total number of fungal colonies (Table 1) experienced a slow increase. The substrate can also affect fungal growth. Ramakrishnan et al., (2019) explains that Local Microorganisms conditions cause oxygen content to be limited, while fungi are aerobic (for survival requires oxygen). Most fungal hyphae have to make contact with air to get oxygen supply. In addition, fungal growth also influenced by factors such as organic matter content, pH, aeration, temperature, light, moisture and chemical compounds in the environment.

The total amount of yeast obtained (Table 1) has increased in the second and third ferments. This caused by giving a substrate suitable for yeast growth. Yeast found in fruit, grain, and sugar-containing substrates. Yeast likes pH 2.5-2.8 reproduction of yeast also influenced by environmental conditions and nutrients available in substrates such as carbohydrates, nitrogen, and oxygen. In all the processes of making Local Microorganisms, there is an increase in the number of microorganism groups. Graph of the increasing number of microorganisms/week for 4 weeks shown in Figure 1.

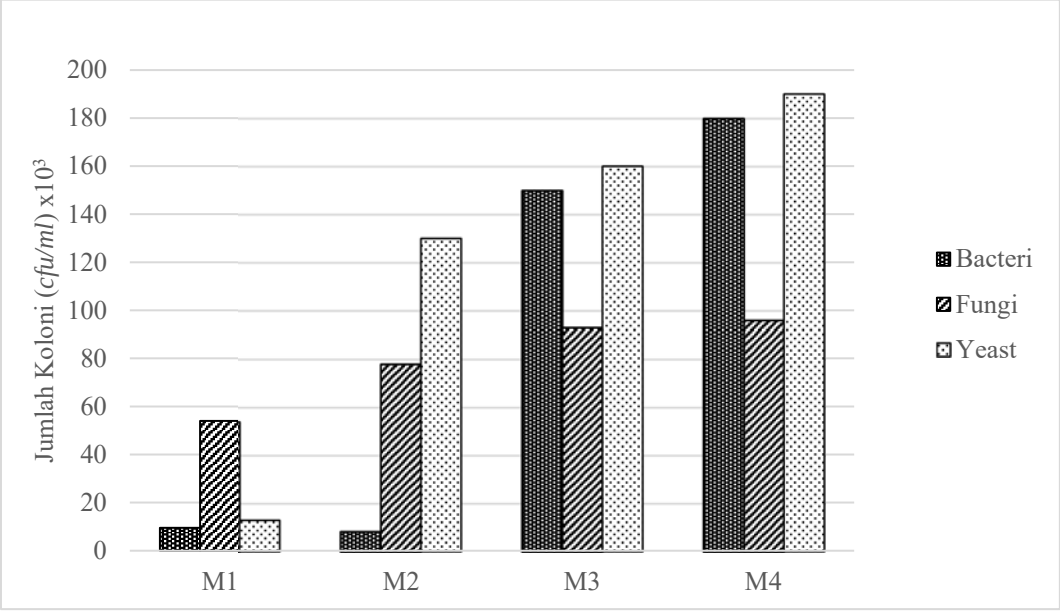

Figure 1. Total Colonies of Each Microbe at Several Stages of Making Local Microorganisms

Description: M1: Phase 1 fermentation process in making Local Microorganisms
M2: Phase 2 fermentation process in making Local Microorganisms
M3: Phase 3 fermentation process in making Local Microorganisms
M4: Phase 4 fermentation process in making Local Microorganisms

Figure 1. shows that each bacterial, fungi and, yeast microbial colony experienced a gradual increase in the number of Local Microorganisms fermentation stage 1 to stage 4 Local Microorganisms fermentation, the most significant increase occurring in the number of bacterial colonies from Local Microorganisms 1 to Local Microorganisms 2. This increase in the number of colonies could be due to sufficient microbial nutrient intake in Local Microorganisms so that microbes can actively reproduce during the fermentation process. Figure 1. shows that the microbial growth phase that occurs is the lag phase or adaptation and exponential phase. Phase lag or adaptation, namely microbes that grow will adjust to the surrounding environment. This adaptation phase influenced by several factors, namely the medium and the growth environment and the number of initial inoculums. The second phase is the Log Phase or Exponential Growth, in this phase, the microbes divide rapidly and constantly following the logarithmic curve. In this phase, growth is strongly influenced by the medium of growth such as pH and nutrient content, as well as environmental conditions including temperature and air humidity

Local microorganisms sourced from organic materials and added with banana peels that have buds that contain a lot of gibberellins and cytokines. Banana skin contains several microorganisms that play a good role in soil fertility. The types of microbes that have been identified in local banana peels include Bacillus sp., Aeromonas sp., Aspergillus niger, these microbes that will break down organic matter. Microbes in banana skin Local Microorganisms will act as decomposers of organic matter to be composted (Sutrisno et al, 2020)

Local microorganisms from organic waste materials can be used to make fertilizer the same as organic solid waste which contains a lot of nutrients (N, P, K) and other organic materials. The use of local microorganisms from organic waste materials can help improve soil structure and quality. Organic waste can not only be made for compost or solid fertilizer but can also be made as liquid fertilizer, the tool needed to make liquid fertilizer is a composter. Composter functions in flowing air (aeration), maintaining humidity, and temperature, so that bacteria and microorganisms can break down organic matter optimally. The difference in microbial growth in each medium caused by the metabolism of organic matter, nutrients, and the microbial environment. Microbial growth influenced by various environmental factors, including temperature, pH, the presence of oxygen, and food availability for these microbes (Kornievskaya et al, 2020).

Comparison of Existence of Microorganism Colonies in Local Microorganisms (M4) and EM4

In Local Microorganisms (M4) microbial groups are found, including bacteria, fungi, and yeast. The number of colonies of each microbial group showm in Table 2:

Table 2. Comparison of Number of Microbes in fermentation Local Microorganisms stage 4 and EM4

Type Of Sample	Number Of Colony (*cfu/ml*)			pH
	Bacteri	Fungi	Yeast	
M 4	1.8 X 10^5	9.6 x 10^4	1.9 x 10^5	4
EM 4	6.8×10^3	1.5×10^4	4.1×10^4	4

Description : M 4 : Phase 4 fermentation process in making Local Microorganisms

EM4 : Control

Table 2. explains that the comparison of the presence of bacteria in Local Microorganisms stage 4 (M4) and EM4 fermentation as control was in Local Microorganisms stage 4 (M4) 1.8 X 10^5 *cfu / ml* fermentation and 6.8×10^3 *cfu / ml* in EM4 as a control. The presence of more bacteria was found in Local Microorganisms stage 4 fermentation, compared to EM4, while the number of fungi in Local Microorganisms fermentation stage 4 (M4) 9.6 x 10^4 *cfu / ml,* and in EM4 the number of fungi was 1.5×10^4 *cfu / ml*, and the amount of yeast in fermentation Local Microorganisms stage 4 (M4) is 1.9 x 10^5 *cfu / ml* and in EM4 is 4.1×10^4 *cfu / ml*. So, from the above data it can be seen in general that the average microbial presence in stage 4 (M4) Local Microorganisms fermentation is more common than EM4. Based on factors that influence the rate of microbial growth, mainly the components of the culture medium, namely, pH of water activity, external factors such as the temperature of oxygen and pressure. In general, in nature there are microbial life cycles, either fungi or bacteria depending on the object to be overhauled. Sometimes the degradation begins with the microbes of the fungus after the degradation process occurs in a certain period of time, the fungi will die but are stored in the spores to be continued by bacteria or vice versa which is initiated by bacterial degradation and followed by fungi.

In Table 3, there were more total bacterial, fungal and yeast colonies found in M4 from EM4. The cause of the higher number of colonies in M4 compared to EM4 is because Local Microorganisms products that have just had an effect on the growth of many microbial colonies because they still have sufficient energy in growth. While microbes in EM4 are less likely to be caused by microorganisms in EM4 in the resting phase or dormancy phase. The phase of microbial dormancy that occurs in a microbial compilation does not die and cannot grow because they do not produce results. This dormancy phase will end the culture of microorganisms in EM4 that are applied, the compilation is applied, soils can quickly become active in overhauling organic matter in the soil. According to Stoffella and Kahn (2001) the longer the fermentation time, the lower the number of microbes followed by the reduced substrate for growth. The use of EM4 products that been purchased before use must be returned back non-organically as well as sugar energy sources so that microbes can be re-developed.

EM4 is a material that helps speed up the process of making organic fertilizers and improves its quality. Besides that, EM4 also used to improve soil structure and texture for the better and to supply nutrients needed by plants. The microorganisms contained therein are genetically original not engineered. Generally, EM4 can made by using materials that are easily available (Nurtjahyani et al., 2020). The composting process with the help of EM4 takes place semi-anaerobically. With this method, the odor produced can be lost if the process goes well. The number of microorganisms in EM4 is very much around 80 genera.

EM4 has properties that are unique because it can neutralize organic or soil material that is acidic or basic. These microorganisms are in a resting phase and when applied can quickly actively remodel organic matter. The results of the organic matter breakdown are in the form of organic compounds, antibiotics (alcohol and lactic acid), vitamins (A and C) and polisakarida. In addition to producing organic compounds, EM4 can also stimulate the development and growth of other beneficial microorganisms such as nitrogen-fixing bacteria, phosphate-soluble bacteria, microbes that are antagonistic to pathogens and can suppress the growth of pathogenic fungi (Kornievskaya et al, 2020).

pH and Temperature Product for Local Microorganisms

The pH value and temperature of Local Microorganisms products obtained with the basic ingredients of organic waste coupled with banana peels can be seen in table 3.

Table 3. pH and Temperature product for Local Microorganisms

M 1		M 2		M 3		M 4	
Temperature	pH	Temperature	pH	Temperature	pH	Temperature	pH
27^0C	3	33^0C	4	28^0C	4	28^0C	4

Based on Table 3. pH values in the first week to last week Local Microorganisms solution ranged from 3-4. The increase in pH occurs in the first week to the second week of the formation of the Local Microorganisms solution. Changes in pH indicate that incubation works well. pH will encourage the activity of bacterial microorganisms optimally. According to Hamed et al (2014), the acidic pH value is influenced by microbial activity in decomposing organic matter which produces CO_2 gas which will form carbonic acid (H_2CO_3) which cheaply breaks down into H + and HCO_3-ions. This H + ion will affect the acidity so that the pH of the Local Microorganisms solution decreases resulting in increased acidity. In addition, acidic conditions are good for the production of phytohormones (auxin, gibberellins, and cytokines) which are known to play a role in increasing vegetative growth, generative and fruit ripening (Widjajanto et al., 2017).

Based on Table 2. the temperature in the fermentation of Local Microorganisms from the first week to the second week experienced an increase, namely from 27^0C-33^0C. The second week until the third week has decreased. Whereas in the third week to the fourth week the fermentation temperature of Local Microorganisms is stable, 28^0C. At the time of making Local Microorganisms fermentation heat release occurs. According to Kornievskaya et al (2020) the increase in temperature related to the activity of microorganisms in decomposing organic matter that produces energy in the form of heat, CO_2, and water vapor. Then the fermentation temperature decreases because the activity in breaking down organic matter decreases.

Quality of Compost Produced Using Local Microorganisms stages 4 (M4) and EM4 (controls)

The quality of compost produced using Local Microorganisms (M4) and EM4 (control) based on organic waste coupled with banana peels shown in Table 4:

Table 4. Compost products produced Using Local Microorganisms (M4) and EM4 (control)

Types	pH	Temperature	Colour	Texture	Scent
Local Microorganisms (M4)	6	28^0C	Brown	Rough,Notcompletely decomposed	No Smell
EM4	5	30°C	Black	Rough,Notcompletely decomposed	No Smell

Based on Table 4. above, it can be seen that there are differences in the composting results between Local Microorganisms (M4) and EM4 including pH. In composting with M4 the pH obtained is 6. Whereas in EM4 compost has a pH of 5. This difference can occur due to the influence of the substrate and decomposers that work on each compost. The pH range of compost piles should be between 7-7.5 according to the pH needed by plants. Based on the table above composting added with a solution of local microorganisms from organic waste materials has a temperature during composting indicating the presence of heat released by microorganisms as a result of oxidation reactions. Based on these tables at the beginning of composting has a high temperature. The temperature increase is caused by the activity of microorganisms in the process of decomposition of organic matter (Colón et al., 2010).

The compost texture added with M4 and EM4 solution has a rough texture and some compost material has not been destroyed. The ripe compost fixed with a smooth texture. At the next observation, the size of the compost becomes smaller. This indicates degradation activity by Azetobacter bacteria in compost. The compost was destroyed more but still a little rough. Compost when held no longer sticky in the hand (crumbs). Compost does not produce water vapor when it mixed with plastic. The aroma of compost in the observation of the smell of compost M4 and EM4 is equally not foul smelling, at the beginning of composting. Scented compost like its raw material, which is a scent of leaf litter, the final stage of composting of both types of compost has no foul odor and is like a litter odor. The compost that cooked has a smell like soil (Ezeagu et al., 2017).

The color produced by the two types of compost is different. For compost, M4 has the color of the brown waste substrate while in EM4 compost has the color of the blackish waste substrate. Color changes occur because of the decomposition of microorganisms that convert organic matter with complex C chains into simple C forms. The decomposition process will cause composted material to lose color pigment so that the color changes to black according to the color of its constituent elements. In the composting processes there will be a decomposition of organic matter by microbial activity, the microbes will take water, oxygen, and nutrients from organic matter which will then decompose and release CO2 and O2 (Mohee and Mudhoo. 2005).

IV. CONCLUSION

The total number of microbes in Local Microorganisms found the presence of microbes such as bacteria, fungi, and yeast with the basic ingredients of organic waste and a banana skin, namely the total bacteria 1.8 x 10^5 *cfu / mL*, total fungi 9.6 x 10^4 *cfu / mL*, and total yeast 1.9 x 10^5*cfu / mL*. The quality of compost produced is different in terms of color and texture but has the same distinctive aroma of compost.

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