

#### Colour, Microbial and Sensory Effects of Food Paste with Jasmine Flower ins Fish Fillet

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#### DECLARATION

I hereby declare that the work embodied in this Report is the result of the original research and has not been submitted for a higher degree to any universities or institutions.

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#### TABLE OF CONTENT

		CONTENT	PAGE
DECLARATION			i
ACKNOWL <mark>EDG</mark>	EMEN1		ii
TABLE OF <mark>CON</mark>	TENT		iii
LIST OF TABLE	S		vi
LIST OF FIGUR	ES		viii
LIST OF ABBRE	VIATIC	ON AND SYMBOLS	ix
ABSTRACT			Х
ABSTRAK			xi
CHAPTER 1	INTI	RODUCTION	
	1.1	Research Background	1
	1.2	Problem Statement	2
	1.3	Hypothesis	2
	1.4	Objective	3
	1.5	Scope of study	3
	1.6	Significant of study	3
CHAPTER 2	LITI	RRATURE REVIEW	
	2.1	Food By-Product	4
	2.2	Persicaria Odorata	5
	2.3	Lemon Citrus Peel	7
	2.4	Jasmine Flower	8
	2.5	Olive Oil	10

2.6	Food Preservation	10
2.7	Marination of Food	11

CHAPTER 3	MAT	TERIALS AND METHODE	
	<mark>3.</mark> 1	Location	13
	3.2	Material	13
		3.2.1 Raw Materials	14
		3.2.2 Equipment	14
		3.2.3 Chemical	14
		3.2.4 Instruments	14
	3.3	Method	
		3.3.1 Preparation of the herbal	15
		paste and fish fillets	
		3.3.2 Fish fillet and herbal paste	16
		preparation	
		3.3.3 Total plate count	17
		3.3.4 Sensory Evaluation	18
		3.3.5 Data Analysis	18
	3.4	Flowchart of Research	19
CHAPTER 4	RES	ULTS AND DISCUSSION	

4.	1 Colou	r Analysis	20
4.	2 pH Ar	nalysis	22
4.	3 Total	Plate Count	31
4.	3 Sensor	ry Evaluation	37

FYP FIAT

#### CHAPTER 5 CONCLUSION 5.1 Conclusion 5.2 Recommendation

**REFERENCES** 

APPENDIX

46

43

43

44

# UNIVERSITI MALAYSIA KELANTAN

#### LIST OF TABLES

		Page
3.1	Formulation of the Food Paste (Gram)	16
3.2	Formulation of the Food Paste (Percentage)	17
4.1	Colour Analysis	20
4.2	pH Analysis	22
4.3	Test of Homogeneity of Variances	37
4.4	ANOVA of Sensory Evaluation	38
4.5	Turkey Test (Colour)	39
4.6	Turkey Test (Aroma)	39
4.7	Turkey Test (Taste)	40
4.8	Turkey Test (Appearance)	41
4.9	Turkey Test (Overall)	41

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## MALAYSIA

#### LIST OF FIGURES

		Page
2.1	Persicaria Odorata Leaves	6
2.2	Lemon Peel	8
2.3	Jasmine Flower	9
3.1	Flowchart of Research	19
4.1	Graph pH Fish	24
4.2	Graph pH Paste	25
4.3	Colour of Control	26
4.4	Colour of F1	27
4.5	Colour of F2	28
4.6	Colour of F3	29
4.7	Colour of F4	30
4.8	Control Sample of TPC	31
4.9	F1 Sample of TPC	32
4.10	F2 Sample of TPC	33
4.11	F3 Sample of TPC	34
4.12	F4 Sample of TPC	35

#### LIST OF ABBREVIATION AND SYMBOLS



FYP FIAT

UNIVERSITI MALAYSIA KELANTAN

#### ABSTRACT

#### Colour, Microbial and Sensory Effects of Food Paste with Jasmine Flower in Fish Fillet

Main objective of the study is to determine the shelf life of the fish pallet incorporated with the herbal paste. To study the effect of different concentration of herbal paste toward the shelf life of fish pallet. In research methodology, marination method is used to determine the how long the shelf life of meat product with and without the herbal paste. The numbers of samples used in the study are 5 samples and each of them have differences concentration. Meat with herbal paste of F2 expected to have longer shelf life compared to others samples. Herbal paste contains citrus lemon peel, jasmine flowers and also Persicaria Odorata. They contain polyphonic content as anti-oxidant and also antifungal compound. Significant testes such as sensory evaluation, total plate count, pH changes and also colour. Significance output from the research project is the meat products with herbal paste produce longer shelf life and give better aroma of meats.

Keywords: Herbal paste, marination, shelf life, sensory.

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#### ABSTRAK

#### Kesan Warna, Mikroba dan Sensor Paste Makanan dengan Bunga Jasmine di Fillet Ikan

Objektif utama kajian ini adalah untuk menentukan jangka hayat pallet ikan yang digabungkan dengan pes herba. Untuk mengkaji kesan kepekatan ubat herba yang berbeza ke arah hayat palet ikan. Dalam metodologi penyelidikan, kaedah perkalian digunakan untuk menentukan sejauh mana jangka hayat produk daging dengan dan tanpa pes herbal. Bilangan sampel yang digunakan dalam kajian adalah 5 sampel dan masing-masing mempunyai perbezaan kepekatan. Daging dengan ubat herba F2 dijangka mempunyai jangka hayat yang lebih lama berbanding sampel yang lain. Pasta herba mengandungi kulit limau, bunga melati dan juga Daun Kesum. Pes mengandungi kandungan polifonik sebagai anti-oksida dan juga sebatian antikulat. Ujian yang signifikan seperti penilaian deria, jumlah plat, perubahan pH dan juga warna. Output penting dari projek penyelidikan ialah produk daging dengan pes herba menghasilkan jangka hayat yang lebih lama dan memberi aroma daging yang lebih baik.

Kata Kunci: Pes Herba, Pemerapan, Jangka Hayat, Deria.



#### **CHAPTER 1**

#### **INTRODUCTION**

#### 1.1 RESEARCH BACKGROUND

This research is about the effect of food paste containing Jasmine Flower on fish fillet. Nowadays, synthetic preservation is widely used in modern technology because of the fast effect and also work effectively. There are several issues regarding the preservation from the consumer especially the side effect of the synthetic preservative. This study is to replace the synthetic preservatives into natural preservatives which can be the alternatives of the synthetic preservation in the food products. Preservation is widely used as the method to prolong the shelf life of a food and also to preserve the quality and avoid from contaminated by the microorganism. This method used to control the quality of a food by removing the factor of the bacteria growth from the food by using the temperature control and also antibacterial herbal properties. The study also to determine the effectiveness of the herbal paste to control the bacterial growth and also to extend the shelf life of the product. Fish used in the experiment is the Rastrelliger Kanagurta. The fish comes from family of Scombridae. The fish is suitable because it is cheap and also easy to collect near with the location of the experiments. The fillet will be tested with the paste and to know about the bacterial effect and also the taste of the products. Sensory Evaluation, pH, Total plate count and colour analysis conducted to know the effect and also respond of the herbal paste toward the meat.

#### **1.2 PROBLEM STATEMENT**

Food preservatives becomes one of the most important things in food and beverages industry nowadays because of its function to extend the shelf life of product. There are two types of food preservatives which are synthetic and also natural food preservatives (Foodadditivesworld.com, 2018). Synthetic nowadays becomes very popular and widely used without knowing it side effect to human health. There are few problem involving with synthetic preservatives such as allergic response and also can lead to cancer problem. This is because of chemical substance used that had side effect to human body. Fish fillet is used to test the effectiveness of the herbal paste and how the paste can react toward the growth of the bacteria. The fish fillet will be divided into a few samples and tested with different condition.

#### 1.3 HYPOTHESIS

- $H_0$  = The presence of herbal paste does not affect the shelf life of fish fillets
- $H_1$  = The presence of herbal paste does affect the shelf life of fish fillets
- $H_0$  = The temperature does not affect the affects the shelf life of fish fillets
- $H_1$  = The temperature affects the shelf life of fish fillets

#### **1.4 OBJECTIVES**

- 1. To determine the shelf life of the fish pallet incorporated with the herbal paste
- 2. To study the effect of  $4^0$  C temperature toward the shelf life of fish pallet

#### 1.5 SCOPE OF STUDY

The scopes of study are using fish fillet as the main dishes with three kind combination of herbal paste. The meat will go preservation process and one fillet used with herbal paste and another one used without herbal paste. Both treatment will be keep under room temperature and also in chiller with different temperature.

#### **1.6 SIGNIFICANT OF STUDY**

Herbal paste is used as the food preservatives and also antibacterial properties since the plant is believed to have the antibacterial properties. The study also to prolong the shelf life of the fish fillet products using the herbal paste combination of the jasmine flower, lemon citrus peel and also Persicaria Odorata. The study could offer an alternatives preservative for food industry and replaced synthetic preservatives to herbs plant which safer and also environmental friendly.

#### **CHAPTER 2**

#### LITERATURE REVIEW

#### 2.1 FOOD BY-PRODUCTS

Food by-products are produced in large amount of wastes and about 38% of food wastes comes from food processing (Helkar & Sahoo, 2016). Food by product usually contain high level of the phenolic compound, glucosinolates, cyanogenic, glycosides, oxylipins, and alkaloids among others (Maldonado et al, 2015). Antimicrobial from the by-product plant will provide suitable chemical that can fight the bacteria and make it suitable to be used as the food preservatives. Antimicrobial usually can be found at outer side of the plant such as mangoes teen plant, banana by product and also lemon citrus (Maldonado et al., 2015). One of the food by product used in the herbal paste is the lemon citrus peel. Food waste usually can be found in animal and also from plant. The disposal of the food waste to environment is not good for the economic, ecosystem and also can cause pollution. Food waste can be used for many purposes and provide extra value for manufacturer. Proper food by product management will create a positive impact to economy and also environmental pollution. Nowadays, consumer looking for safe food and avoid from chemical product which can give side effect of their health. Food by product is one of the arising demands since it is natural and also health food sources. Food waste can be found in edible food product and can be divided into several group such as

- Fruit and vegetable industry
- Grain processing industry
- Brewery and winery industry
- Marine industry
- Meat industry
- Dairy industry

This are few of categories of food waste of food by product commonly used in the industry to be processed. (Bharat & Sahoo, 2016)

#### 2.2 PERSICARIA ODORATA

P. Odorata is the herbs plant widely used in the medicine and also in cooking. It is belonging to the group of fresh culinary herbs. P. Odorata usually used as the culinary herbs in cooking. In Malaysia, P. Odorata was named as 'Daun Kesum'. In Persicaria Odorata leaves, it contains the phenolic of aldehyde and terpene which has antibacterial properties. In his study also stated that the extract of Persicaria Odorata has high potential natural antibacterial since it can inhibit the growth of the Staphylococcus aureus, Escherichia coli and Enterococcus faecalis effectively (Abu bakar et al, 2015). Persicaria odorata can be classified under family of polygonaceae and genus of persicaria. The plant also grows in tropical and subtropical zones. In stable condition, they also can grow 15 – 30 cm. The physical properties of the plant are dark green and the stem jointed of each leaf (Ridzuan et al, 2013). Persicaria Odorata claimed to have antioxidant, antibacterial, anti-fungal, anti-diarrheal, anti-inflammatory, ant cytotoxic, antiulcer, and antigen-toxicity activities (Uyub et al, 2010). Persicaria Odorata also known as Vietnamese coriander and commonly used for various purposes such as culinary herbs and for traditional medicine to fight inflammation, sores, ulcer and wounds. The plant also has phytochemical activities such as algaecide, antidiabetic and also antifungal, antioxidant and also can be used for anticancer activities. (Kumar, Dash, & Zakaria, 2016). In a study to determine the microbial activity of leaf Vietnamese with "gulai tempoyak", total plate count (TPC) used to know the growth rate of yeast and bacteria. The abundance of the biologically active compound in the plant such as antimicrobial, antifungal and also anti-inflammatory was detected. From the resulted showed that the Persicaria Odorata effectively can reduced the growth rate of bacteria. (Aris, 2015).



#### 2.3 LEMON CITRUS PEEL

Citrus is a very popular due to its multifunctional properties such as for medicinal, culinary herbs and others. It is also popular in term of aroma, taste and also health benefits. Citrus Limon is botanical name of lemon plant and perhaps originated from Asia. The colour of lemon may varies depend on the maturity level and also type of cultivar (Al-Juhaimi, 2014). Citrus peel by-products have many benefits and also function. These by-products can be used as pectin and flavonoids production (Ahmed et al., 2016). These by product can be used in animal feed and also for green energy production (Al-Juhaimi, 2014). A study was conducted to know the content of the lemon peel. Lemon peel contains rich sources of the phenolic compounds and dietary fibre; citrus fruit residues or waste also can act as potential nutraceutical resources. Citrus peel also rich in bioactive compounds and also can be recycle back to get the benefit and also food supplement that provide many function in food industry. Citrus peel also used for food natural antioxidant to prevent oxidation of some foods. The antioxidant in the peel is because of the ability of the phytochemical to donate an electron or hydrogen from phenolic hydroxyl group. The antioxidant is mainly because of the ability of phenolic compound to scavenge free radicals and break radical chain reaction and chelate metals. The function of the antioxidant is for food additives and improve their shelf life by preventing lipid per-oxidation and also protect oxidative damage (Ahmed et al., 2016). The citrus peel also functions for non-caloric bulking agent, enhance water and also oil retention. Apart from that it also can improve oil retention and prevent from any disease from oxidative issues (Rafiq et al, 2016). Citrus fruit is an important medicinal plant that belongs to the family Rutaceae and it has many functional properties such as for medicinal purposes, highly nutritious and contain only 0.9% of total daily calories

(Amutha Kavusik, 2017). Some of the important bioactive compound contain in the citrus fruits are ascorbic acid, flavonoids and also pectin which is very important for the nutrients. Some of the flavonoid present in the plant are Hesperidin, Narirutin, Naringin and Eriocitrin. In the peel, it contains and rich in polymethoxylated flavones (Amutha Kavusik, 2017). The citrus flavonoid has been found to have a health related property such as anticancer, antiviral and also anti-inflammatory activities and also, reduce capillary fragility and restricts human platelet aggregation. Apart from that, Citrus is one of the most popular world fruit crop and active in biological activities that can give health protection for human. It also can provide vitamin C, folic acid, potassium and also pectin. (Rafiq et al., 2018).Vitamin C is the major in citrus and rich in peel with efficient scavenge diversity of reactive oxygen (Ahmed et al., 2016). Dietary fibre also can be obtained in the peel of the lemon citrus. It helps in evading hydrolysis, digestion and absorption in the human small intestine (Rafiq et al., 2018).



Figure 2.2: Lemon Peel

#### 2.4 JASMINE FLOWER

Jasmine flower is widely used for cooking in culinary herbs. Jasminum sambac also known as Nyctanthes sambac is classified under the genus of Jasminum. This plant mainly in the southerwestern and southern Asia. It is small evergreen vine and reaching up to 0.5-3m tall (Ahmed et al, 2015). The Jasmine flower is widely used for the perfumes and also making tea. Apart from that, jasmine flower also used for decoration purposes and also useful for pharmaceutical industries since of the bioactive compound contain in the flower and also react as antifungal properties. Jasmine flower also used for the stimulating effect for the relief of depression and also stress. Apart from that, jasmine flower also can be used for antioxidant activity. This is because of the flower contain linalool that can be used for prevention of oxidation of food. Linalool also can be used with other component of essential oil to form an effective preservatives agent. Linalool also has been certified by Food Drug Association (FDA) that it is safe to be used and can be used also for flavouring agent of food products (Ahmed et al., 2015). This type of herbs gives very good fragrance and suitable used for its aromatic flavour. This natural flavour used in many industries such as food and also fragrance industry. Jasmine function as the calming and relaxing. It is very effective to release the stress and reduced the depression. Jasmine also used for the skin irritation and function as antioxidant properties for the human cells (AlKhazraji & Prof, 2015).



Figure 2.3: Jasmine Flowe

#### 2.5 OLIVE OIL

Olive oil is widely used in cooking purposes. It has high content of oleic acid and give many benefits. It has polyphenols, tocopherols and phytosterols as the antioxidant. Apart from that olive oil also play important role as anti-inflammatory and also can prevent from various types of cancer. Oleic acid can reduce the effect of oxidative stress in the human body and also can reduced the risk of the heart attacks. Oleic acid also can act as antiitumor agent and also can reduce the risk of prostate cancer. (Salazar-garc & Town, 2017). Olive oil has many benefits such as it is very rich in health monounsaturated fats called oleic acid which about 73% of the total oil content. Apart from that, olive oil also contains large amount of the antioxidants properties. Apart from it can fight inflammation it also can reduce the risk of heart disease. Olive oil also can help to prevent strokes that cause by the disturbance of blood flow to our brain by the blood clotting. Consuming of the olive oil not cause the increase of weight and also obesity ("11 Proven Benefits of Olive Oil", 2018).

#### 2.6 FOOD PRESERVATION

Food preservatives are chemical and biological agents used to ensure safety, improve the appearance or extend the shelf life of foods (Shao et al, 2011). There are many method nowadays used to preserve the food in conventional technologies and also traditional ways. One of the way to preserve the food product by using the thermal processing. Thermal processing is the most common technique used to preserve the food. Food is heated to kill the bacteria at certain temperature. (Augusto et all, 2018). Another method is by cooling techniques. Cooling or chilling is the process to extend the shelf life of the food product by inactivate the bacterial activity. Cooling also can preserve the nutritional changes and slowing the process. Cooling not just reduce the microbial growth but also prevent the development of certain microorganism. Another method that can be used is freezing techniques. Freezing is a method where food product is exposed to very cool temperature until 0<sup>o</sup> C. By solidifying the water, this will kill or slowing down the activity of the bacterial growth.(Augusto et al., 2018). Apart from that, water activity also plays very important function for the food preservation. By reducing the water activity, it will also slow down the bacterial growth and also can extend the shelf life of the food. Dry product will have longer shelf life compare to wet product. The temperature used in this research is 4<sup>o</sup> C for a week of storage. In this study preservation method is used to extend the shelf life of the fish fillet by using the herbal paste.

#### 2.7 MARINATION OF FOOD

Herbal paste with antibacterial are selected to test the quality and also the shelf life of fish fillet. There are many methods used in the preservation of food. Instead of using the synthetic preservation, the green method by using the herbal paste is used to replace the synthetic method and to determine the effectiveness of the herbal paste. Food product easily contaminated if they are exposed to the outside air because of the pathogen. Herb and spices have many function in cooking also for food preservative and also additives in term of aromatic or flavours. Marination is another technique used to tenderize and improve the cooked meat juiciness. The word marinade comes from the Latin "marinara" which means "of the sea". Marinade consist of cooking oil, acid and also spices. The main purposes are to tenderize and flavour. Some of the material used for margination process are olive oil, salt, herbal paste and also onion with garlic. Acid work to soften and flavour the meat by denaturing the meat. Oils used to moisten the meat and add flavour (Christensen et al, 2009).



#### **CHAPTER 3**

#### **METHODOLOGY**

#### 3.1 LOCATION

Herbal paste of Citrus Peel, Jasmine Flower, Persicaria Odorata and also marination herbs such as onion, garlic, olive oil and also salts. These herbal sources from local market in Jeli Kelantan. The study was conducted at food laboratory, Faculty of Agro Based Industry, University Malaysia Kelantan Jeli

### 3.2.1 RAW MATERIALS

List of raw materials used in food preservation are Indian Mackerel fish, Jasmine Flowers, Citrus Lemon peel, Persicaria Odorata, Onion, Garlic, Olive Oil, Salt and distilled water.



#### **3.2.2 EQUIPMENT**

List of equipment will be used in this experiment such measuring cylinder, knife, electronic weighing balance, petri dish, Nutrient, spoon, micropipette (1000µl), test tube rack, test tube, dropper, media bottle, filter funnel, beaker, measuring cylinder, aluminium foil and media bottle.

#### 3.2.3 CHEMICAL

Ethanol, Nutrient Agar, and distilled water, Nutrient Agar Powder.

#### 3.2.4 INSTRUMENTS

CR-400 Chromameter (Konika Minolta Sensing Ameicas, USA), SES6202 Saffron Precision weighing balance (Saffron Electronic Scales, Varachha Road, Surat, Gujrat, India),

#### 3.3.1 PREPARATION OF THE HERBAL PASTE AND FISH FILLETS

The raw materials of herbal paste used for marination basically from the local market near with UMK Jeli at Pantai Timur. The plants were Jasmine Flowers, Citrus Lemon peel, Persicaria Odorata, Onion, Garlic and Olive Oil used for marination process. All the plants washed and cleaned to ensure there is no dirt on the plant. Next, the herbal plant rinsed for 30 minutes and dried. After drying process then the plant was blended for 5 minutes. The paste mixed together with different ratio according to different sample in the table 3.1. There are two different criteria conducted that were herbal paste and herbal paste with fish fillet. Preparation of fish fillet firstly fresh fish was cleaned and sliced. The bones were fully removed and the flesh was cut into 12 cubes. 2g of herbal paste applied at the fish cube. The fish cube with herbal paste and herbal paste were packed using plastics sachet stored at temperature of 4 °C for 1 week. The assays carried out using total plate count, pH, colour and also sensory evaluation.



#### 3.3.2 FISH FILLET AND HERBAL PASTE PREPARATION

Five fish will be used to get 60 fish fillet cube to use with the preservation process. The preservation process use temperatures of 4 °C with different concentration of the herbal paste and also the fish fillet. Herbal Paste and Fish Fillet uses same formula to study the reaction of the paste and the paste toward the fish.

INGREDIENT	CONTROL	F1	F2	F3	F4
Jasmine Flower (g)	0	0	0	60	20
Persicaria Odorata (g)	0	0	60	0	20
Lemon Peel (g)	0	60	0	0	20
Onion (g)	25	25	25	25	25
Garlic (g)	5	5	5	5	5
Olive Oil (g)	5	5	5	5	5
Salt (g)	5	5	5	5	5
Water (g)	40	40	40	40	40
Total (g)	80	140	140	140	140

Table 3.1 Formulation of the Food Paste (Gram)



INGREDIENT	CONTROL (%)	F1(%)	F2(%)	F3(%)	F4(%)
Jasmine Flower (g)	0	0	0	42.9	14.3
Persicar <mark>ia</mark> Odorata (g)	0	0	42.9	0	14.3
Lemon Peel (g)	0	42.9	0	0	14.3
Onion (g)	31.25	17.9	17.9	17.9	17.9
Garlic (g)	6.25	3.6	3.6	3.6	3.6
Olive Oil (g)	6.25	3.6	3.6	3.6	3.6
Salt (g)	6.25	3.6	3.6	3.6	3.6
Water (g)	50	28.4	28.4	28.4	28.4
Total (g)	100	100	100	100	100

 Table 3.2 Formulation of the Food Paste (Percentage)

#### **3.3.3 TOTAL PLATE COUNT TEST**

Total plate count was the assays to count the bacteria growth from the food samples. Firstly, the agar plate is prepared using the nutrient agar powder and also petri dish. The technique used is spreading on agar plates to estimate the total bacteria grow on the plate for each concentration with serial dilution. Spreader is sterilizing by the ethanol and heat with Bunsen burner flame. Then let it cool for a while before spread the samples into the plate. The plate incubated at 27 °C for 12 hours to promote the growth of the colonies (Eyler, 2013).

#### 3.3.4 SENSORY EVALUATION

The sensory test was conducted to determine the consumer likeness towards herbal paste with fish fillet formulation. Fish fillet incorporated with herbal paste was evaluated in terms of colour, aroma, taste, appearance and overall acceptability by thirty random (n=30) students from Universiti Malaysia Kelantan Jeli Campus. The samples were evaluated based on 7-point hedonic scale (1= extreamly dislike and 7= extreamly like). Sensory consumers received a cup of water and a sensory evaluation form. Consumers were given one sample at a time. The fish with herbal paste fried for 5 minutes and the samples cut into 5 cm cube and packed into a plastic.

#### 3.3.5 DATA ANALYSIS

All the data will be subjected to one-way analysis of variance (ANOVA) subjected by Duncan multiple range test with significance (P<0.05). The statistical analyses will be carried out using the SPSS Software Program for Windows, Version 24.0.

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#### 3.4 FLOWCHART OF RESEARCH ACTIVITIES



Raw material washed to remove all the dirt from the peel

The herbal plant will be rinsed for 30 minutes and let it dried before blend

Then, the paste will be mixed together with different ratio according to different sample in the Table 3.1

The fish fillet and herbal paste stored at temperature of 4 °C. The fish fillet and herbal paste separated according to different concentration of herbal paste and without herbal paste as control sample.

The fish fillet will be kept in the storage for about 1 week and the antimicrobial activity will be recorded based on the diameter of the microorganism growth.

Data will be recorded and analysed

Figure 3.1: Flowchart of research activities



#### **CHAPTER 4**

#### **RESULTS AND DISCUSSION**

#### 4.1 COLOUR ANALYSIS

			Colour analysis	
Paste	Day	<i>l</i> *	<i>a</i> *	<i>b</i> *
		Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD
Control	1	56.35±1.72	3.15±0. <mark>46</mark>	15.29±2.41
	3	55.46±3.24	3.16±0.48	13.80±0.51
	5	53.01±9.36	2.06±0.85	15.37±6.61
	7	49.83±5.17	$1.47 \pm 0.28$	13.50±2.74
F1	м / <sup>1</sup> м	57.31±4.84	9.19±1.11	19.55±1.80
	3	57.28±2.73	8.95±1.18	18.71±0.74
	_			
	5	59.64±3.97	7.89±1.52	17.06±2.34

	7	57.26±2.66	6.33±1.04	18.84±1.00
F2	1	26.86±1.48	-4.42±0.48	17.27±1.62
	3	33.76±2.84	-5.87±0 <mark>.69</mark>	21.29±1.39
	5	35.24±2.08	-2.67±0 <mark>.36</mark>	16.55±0.80
	7	35.31±1.45	-2.08±0.13	15.78±2.10
F3	1	35.33±1.45	-2.08±0.13	15.78±2.10
	3	39.95±2.32	2.25±0.31	11.24±1.95
	5	57.92±1.52	3.09±0.38	16.42±0.66
	7	56.81±1.47	2.94±0.78	18.29±2.72
F4	1	36.22±5.45	-5.52±1 <mark>.22</mark>	22.12±3.08
	3	36.94±3.37	-5.75±0. <mark>42</mark>	21.76±1.67
	5	38.48±2.81	-2.85±0.35	19.77±2.66
	7	37.89±0.95	-2.38±0.24	19.95±1.05

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#### 4.2 pH ANALYSIS

		рН		
Paste	Day	fish	paste	
		Mean ± SD	Mean ± SD	
Control	1	$2.28\pm0.88$	$2.283 \pm 0.88$	
	3	2.93±0.72	6.63±0.17	
	5	7.30±0.10	2.27±0.015	
	7	5.67±0.03	2.47±0.33	
F1	1	$2.73 \pm 0.75$	2.73±0.75	
	3	7.2±0.15	5.40±0.10	
	5	6.46±0.30	3.30±0.15	
	7	4.80±0.06	2.52±0.54	
F2	NIV	5.27±0.06	5.27±0.06	
	3	7.16±0.20	6.50±0.00	
	5	7.20±0.10	5.33±0.27	
	7	5.30±0.17	5.17±0.07	
F3		5.30±0.17	5.17±0.07	
	3	7.36±0.35	6.13±0.05	

	5	7.10±0.10	2.36±0.02
	7	4.91±0.23	2.58±0.66
F4	1	2.40±1.46	2.4 <mark>0±1.46</mark>
	3	6.86±0.15	5.36±0.05
	5	6.60±0.17	3.58±0.32
	7	4.69±0.24	3.75±0.25





#### Figure 4.1: pH Fish

The graph above showed the results of the pH against the time interval. pH is one of the indicator to determine the growth of bacteria. The internal pH of bacteria ranges from 6.5 to 7.0 in acidophilus, 7.5 to 8.0 in neutrophils and 8.4 to 9.0 in halophiles.(Mayo & Noike, 1996). The graph showed that almost all the samples were increased their pH values. The last days showed some decreased in the value of pH. The experiment conducted with different concentration of herbal paste and marinated for 1 week to study the effect of temperature and also pH of the paste to the bacterial growth. Within 1 week, the changes of the pH were recorded to study the effect of pH against the growth rate of bacteria in 4° C of temperature. Most Bacteria are neutrophils and optimally grow at pH within 7.5 to 8.0. Most common bacteria such as Escherichia coli, staphylococci, and Salmonella spp. are neutrophils and do not function well in the acidic condition. ("The Effects of pH on Microbial Growth | Microbiology", 2018). On the first day, F2(5.27) and F3(4.27) have almost same values. F2(5.27) and F3(4.27) has higher value of pH compare to Control, F1(2.28) and also F4(2.40). All of the samples tested were in acidic condition.



Figure 4.2: pH Paste

From graph above, it showed the pH readings of the herbal paste against the time for 1 week. Herbal pastes had reading between range of 7.00 to 2.00 pH value. Mostly the pH reading for herbal paste was in acidic condition. On the 3<sup>rd</sup> day, the graph showed the reading start to increased. On the 1<sup>st</sup> day, pH reading of F2 and F3 has higher pH value compared to Control, F1 and F4. This indicates that Control, F2 and F3 is more neutral condition compared to others samples which are more acidic. Acidity will affect the growth of the bacteria in Total Colony Unit.



Figure 4.3: Colour of Control

Colour of the control sample not too much changes and the graph just slightly change during the first day until last day. 1\* indicates the lightness of the samples, a\* indicates redness and b\* indicates the yellowness of the samples. ("Let's look at some color spaces. ...I - Part I - Precise Colour Communication | KONICA MINOLTA", 2018). The 1\* value start with 56.35 for first day and drop to 55.47 on 3<sup>rd</sup> day. On 5<sup>th</sup> day the value keep decreased to 53.01 and lastly on 7<sup>th</sup> day 1\* value decreased to 49.82. The redness a\* of the control reach peak level at 3.16 on third day and drop until 1.42 at the end of the week. The yellowness of the b\* has the highest point at 15.37 and the lowest point is 13.51.





Figure 4.4: Colour of F1

The Colour samples of F1 show the same pattern with control sample and had constant value and slightly drop as the time pass. The samples of F1 has highest value of lightness 1\* and has lower value of redness a\*. F1 has the mixture concentration of the 60g of lemon peel, 25g onion, 5g each of garlic, olive oil and salt. Lastly distilled water of 40g. F1 has the highest concentration of the lemon peel compared to others herbal plants.





Figure 4.5: Colour of F2

The colour of the samples F2 was slightly different each other. Lightness 1\*, redness a\* and b\* value showed constant value and only slight changes along the period of assay. The lowest value of colour samples is redness and the highest colour value is the lightness 1\*. F2 has the highest concentration of the Persicaria Odorata in the sample. The herbs showed the dark green colour with the respective value of 1\*, a\*, b\* on the chart above. Lightness of the colour sample F2 was very high of colour value and lowest for redness and medium for yellowness.





Figure 4.6: Colour of F3

The samples of F3 has showed increased in the lightness value within the period of assay. The lightness starts from 37.42 and increased to 57.92 on the 5<sup>th</sup> day and drop again at 56.81 on 7<sup>th</sup> day. F3 has the highest concentration of the Jasmin herbal compared to other herbs. Jasmin flower showed changes of the colour along the time of experiments. The changes not too significant from the initial and the final value of colour analysis.





Figure 4.7: Colour of F4

Colour of samples F4 also same with the others samples. There were no major changes in the lightness 1\*, redness a\* and also yellowness b\*. For redness a\*, the value is -5.52 on 1<sup>st</sup> day and increased to -2.38 on the last day. Lightness of the samples has the highest value compared to redness a\* and also yellowness b\*. on 1<sup>st</sup> day, the a\* values was 36.22 and increased to 38.48 on 5<sup>th</sup> day and slightly drop to 37.89 on 7<sup>th</sup> day. For F4, herbal paste of jasmine flower, Persicaria Odorata and lemon peel has the same concentration with 20g each samples before mixed together.



#### 4.3 Total Plate Count

Sample dilution	Control
10 <sup>-3</sup>	Carthol - 1 Press
Colony Forming Unit(CFU)	0 cfu ml <sup>-1</sup>
10-2	
Colony Forming Unit(CFU)	0 cfu ml <sup>-1</sup>
10 <sup>-1</sup>	
Colony Forming	1 cfu ml <sup>-1</sup>

#### Table 4.8 Control Sample

Sample dilution F1 10<sup>-3</sup> Colony Forming Unit(CFU) 0 cfu ml<sup>-1</sup> Findau 10**-2** Colony Forming 0 cfu ml<sup>-1</sup> Unit(CFU) I! 10-1 Colony Forming 0 cfu ml<sup>-1</sup> Unit(CFU)

Table 4.9 F1 Sample



Sample dilution F2 10<sup>-3</sup> Colony Forming Unit(CFU) 0 cfu ml<sup>-1</sup> 10-2 Colony Forming Unit(CFU) 0 cfu ml<sup>-1</sup> 10-1 Colony Forming Unit(CFU) 0 cfu ml<sup>-1</sup>

Table 4.10 F2 Sample





Table 4.11 F3 Sample

Sample dilution F4 10<sup>-3</sup> Colony Forming Unit(CFU) 0 cfu ml<sup>-1</sup> Findas 10-2 Colony Forming Unit(CFU)  $1 \ge 10^3 \text{ cfu ml}^{-1}$ 10**-1** Colony Forming Unit(CFU)  $1 \ge 10^2 \text{ cfu ml}^{-1}$ 

Table 4.12 F4 Sample

Total Plate Count (TPC) was used as an indicator to know the growth of the bacterial population on the tested samples. It is also called standard plate count or mesophilic count. ("Aerobic Plate Count - Murray Brown Labs", 2018). In this experiment, TPC was used to know the Colony forming unit of the bacteria from the samples. The techniques used serial dilution method and the samples diluted until 10<sup>-3</sup> of the concentration. First the samples were prepared and take out from the storage. Next, distilled water was used to prepare the stock solution of each samples about 10 ml in the media bottle mixed together with the samples. Next the stock solution pipetted into the test tube for 1 ml each into 3 different test tube according to 10<sup>-3</sup> of serial dilution respectively and each test tube was labelled. Agar plate was prepared using the nutrient agar powder with distilled water and autoclaved. Then, the samples were spread into the petri dish with agar plate. The most effective sample from the experiment was F2 which has the lowest growth of the bacteria on the plate. From the result above, there was no colony forming unit on F2 plate which will be the most efficient combination of the herbal paste. F2 had showed the longest shelf life compared to others concentration and of samples of F1, F3, F4 and also control. The Herbal plant used in F2 were Persicaria Odorata, Onion and also marinated materials such as garlic, olive oil, salt and also water. Odorata leaves, it contains the phenolic of aldehyde and terpene which has antibacterial properties. In his study also stated that the extract of Persicaria Odorata has high potential natural antibacterial since it can inhibit the growth of the Staphylococcus aureus, Escherichia coli and Enterococcus faecalis effectively (Abu bakar et al, 2015). This indicates that the Persicaria Odorata has the most effective of antibacterial inhibition. This is proven that these herbs can fight the bacterial from growth.

#### 4.3 SENSORY EVALUATION

TEST OF HOMOGENEITY OF VARIANCES							
	Levene Statistic	df1	df2	Sig.			
colour	.640	4	145	.635			
aroma	3.599	4	145	.008			
taste	.821	4	145	.514			
appearance	3.623	4	145	.008			
overall	.529	4	145	.715			

#### Table 4.3: Test of Homogeneity of Variances



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Attribu tes	Groups	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	10.507	4	2.627	2.56 4	.041
colour	Within Groups	148.567	145	1.025		
	Total	159.073	149			
	Between Groups	9.907	4	2.477	3.59 2	.008
aroma	Within Groups	99.967	145	.689		
	Total	109.873	149			
	Between Groups	5.227	4	1.307	2.45 8	.048
taste	Within Groups	77.067	145	.531		
	Total	82.293	149			
	Between Groups	6.827	4	1.707	4.50 2	.002
appeara nce	Within Groups	54.967	145	.379		
	Total	61.793	149			
	Between Groups	7.907	4	1.977	4.31 2	.003
overall	Within Groups	66.467	145	.458		
	Total	74.373	149			

ANOVA

colour						
Tukey HSD						
		Subset for alpha = 0.05				
formulation	Ν	1				
F3	30	3.70				
F1	30	4.00				
Control	30	4.07				
F2	30	4.40				
F4	30	4.40				
Sig.		.062				
Means for groups in ho displayed.	mogeneou	is subsets are				
a. Uses Harmonic Mean S	ample Siz	e = 30.000.				

#### Table 4.5: Turkey Test (Colour)

### Table 4.6: Turkey Test (Aroma)

	A	roma	
Tukey HSD			
ΓΛΙ	Λ	Subset for a	lpha = 0.05
formulation	Ν	1 4	2
F2	30	3.90	
F3	30	4.00	4.00
F1	30	4.30	4.30
F4	30		4.50

Control	30		4.53				
Sig.		.341	.099				
Means for groups in homogeneous subsets are displayed.							
a. Uses Harmor	nic Mean	Sample Siz	e = 30.000.				

#### Table 4.7: Turkey Test (Taste)

Taste						
Tukey HSD						
		Subset for				
		alpha =				
		0.05				
formulation	Ν	1				
F1	30	3.80				
F2	30	3.80				
F3	30	3.80				
Control	30	3.80				
F4	30	4.27				
Sig.	171	.101				
Means for groups in ho	mogeneou	is subsets are				
displayed.						
a. Uses Harmonic Mean S	ample Siz	e = 30.000.				



appearance							
Tukey HSD		/					
	Subset fo <mark>r alpha = 0.05</mark>						
formulation	Ν		1	2			
F3	30		4.00				
F1	30		4.40	4.40			
F2	30		4.40	4.40			
Control	30			4.57			
F4	30			4.60			
Sig.			.093	.717			
Means for groups in homogeneous subsets are displayed.							
a. Uses Harmonic	Mean	Saı	mple Size = <mark>30</mark>	.000.			

#### Table 4.8: Turkey Test (Appearance)

#### Table 4.9: Turkey Test (Overall)

overall						
Tukey HSD						
лат		Subset for al 0.05	pha =			
formulation	Ν	Y I	2			
F3	30	4.30				
F1	30	4.50				
Control	30	4.53	4.53			
F2	30	4.60	4.60			
F4	30		5.00			

Sig.		.427	.064
Means for grou displayed.	ps in ho	omogeneous subs	ets are
a. Uses Harmoni	c Mean S	ample Size = 3 <mark>0.0</mark>	)00.

From the Table 4.3.3 (ANOVA of Sensory Evaluation), the significant different of the colour attribute was 0.041 which is lower than 0.05 so the null hypothesis was rejected. From the aroma attribute the significant different also lower than 0.05 which was 0.008 and for taste attribute, the significant different also lower which was 0.048. for appearance, the significant different was very low that was 0.002 and for overall attribute the significant different was 0.003 which is lower than 0.005 of significant different. So all the null hypothesis was rejected and the data was valid to be used. From the sensory evaluation also the most preferred samples was F4 since it has highest mean value of the mark given by the panellist compared to others samples.

## UNIVERSITI MALAYSIA KELANTAN

#### **CHAPTER 5**

#### 5.1 CONCLUSION

The aim of this study is to determine the effectiveness of the herbal paste to extend the shelf life of the meat and food product. The result showed that each concentration plays important role in food presercation. The most efficient of herbal paste was F2 since it can lower the growth of the bacteria and also slow down the growth rate based on the Total plate count analysis. Apart from that, the most effective herbal plant is the Persicaria Odorata which have the antibacterial properties and the herbs also widely used in the culinary herbs for cooking and also for medicinal purposes. Different treatment was conducted such as Total Plate Count, sensory evaluation, pH, and also colour analysis. From sensory evaluation in term of taste, aroma and also overall likeness, F4 is preferred compared to others samples.

#### 5.2 RECOMMENDATION

Further research need to be done to make sure that the result was better and more accurate. In the experiment there were a few errors detected such as uncallibrated pH meter that caused the graph of the pH not too good.

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#### Table 4.3.1: Descriptive Sensory Evaluation

		N	Mean ±SD	Std. Error	95% Confi Interval for Lower Bound	dence r Mean Upper Bound	Mini mum	Maxi mum	Between- Compone nt Variance
	F1	30	4.00± 1.114	.203	3.58	4.42	2	6	
colour	F2	30	4.40± 1.003	.183	4.03	4.77	2	6	
	F3	30	3.70± 0.794	.145	3.40	4.00	2	5	
	F4	30	4.40± 0.932	.170	4.05	4.75	2	6	
	Control	30	4.07± 1.172	.214	3.63	4.50	2	6	
	Total	150	4.11± 1.033	.084	3.95	4.28	2	6	

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	Model	Fixed Effects		1.012	.083	3.95	4.28			
		Random Effects			.132	3.75	4.48			.053
	F1		30	4.30± 0.877	.160	3.97	4.63	3	6	
	F2		30	3.90± 0.607	.111	3.67	4.13	3	5	
	F3		30	4.00± 0.788	.144	3.71	4.29	3	6	
aroma	F4		30	4.50± 0.861	.157	4.18	4.82	3	6	
	Control		30	4.53± 0.973	.178	4.17	4.90	3	6	
	Total		150	4.25± 0.859	.070	4.11	4.39	3	6	
	Model	Fixed Effects	ľ	0.830	.068	4.11	4.38			

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		Random Effects			.128	3.89	4.60			.060
	F1		30	3.80± 0.847	.155	3.48	4.12	2	5	
	F2		30	3.80± 0.761	.139	3.52	4.08	2	5	
	F3		30	3.80± 0.610	.111	3.57	4.03	3	5	
taste	F4		30	4.27± 0.640	.117	4.03	4.51	3	5	
	Control		30	3.80± 0.761	.139	3.52	4.08	2	5	
	Total		150	3.89± 0.743	.061	3.77	4.01	2	5	
	Model	Fixed Effects		0.729	.060	3.78	4.01			
		Random Effects	ľ	/A]	.093	3.63	4.15			.026

	F1		30	4.40± 0.675	.123	4.15	4.65	3	6	
	F2		30	4.40± 0.498	.091	4.21	4.59	4	5	
	F3		30	4.00± 0.587	.107	3.78	4.22	3	5	
appear	F4		30	4.60± 0.621	.113	4.37	4.83	4	6	
ance	Control		30	4.57± 0.679	.124	4.31	4.82	3	6	
	Total		150	4.39± 0.644	.053	4.29	4.50	3	6	
	Model	Fixed Effects		0.616	.050	4.29	4.49			
		Random Effects	l	JNI	.107	4.10	4.69			.044
overall	F1		30	4.50± 0.630	.115	4.26	4.74	3	6	

F2		30	4.60± 0.724	.132	4.33	4.87	3	6	
F3		30	4.30± 0.596	.109	4.08	4.52	3	5	
F4		30	5.00± 0.743	.136	4.72	5.28	4	6	
Control		30	4.53± 0.681	.124	4.28	4.79	3	6	
Total		150	4.59± 0.707	.058	4.47	4.70	3	6	
Model	Fixed Effects		0.677	.055	4.48	4.70			
	Random Effects		IBIII	.115	4.27	4.91			.051

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#### Table 4.3.4: Post Hoc Tests

		М	ultiple Compar	isons			
			Tukey HSD				
Deper	ndent Variabl	e	Mean Difference		Sig.	95% Confidence Interval	
-			(I-J)	Error		Lower Bound	Upper Bound
	F1	F2	400	.261	.544	-1.12	.32
		F3	.300	.261	.781	42	1.02
		F4	400	.261	.544	-1.12	.32
colour		Control	067	.261	.999	79	.66
		F1	.400	.261	.544	32	1.12
	F2	F3	.700	.261	.062	02	1.42
		F4	0.000	.261	1.000	72	.72

		Control	.333	.261	.707	39	1.06
		F1	300	.261	.781	-1.02	.42
	F3	F2	700	.261	.062	-1.42	.02
		F4	700	.261	.062	-1.42	.02
		Control	367	.261	.627	-1.09	.36
		F1	.400	.261	.544	32	1.12
	F4	F2	0.000	.261	1.000	72	.72
		F3	.700	.261	.062	02	1.42
		Control	.333	.261	.707	39	1.06
		F1	.067	.261	.999	66	.79
	Control	F2	333	.261	.707	-1.06	.39
		F3	.367	.261	.627	36	1.09
		F4	333	.261	.707	-1.06	.39
aroma	F1	F2	.400	.214	.341	19	.99
		F3	.300	.214	.629	29	.89

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	F4	200	.214	.884	79	.39
	Control	233	.214	.812	83	.36
	F1	400	.214	.341	99	.19
F2	F3	100	.214	.990	69	.49
	F4	600*	.214	.045	-1.19	01
	Control	633*	.214	.030	-1.23	04
	F1	300	.214	.629	89	.29
F3	F2	.100	.214	.990	49	.69
	F4	500	.214	.141	-1.09	.09
	Control	533	.214	.099	-1.13	.06
	F1	.200	.214	.884	39	.79
F4	F2	.600*	.214	.045	.01	1.19
	F3	.500	.214	.141	09	1.09
	Control	033	.214	1.000	63	.56
Control	F1	.233	.214	.812	36	.83

		F2	.633*	.214	.030	.04	1.23
		F3	.533	.214	.099	06	1.13
		F4	.033	.214	1.000	56	.63
		F2	0.000	.188	1.000	52	.52
	F1	F3	0.000	.188	1.000	52	.52
		F4	467	.188	.101	99	.05
		Control	0.000	.188	1.000	52	.52
		F1	0.000	.188	1.000	52	.52
taste	F2	F3	0.000	.18 <mark>8</mark>	1.000	52	.52
		F4	467	.188	.101	99	.05
		Control	0.000	.188	1.000	52	.52
		F1	0.000	.188	1.000	52	.52
	F3	F2	0.000	.188	1.000	52	.52
		F4	467	.188	.101	99	.05
		Control	0.000	.188	1.000	52	.52

		F1	.467	.188	.101	05	.99
	F4	F2	.467	.188	.101	05	.99
		F3	.467	.188	.101	05	.99
		Control	.467	.188	.101	05	.99
		F1	0.000	.188	1.000	52	.52
	Control	F2	0.000	.188	1.000	52	.52
		F3	0.000	.188	1.000	52	.52
		F4	467	.188	.101	99	.05
		F2	0.000	.159	1.000	44	.44
	F1	F3	.400	.159	.093	04	.84
		F4	200	.159	.717	64	.24
appearance		Control	167	.159	.832	61	.27
		F1	0.000	.159	1.000	44	.44
	F2	F3	.400	.159	.093	04	.84
		F4	200	.159	.717	64	.24

	Control	167	.159	.832	61	.27
	F1	400	.159	.093	84	.04
F3	F2	400	.159	.093	84	.04
15	F4	600*	.159	.002	-1.04	16
	Control	567*	.159	.004	-1.01	13
	F1	.200	.159	.717	24	.64
E4	F2	.200	.159	.717	24	.64
Г4	F3	.600*	.159	.002	.16	1.04
	Control	.033	.159	1.000	41	.47
	F1	.167	.159	.832	27	.61
Control	F2	.167	.159	.832	27	.61
Control	F3	.567*	.159	.004	.13	1.01
	F4	033	.159	1.000	47	.41
	F2	100	.175	.979	58	.38
FI	F3	.200	.175	.783	28	.68
	F3 F4 Control F1	$\begin{tabular}{ c c c c } \hline Control & F1 & F2 & F3 & F4 & F4 & F4 & F1 & F2 & F1 & F2 & F1 & F1 & F2 & F1 & F1$	$\begin{tabular}{ c c c c c } \hline Control &167 \\ \hline F1 &400 \\ \hline F2 &400 \\ \hline F2 &400 \\ \hline F4 & F2 &600^* \\ \hline Control &567^* \\ \hline F4 & F1 & .200 \\ \hline F4 & F2 & .200 \\ \hline F4 & F3 & .600^* \\ \hline Control & .033 \\ \hline F1 & .167 \\ \hline F2 & .167 \\ \hline F2 & .167 \\ \hline F4 &033 \\ \hline F4 &033 \\ \hline F1 & F2 &100 \\ \hline \end{array}$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$

	F4	500*	.175	.038	98	02
	Control	033	.17 <mark>5</mark>	1.000	52	.45
	F1	.100	.17 <mark>5</mark>	.979	38	.58
F2	F3	.300	.175	.427	18	.78
12	F4	400	.175	.155	88	.08
	Control	.067	.175	.995	42	.55
	F1	200	.175	.783	68	.28
F3	F2	300	.175	.427	78	.18
1.2	F4	700*	.17 <mark>5</mark>	.001	-1.18	22
	Control	233	.175	.670	72	.25
	F1	.500*	.175	.038	.02	.98
E4	F2	.400	.175	.155	08	.88
Г4	F3	.700*	.175	.001	.22	1.18
	Control	.467	.175	.064	02	.95
Control	F1	.033	.175	1.000	45	.52

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	F	2	067	.175	.995	55	.42		
	F.	3	.233	.175	.670	25	.72		
	F	1	467	.175	.064	95	.02		
*. The mean difference is significant at the 0.05 level.									

