Extraction of Spear Grass (*Imperata Cylindrica*) As Pro-Oxidant In Polymer Blends

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Abstract. Packaging material such as plastic bags is one of the main factors that contribute to the environmental pollution due to slow degradation. The usage of metal oxide as pro-oxidant has been proven to accelerate the degradation of these materials, but the excessive usage of this pro-oxidant will be harmful to the human body. Therefore, in this research, spear grass is investigated to be used as natural based pro-oxidant that can increase the degradation rate of the polymers. In terms of that, spear grass is extracted by using pressurized hot water extraction (PHWE) to obtain the metal element such as zinc (Zn) and ferum (Fe). PHWE is using water as a solvent which is highly favourable due to non-toxicity and non-flammable characteristics that make it easy to handle. Box-Behnken design is used to optimize the temperature, extraction time, and sample-to-solvent ratio to get the maximum amount of Zn and Fe concentration from the extracted spear grass. As a conclusion, the leaf of spear grass contributed the highest amount of Zn and Fe concentration. The highest amount of Zn and Fe concentration is achieved at 150 °C, 20 minutes, and 3 g of sample to 45 ml of water.

1 Introduction

From the last decades, the search for solutions to minimize environmental problem due to polymeric materials such as polyethylene has increase necessity when it disposed in an appropriate environment [1]. Polyethylene represents 64% of plastic materials produced as packaging and bottles, which usually discarded after only brief use due to easily contaminated, recycling these materials is not effortless and recovery cost is higher than cost producing. As the effects, several thousand tonnes of goods made from plastic materials are sent to landfill, and increasing the amount of municipal waste [2]. Due to their low degradability, generating pollution and taking space in landfills, plastics accumulate in environment.

Polyethylene is considered as an inert bulk polymer which is slowly degradable. Polyethylene in its pure form is extremely resistant to environmental degradation. Khabbaz et al. (2001) has estimated that polyethylene would degrade less than 0.5% in 100 years, and 1% if exposed to sunlight for 2 years before biodegradation [3]. Roy et al. (2007) stated that to produce degradable polyethylene is to mix it with pro-oxidant additives that can effectively improve the degradability of these materials [4]. Metallic salt is comprised in the commercial polyethylene as the most additives being used which is harmful to human body if used in excessive quantity. The usage of metal oxides as chemical based pro-oxidant such as is cobalt stearate that led to toxicity that can cause damage to major constituents of biological system.

Imperata cylindrica also known as spear grass in Nigeria, alang-alang in Asia, and cogongrass in America is a perennial rhizomatous grass which has the high up to 1.2 m. Spear grass is the most dominant, competitive, and difficult weed to control in Asia, West Africa, and Latin America [5]. Spear grass can act as natural pro-oxidant that can increase the degradation of the polymer by reducing their molecular weight. In order to do that, spear grass need to be extracted to obtain essential ingredients that can make it act as pro-oxidant. Ferum (Fe) and zinc (Zn) are metal elements that an important content in spear grass that can accelerate the degradation.

In this project, spear grass as pro-oxidant will be extracted by using pressurized hot water extraction (PHWE). PHWE is feasible green solvent extraction that used water as extraction solvent as it utilized pressurized water at elevated temperature and controlled pressure conditions. Water as solvent is highly favourable because water is non-toxicity and non-flammable characteristics that make it easy to handle. In the various report, the polarity of water can be varied close to those of alcohols at certain temperature and applied pressure. Thus water can dissolve a large range of medium and categorized as low polarity analytes [6-7].

2 Materials and Procedures

1.1 Materials

The spear grass was collected from Kuala Perlis. The spear grass was grounded to fine powder. Distilled water was used as solvent for extraction process.

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1.2 Procedures

1.2.1 Preparation of sample

The plant which is spear grass (*Imperata cylindrica*) was collected from Kuala Perlis, Perlis. The spear grass was separated to different parts which were leaf, root, and straw. It was washed thoroughly under running tap water to get rid dirt, dust, and soil. Then, the plant was undergoing drying process by using oven at 50 ˚C for 24 hours. After that, the spear grass was crushed by using grinder and sieved to 63 µm particle sized to get powder form.

1.2.2 Extraction of spear grass

To identify the best extraction method, three type of extraction which is cold extraction, hot extraction, and pressurized hot water extraction (PHWE) was conducted. For cold extraction, 2 g of spear grass was mixed with 20 ml of water and leaved at room temperature for about 3 hours. For hot extraction, 2 g of spear grass was mixed with 20 ml water and was stirred using hot plate stirrer for about 3 hours at 100 ˚C. For PHWE, 2 g of spear grass was mixed with 20 ml water and the sample was extracted at 150 ˚C for 20 minutes. After that, the extracted sample was analyzed using atomic absorption spectroscopy (AAS) and the best extraction method was proceeding for screening process.

1.2.3 Screening different part of spear grass

After the extraction process, 2 g of different part of spear grass (leaf, root, and straw) were undergone screening process by using pressurized hot water extraction (PHWE) at constant temperature, extraction time, and sample-to-solvent ratio.

After the extraction process has done, the amount of metal element contains in spear grass was determined by using Atomic Absorption Spectroscopy (AAS). The screening process was repeated for three times.

2.2.4 Extraction of selected part using PHWE

The selected part of spear grass with the highest amount of Fe and Zn that obtain from the screening process was extracted using pressurized hot water extractor (PHWE) at different temperature, extraction time, and sample-to-solvent ratio. The extracted spear grass was centrifuge at 4000 rpm at 4 ˚C for 10 minutes to separate the residue and the liquid. After that, the sample was filtered using syringe filter 0.2 µm to ensure that no residue in the sample. After the extraction process, Atomic Absorption Spectroscopy (AAS) was used to measure and analyze the concentration of Zn and Fe contains in spear grass.

2.2.5 Determining optimum conditions by using Response Surface Methodology (RSM)

The choice of design of experiment can have a large influence on the accuracy of the approximately and the cost of constructing the response process [8]. In this study, Response Surface Methodology (RSM) was used to determine the effect of different parameters that generate maximum response which is the amount of Fe and Zn. The Box-Behnken design was used to optimize the temperature, extraction time, and solvent-to-sample ratio for extraction of selected part of spear grass. Table 1 shows experimental design for optimization under Box-Behnken design.

Table 1. Experimental Design for Optimization under Box-Benhken Design

Standard run	Parameters					
	Temperature		Extraction time		Sample:solvent	
	Coded	Actual (celcius)	Coded	Actual (minute)	Coded	Actual (g/ml)
1	+1	200.00	+1	30.00	0	15.00
2	0	150.00	-1	10.00	+1	20.00
3	+1	200.00	0	20.00	-1	10.00
4	0	150.00	-1	10.00	-1	10.00
5	0	150.00	0	20.00	0	15.00
6	0	150.00	+1	30.00	+1	20.00
7	+1	200.00	-1	10.00	0	15.00
8	0	150.00	0	20.00	0	15.00
9	0	150.00	0	20.00	0	15.00
10	-1	100.00	+1	30.00	0	15.00
11	0	150.00	+1	30.00	-1	10.00
12	0	150.00	0	20.00	0	15.00
13	0	150.00	0	20.00	0	15.00
14	-1	100.00	0	20.00	-1	10.00
15	+1	200.00	0	20.00	+1	20.00
16	-1	100.00	-1	10.00	0	15.00
17	-1	100.00	0	20.00	+1	20.00

3 Results and Discussions

3.1 Comparison of extraction method

Pressurized hot water extraction (PHWE), hot extraction, and cold extraction for the extraction of spear grass, are compared in terms of temperature, extraction time, and sample-to-solvents ratio. Table 2 represents the concentration of zinc (Zn) and ferum (Fe) of spear grass by using different extraction methods.

Based on the Table 2, extraction of spear grass using PHWE is contributed the highest concentration of Zn and Fe with 0.7260 g/mL and 3.4827 g/mL respectively. It indicated that extraction by using PHWE was more efficient compared to cold and hot extraction. One of the greatest advantages of the PHWE is rapidity. An extraction time of 10 minutes provides comparable yield to those 3 hours of cold and hot extraction. The ultimate yield of concentration of Zn and Fe by PHWE was the highest which means PHWE method are 2 times more efficient than cold and hot extraction. This result means a substantial saving time, energy and plant material by PHWE.

3.2 Screening different part of spear grass

The screening process was performed using the pressurized hot water extraction (PHWE). The constant parameters which are 150 ˚C of temperature, 20 minutes of extraction time, and 15 mL of sample-to-solvents ratio was used to investigate which part of spear grass contributed the highest amount of zinc(Zn) and ferum (Fe) concentration. Table 3 and table 4 represent the average concentration of Zn and Fe after three set of replication.

Table 3. Concentration of zinc, Zn

Sample part	Concentration of zinc, Zn (g/mL)			
	First set	Second set	Third set	Average
Leaf	0.9639	0.8092	0.5627	0.7786
Straw	1.0472	0.6370	0.4092	0.4495
Root	0.1305	0.1408	0.1606	0.1439

Table 2. Concentration of Zn and Fe using different extraction methods.

Extraction method	Parameters			Concentration (g/mL)	
	Temperature (˚C)	Extraction time (min)	Sample:solvent (mL)	Zn	Fe
PHWE	150	10	10.00	0.7260	3.4827
Cold	37	180	10.00	0.2111	1.2211
Hot	100	180	10.00	0.3423	0.5404

Table 4. Concentration of ferum, Fe

Sample part	Concentration of ferum, Fe (g/mL)			
	First set	Second set	Third set	Average
Leaf	2.9646	3.8157	4.9571	3.9125
Straw	1.2746	2.2779	1.2762	1.6096
Root	3.8091	2.8215	3.8157	3.4821

Based on the result represents in Table 3 and Table 4, it indicates that the leaf part from the spear grass contributed the highest amount of zinc and ferum with 0.7786 g/mL and 3.9125 g/mL, respectively. Hence, the leaf part of spear grass was chosen to proceed for optimization of spear grass.

3.3 Optimization of spear grass using pressurized hot water extraction (PHWE)

3.31 Regression analysis in Box-Behnken Design

Response Surface Methodology (RSM) is a model which comprises of statistical and mathematical techniques to study the effect of variables and to determine the optimum conditions that generates the maximum response. In the present study, by using the Design Expert 7.1.5 software, the experimental design and the data analysis were easier to observe and interpreted. . From the screening different part of spear grass, the leaf part was chosen as a result of highest concentration of zinc (Zn) and ferum (Fe). A Box-Behnken Design which is one of the components of RSM was applied to analyze the interaction between three parameters with Zn and Fe content in spear grass as a response. Table 5 represents the result based on Box-Behnken design for Zn and Fe concentration.

Table 5. Zinc, Zn concentration (g/ml)

Run no.	Factor			Response	
	A Temperature (Celsius)	B Extraction time (minute)	C Sample:solvent (mL)	Zn Concentration (g/mL)	Fe concentration (g/mL)
1	200.00	30.00	15.00	1.2338	2.9361
2	150.00	10.00	20.00	1.0158	1.3595
3	200.00	20.00	10.00	0.9591	2.2742
4	150.00	10.00	10.00	0.6593	3.1827
5	150.00	20.00	15.00	1.7732	3.0423
6	150.00	30.00	20.00	0.9401	2.4251
7	200.00	10.00	15.00	0.4402	1.2554
8	150.00	20.00	15.00	1.5369	2.9811
9	150.00	20.00	15.00	1.5543	3.0323
10	100.00	30.00	15.00	0.6552	1.6453
11	150.00	30.00	10.00	0.8358	2.3407
12	150.00	20.00	15.00	1.5482	3.0353
13	150.00	20.00	15.00	1.5537	3.0158
14	100.00	20.00	10.00	0.7426	2.2684
15	200.00	20.00	20.00	0.7377	2.0684
16	100.00	10.00	15.00	0.6772	1.7396
17	100.00	20.00	20.00	0.6708	1.2367

3.32 Regression analysis of zinc (Zn) concentration

A regression analysis was performed to fit the empirical model based on the second order equation. The regression equation model is given as equation 1.

$$Y = 1.59 + 0.078A + 0.011B + 0.021C - 0.037AC - 0.063BC - 0.46A^2 - 0.038B^2 - 0.035C^2 \qquad (1)$$

Y was referred as the response of zinc (Zn) concentration (g/mL), A represent temperature, B represent extraction time, and C represent sample-to-solvent ratio. The variable of AB is indicated the interaction effect between temperature and extraction time, AC is indicated interaction effect between temperature and sample-to-solvent ratio, while BC is indicated the interaction effect between extraction time and sample-to-solvent ratio. A^2, B^2, and C^2 were quadratic effects to show the present of curvature in the model.
Table 6 illustrates the statistical data to test the fit of the model.

Table 6. Test of significance for regression square

Standard deviation	0.16
R^2	0.9391
Adjusted R^2	0.8609
Predicted R^2	0.7385
Adequate precision	9.050

For testing the goodness of fit of regression equation, the determination coefficient (R^2) was evaluated. Hence, the value of determination coefficient, R^2 = 0.9391 showed that 93.91% of the data obtained were distributed evenly in this quadratic model. The R^2 value $\geq$ 0.8 was acceptable. The R^2 value which more than 80% represent the good agreement between predicted data and experimental data. Adequate precision measures the signal to noise ratio. A ratio greater than 4 is desirable. Ratio of 9.050 indicates an adequate signal.

3.33 Regression analysis of ferum (Fe) concentration

A regression analysis was performed to fit the empirical model based on the second order equation. The regression equation model is given as equation 2.

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$$Y = 3.02 + 0.21A + 0.23B - 0.37C + 0.44\,AB + 0.21AC + 0.48BC - 0.75A^2 - 0.38B^2 - 0.31C^2 \qquad (2)$$

Y was referred as the response of ferum (Fe) concentration (g/mL), A represent temperature, B represent extraction time, and C represent sample-to-solvent ratio. The variable of AB is indicated the interaction effect between temperature and extraction time, AC is indicated interaction effect between temperature and sample-to-solvent ratio, while BC is indicated the interaction effect between extraction time and sample-to-solvent ratio. A^2, B^2, and C^2 were quadratic effects to show the present of curvature in the model.
Table 7 illustrates the statistical data to test the fit of the model.

Table 7. Test of significance for regression square

Standard deviation	0.19
Mean	2.34
R^2	0.9653
Adjusted R^2	0.9208
Predicted R^2	0.4500
Adequate precision	12.328

For testing the goodness of fit of regression equation, the determination coefficient (R^2) was evaluated. Hence, the value of determination coefficient, $R^2 = 0.9653$ showed that 96.53% of the data obtained were distributed evenly in this quadratic model. The R^2 value ≥ 0.8 was acceptable. The R^2 value which more than 80% represent the good agreement between predicted data and experimental data. Adequate precision measures the signal to noise ratio. A ratio greater than 4 is desirable. Ratio of 12.328 of indicates an adequate signal and good agreement between the signals to the noise ratio.

3.34 Summary

Based on the three parameter involved in the research, there were many factors that identified as the significant factor. The concentration of zinc and ferum on PHWE was successfully demonstrated by the RSM. Figure 1 and Figure 2 represents the model analysis of Zn and Fe concentration. The model obtained was good as the R^2 value is 0.9391 for Zn concentration and 0.9653 for Fe concentration. It implies the good agreement. From the model analysis, it showed that each parameter affected the concentration of Zn and Fe concentration. Based on Figure, the red dot in the elliptical indicates the highest concentration of Zn concentration which is 1.55433 g/mL and the highest concentration of Fe which is 3.03237 g/mL.

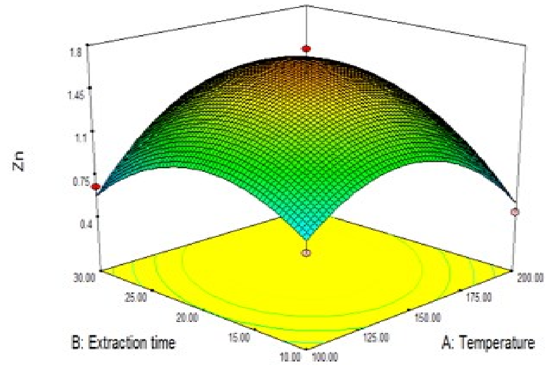

Fig. 1. Highest concentration of zinc, Zn

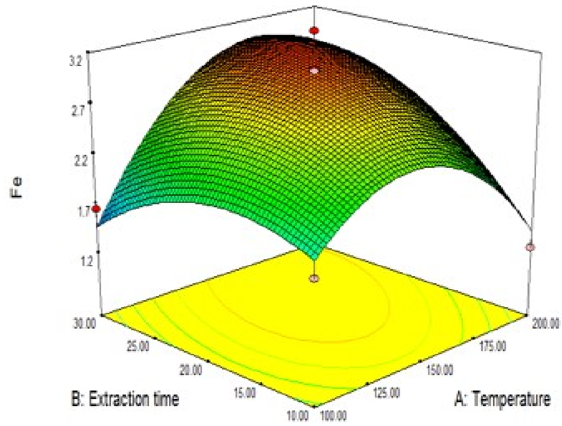

Fig. 2. Highest concentration of ferum, Fe

The sign of regression coefficients indicates that all three parameters have an effect on Zn and Fe concentration. The surface response curves in the both Figure show that the concentration of Zn and Fe increase with temperature, extraction time, and sample-to-solvents ratio, however after achieve the maximum level, the concentration of Zn and Fe will be decrease. At 150 ˚C of temperature, 20 minutes of extraction time, and 15 mL of sample-to-solvent ratio, the optimum conditions has achieved.

4 Conclusion

Spear grass sample were extracted to obtain metal elements such as zinc and ferum. These type of metal elements can be categorized as natural based pro-oxidant. Pressurized hot water extraction required a shorter time than cold and hot extraction method, provides the quality extracted spear grass. Therefore, in this research, the spear grass was extracted by using pressurized hot water extraction.

After the screening process, the leaf part from the spear grass contributed the highest amount of Zn and Fe concentration. The box-behnken design method was used to optimize the operating conditions parameters. The optimum conditions of temperature, extraction time, and sample-to-solvent ratio were adjusted at 150 °C, 20 min, and 3 g of sample to 45 mL of water, respectively, to achieve the highest amount of Zn and Fe concentration.

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