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THE MORPHOLOGICAL AND CHEMICAL CHARACTERIZATION OF ENCAPSULATED POWDER IN RODENT TUBER MUTANT PLANT (*Typhonium flagelliforme*) EXTRACT

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ABSTRACT
The rodent tuber mutant plant (*Typhonium flagelliforme*) has anticancer properties. This plant contains several bioactive compounds such as hexadecanoic acid, alpha-tocopherol, octadecanoic acid, stigmasterol, and eicosane. However, the utilization of rodent tuber mutant plant extract as a functional food is not optimal yet. One of the things that need to be done to make processed products from rodent tuber mutant plant extract is a functional beverage that has benefits like cancer prevention through emulsions. This study was used as a spray drying method, which is a process for making microparticles from necessary materials with a coating to protect active ingredients from the outside environment. The sample used was the rodent tuber mutant plant extract. The encapsulate agents were used maltodextrin, whey protein concentrate (WPC), and soy protein isolate (SPI). This study aimed to investigate the bioactive compounds and microparticles characteristics in rodent tuber mutant plant extract powders. Nanoemulsion produced from rodent tuber mutant extract was applied for the encapsulation process with several different composition ratios. The compositions were consists of maltodextrin (3), maltodextrin, WPC, SPI (2:1:1), maltodextrin & WPC (2:1), maltodextrin & SPI (2:1). The results revealed that encapsulation using maltodextrin, WPC & SPI (2:1:1) had better microparticle characteristics, and still contained bioactive compounds which were used as an anticancer such as hexadecenoic acid/ palmitic acid (6.23%), octadecadienoic acid/ stearic acid (5.24%), and oleic acid (2.15%).
Keywords: Nanoemulsion, Bioactive Compounds, Spray Drying, Scanning Electron Microscopy, GC-MS.
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INTRODUCTION

Rodent tuber (*Typhonium flagelliforme*) from *Araceae* family grows in Indonesia. Rodent tuber is an herbal plant that has a role as an anticancer and antioxidant. Rodent tuber plays an active role in anticancer activity in breast, leukemia, and lungs.[31,28,29,24] Bioactive compounds from rodent tuber mutant plants have been investigated to increase higher than the mother plant.[19,42] According to the study of Sianipar *et al.*[43] have revealed that rodent tuber mutant extracts have an IC_{50} range of MCF-7breast cancer cells of 2-13µg/mL^{-1}. Bioactive compounds in rodent tuber mutant plants that have been found include hexadecanoic acid, alpha-tocopherol, octadecanoic acid, stigmasterol, and eicosane.[41] Rodent tuber mutant extracts are still not developed optimally and useful as functional food products. The purpose of this research will become an early stage to produce functional beverage products that have efficacy as a cancer prevention agent. One of them is the development of rodent tuber mutant extract products by using the encapsulation method. In general, encapsulation is one of the product development strategies to deal with a large number of deficiencies when using systemically bioactive compounds. Encapsulation is used to prevent the loss of

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bioactive compounds.[6] The encapsulation method can provide several advantages, including protecting encapsulated compounds from degradation and adverse environment, increasing the physicochemical characteristics of bioactive compounds, maintaining stabilization, having controlled solubility, and release of bioactive compounds.[23,37] Encapsulation can be defined as the process of trapping one substance (active substance) in another substance (wall material). Encapsulated substances, except active substances, can be determined as the core, fill, active, internal, or charge phases.[30] Substances that encapsulate are described as coatings, membranes, shells, capsules, carriers, external phases, or matrices.[48,49,12] Encapsulation refers to a technology in which bioactive compounds are completely enveloped, covered, and protected by a barrier physical.[47] Various types of coatings have been used including microencapsulation including polysaccharides (maltodextrin, arabic gum, starch, and corn syrup), lipids (mono and diglycerides), and proteins (casein, whey protein, gelatin, and protein soybean).[10,13,14,7] Whey protein concentrate (WPC) exhibits the emulsifying properties and provides protection against oxidation as an encapsulation material and increases the efficiency of encapsulation as well as provides stabilizing, emulsifying, and thickening properties to the product.[51] Maltodextrin is one of the polysaccharides that is often applied to the encapsulation process as an encapsulant material and serves to protect the active compound from one of the easily damaged materials.[40] Similarly, maltodextrin also protects the encapsulated material from oxidation.[9] Soy protein isolate (SPI) has been used to develop nanoscale delivery systems for bioactive compounds, water solubility, and balance amino acid compounds as well as forms of protein interaction with functional food.[38] Soy protein isolate also has many functional groups such as carboxyl groups and amides/ amines, which are modified to various extents by existing methods to change their interactions with other natural materials, thus leading to microparticles synthesized with SPI and polysaccharides using different methods.[46] Spray drying method has been proven to be an effective technology for protecting bioactive compounds.[40] This technology converts suspensions into powder microparticles, consisting of walls and core materials. Carbohydrates, such as maltodextrin, are among the main wall materials used as encapsulation materials that protect the core.[20] Spray drying is a challenge to combine food components, bioactive compounds, dyes, cells, and enzymes in powder form in food products.[37] The spray drying method is one of the most widely used for the food industry because it has a low cost, is flexible, and increases the shelf life of the product.[12,37] This research aims to characterize and evaluate the bioactive compounds contained in the encapsulation powder product by using different encapsulant ingredients from rodent tuber mutant extract.

EXPERIMENTAL

Encapsulation Process

Rodent Tuber Mutant Plant Extract

The rodent tuber mutant plant of KB 6-1-2 (Sianipar & Purnamaningsih's collection) was macerated in 96% ethanol overnight after it had been dried.[58,59] A Whatman filter paper No. 1 was used to remove the solvent after it was filtered. Supernatants were collected under a rotary evaporator and evaporated to dryness. An emulsion was prepared from the concentrated extract.

Preparation of Nanoemulsion

A method described in this paper was used to prepare nanoemulsions using inversion. Phase inversion is the technique used to create an oil in water emulsion from water in oil emulsion. A plant extract from a mutant rodent tuber plant, KB 6-1-2 (0.01% of the final emulsion), was combined with DMSO (0.1% of the final emulsion) and tween 80 (0.01% of the final emulsion) in a beaker until dissolved. Under magnetic stirring, glycerol concentration was added at 2.5% under 700 rpm. Table-1 shows a nanoemulsion formula for water. A final volume of emulsion containing 300 mL of the weight of the formula was added to stir for 30 min to form an organic phase. In order to use the nanoemulsion for encapsulation, sample bottles were placed within them.

Preparation of Encapsulation

In the spray dry technique, a nanoemulsion of rodent tuber mutant extract was prepared with the finest particle size, polydispersity index, and zeta potential. As shown in Table-2, a total of 300 mL of nanoemulsion was added along with various encapsulation ratios. Drying took place at a flow rate of 20 mL/min at 170°C with the mix inlet temperature. A tube containing the encapsulated powder was then

passed through the cyclone one and the second, which is then weighed in order to determine the yield of the encapsulation process.

Table-1: Nanoemulsion Formulas with Different Concentrations of Extract and Surfactant

Formula Nanoemulsion	Encapsulation Emulsifier (%)			Ratio
	Maltodextrin	WPC	SPI	
F22	30	0	0	3:0:0
F22	20	5	5	2:1:1
F22	20	10	0	2:1:0
F22	20	0	10	2:0:1

The research study used maltodextrin, whey protein concentrate, and soy protein isolate as encapsulate materials. Encapsulation formulas were prepared for nanoemulsions using the inversion technique at 700 rpm.[60] The nanoemulsions were added with encapsulate materials and stayed under magnetic stirring overnight. In the spray drying process, nanoemulsion was converted into small fluid droplets by using an atomizer and dried with hot air flowing into a drying chamber.[4] The powder was stored in glass bottles coated with aluminum foil. It is impermeable to light for further analysis. The type of spray dryer used in this study was BUCHI-B19.

Table-2: Encapsulation Ratios with Different Emulsifiers

Formula	Nanoemulsion Concentration (%)				
	Extract	DMSO	Tween 80	Glycerol	Water
F22	0.1	0.1	0.1	2.5	96.3

Physical Characterization

Measurement of Particle Morphology Structure

The particle size and morphology surface were observed using Scanning Electron Microscopy (SEM) (Evo 50 Series, ZEISS, Germany) and the representative SEM images of each encapsulation formulation. The working principle of SEM is to shoot high-energy electrons on the surface of an object to imagine its surface morphology. SEM has a sharp imaging image with the surface of the object that must be an electron reflector or can release secondary electrons equipped with electrons.[35,54]

GC-MS Analysis

The GC column contained encapsulation formulae. A split ratio of 5:1 was used, and the injection temperature was 250°C. A velocity of 0.8 L per min was obtained with helium as a carrier gas. The column temperature was set at 70°C with 5°C per min. Temperatures reached 200°C after 1 min of constant heat, followed by an increase of 20°C per minute until 280°C was reached. For 28 min, the heat remained constant. In electron impact ionization mode, the mass spectrometer was powered at a voltage of 70 eV. A 90% fitting factor was applied to compare these mass spectra to the NIST database for identification of the compounds.

RESULTS AND DISCUSSION

Bioactive Compounds in Encapsulation Powder and Rodent Tuber Mutant Plant Extract based on GC-MS Analysis

Encapsulation was carried out on four formulas with different encapsulated material compositions as maltodextrin, whey protein concentrate (WPC), and soy protein isolate (SPI). The four encapsulation formulas were analyzed to determine the chemical characterization contained in the encapsulation powder. Bioactive compounds that have the potential as anticancer are still detected in all encapsulated powder formulations. In Table-3, it can be seen that the composition of the encapsulated formula with a combination of maltodextrin encapsulate, WPC, and SPI contains hexadecanoic acid/ palmitic acid (6.23%), octadecadienoic acid/ stearic acid (5.24%), oleic acid (2.15%). Hexadecanoic acid (6.87%), tridecenoic acid (5.59%), and dipalmitin (2.65%). compounds were detected in the composition of the maltodextrin formula and SPI (2: 1) (Table-4). The composition of the maltodextrin encapsulation formula and WPC (2:1), as seen in Table-5, detected bioactive compounds that act as anticancer are palmitic acid(1.19%), octadecadienoic acid (85.16%)).. The composition of the encapsulation formula using the only maltodextrin detected the type of bioactive compound, hexadecanoic acid/ palmitic acid (31.89%), octadecadienoic acid/

stearic acid (57.61%), eicosane (1.45%), tricosane (1.4%), heptacosane (1.71%), nonacosane (0.83%), and stigmasterol (1.07%) are included in the alkane class as shown in Table-6. The alkanes have potential as anticancer agents. When compared between the bioactive compounds produced by rodent tuber mutant extracts and encapsulation powders, some bioactive compounds that play an active role as anticancer compounds have some bioactive compounds that are missing or not detected. This can be indicated that some compositions of bioactive compounds are lost when the drying process becomes powder.

Table-3: The Bioactive Compounds of Encapsulation Rodent Tuber Mutant Plant Extract Using Encapsulated Maltodextrin, WPC and SPI with Ratio (2:1:1)

RT	Bioactive compound	Abundance (%)
4.338	Glycerol	17.02
4.359	Glycerol	15.74
4.531	1,2,3,4-Butanetetrol	4.23
4.911	Pentanal	26.02
6.062	Glycerol	5.86
6.255	Glycerin	1.99
9.372	2-Hexene, 2-methoxy	3.83
26.376	2-Methyl-3-(Methylcarbamyl) Bicyclo [2.2.1] heptane	1.44
27.189	2-Methyl-3-(Methylcarbamyl) Bicyclo [2.2.1] heptane	4.96
27.562	Hexadecenoic acid, methyl ester	1.09
28.134	Hexadecanoic acid	5.14
29.230	6-Octadecanoic acid	5.24
29.341	Oleic acid	2.15
34.726	Cholesterol	2.96

Table-4: The Bioactive Compounds of Encapsulation Rodent Tuber Mutant Plant Extract Encapsulated Using Maltodextrin, SPI, and WPC with Ratio (2:0:1)

RT	Bioactive compounds	Abundance (%)
4.339	Glycerin	1.72
4.414	Glycerol	2.37
5.138	Glycerin	14.19
5.959	Glycerol3,5-dihydroxy-6-methyl-2,3-dihydro-4h-pyran-4-one	11.13
27.190	Z-(13, 14-Epoxy) tetradic-11-en-1-ol acetate	2.84
27.307	Cyclododecanone, oxime	2.84
27.562	Hexadecanoic acid, methyl ester	2.14
27.886	Cyclo	3.37
28.100	Glycyl-L-proline	2.39
28.134	2-Tridecenoic acid	5.59
28.817	Oxacylohexadecan-2-one	2.21
29.231	9, 12 Octadecadienoic acid	3.41
29.341	9, 17-Octadecadienal	3.10
29.603	Decahydro-1-naphthalenol	4.64
29.672	Hexadecanoic acid	4.73
29.886	Acetic acid, 2-(4-acetoxy-butyl)-cyclohexyl ester	2.25
30.051	Cyclopropeneoctanal, 2-octyl	1.61
30.389	Longiborneol	2.78
30.472	Phenylalanine-proline-diketopiperazine	6.55
30.534	Dipalmitin	2.65
30.589	2,4-Dimethylaniline, N N-di(trifluoroacetyl)	1.03
30.692	2-Phenyl-4(ethoxycarbonyl)-6-methyl-3H-imidazole [1,5 B] pyridazine-5,7-dione	5.06
30.748	Podocarpa-6,8,11,13-tetraen-15-oic acid, 13-isopropyl-, methyl ester	2.53

Based on Fig.-1 (A-D), GC-MS chromatogram analysis revealed that the major bioactive compounds of the rodent tuber mutant extract and the encapsulation formula is hexadecanoic acid/ palmitic acid and

octadecadienoic acid/ stearic acid. GC-MS analysis showed that the major bioactive compounds in the anticancer potential were fatty acid compounds.

Table-5: The Bioactive Compounds of Encapsulation Rodent Tuber Mutant Plant Extract Encapsulated Using Maltodextrin, WPC, and SPI with Ratio (2:1:0)

RT	Bioactive compound	Abundance (%)
29.355	9,12-Octadecadienoic acid	40.89
29.340	9,12-Octadecadienoic acid	13.76
29.644	9,12-Octadecadienoic acid	19.10
30.279	9,12-Octadecadienoic acid	1.36
30.396	9,12-Octadecadienoic acid	2.43
30.616	9,12-Octadecadienoic acid	7.62
30.734	Cyclopropaneoctanal, 2-octyl	1.79
31.733	9,12-Octadecadienoic acid	1.65
26.215	Palmitic acid vinyl ester	1.19
39.174	(Z,Z)-Heptadeca-8,11-dien-1-YL Iodide	2.32
43.476	Tricyclo [20.8.0.0 (7(16)-diepoxy	1.59

Table-6: The Bioactive Compounds of Encapsulation Rodent Tuber Mutant Plant Extract Encapsulated Using Maltodextrin with Ratio (3:0:0)

RT	Bioactive compound	Abundance (%)
31.912	Hexadecanoic acid, ethyl ester	8.26
31.968	Hexadecanoic acid	22.8
32.568	9,12-octadecadienoic acid, methyl ester (linoleic acid, methyl ester/ methyl linoleate)	1.41
32.954	Ethyl (9Z,12Z)-9,12-octadecadienoate	6.67
33.03	9,12-octadecadienoic acid	24.29
33.112	(9E,12E)-9,12-octadecadienoic acid	23.4
33.616	Tricosane	1.4
34.064	Z,Z-10,12-hexadecadien-1-ol acetate	0.83
34.147	9,12-octadecadienoic acid	1.84
34.505	Eicosane	0.56
35.464	Eicosane	0.89
36.263	(6E,10E,14E,18E)-2,6,10,15,19,23-hexamethyl-2,6,10,14,18,22-tetracosahexaene	1.05
36.705	Heptacosane, 1-chloro	1.71
38.456	Nonacosane	0.83
41.063	Stigmasterol	1.03
41152	Stigmasterol	0.04

The bioactive compounds detected in the rodent tuber mutant extract of KB 6-1-2 was fatty acids(Table-7). Fatty acid groups were identified as hexadecanoic acid (1.50%), and oleamide (1.71%). In *Centratherum punctatum* herbal plants containing eicosane bioactive compounds using GC-MS analysis has a cytotoxic effect on Ehrlich Ascites Carcinoma (EAC) cancer cells.[44] Eicosane can provide cytotoxic effects on ovarian carcinoma cells in vitro and *in vivo*.[50] Tricosane has shown that it has cytotoxic activity and apoptotic ability against HCT 116 hepatocellular cancer cells within 48 hr.[1] Simultaneously tricosane modulates the activation of multiple signaling pathways and the expression of several genes associated with negative regulation of cell proliferation and cell cycle development, and positive regulation of apoptosis in human cancer cells.[1] According to the research of Bhagat *et al.*[5] that stigmasterol has potential as an antitumor.

The majority of the bioactive compounds between the rodent tuber mutant extract and the encapsulation formula have been detected to be fatty acid-containing compounds, as shown by the GC-MS analysis summarized in Table-8. Several bioactive compounds, such as hexadecanoic acid and octadecadienoic acid, are potent against cytotoxicity in cancer cells. Hexadecanoic acid inhibits growth and induces apoptosis in human gastric cancer cell line (SGC-7901) and colorectal cancer cells, demonstrating anticancer potential in cervical cancer cell lines, MOLT-4 leukemia cell line, and MOLT-4 leukemia cell line.[15,34,52,53]

Table-7: Bioactive Compounds of Rodent Tuber Mutant Plant Extract KB 6-1-2 Based on GC-MS Analysis.[42]

RT	Bioactive compounds	Abundance (%)
27.155	4-methyl-2,7-dioxa-tricyclo [4.4.0.0 (3.8)] decane	4.75
27.307	Furan, 3,4-Dimethoxy-2,5-dimethyl	13.18
27.562	Hexadecanoic acid, methyl ester	1.50
27.920	L-Proline, N-valeryl-, hexyl ester	4.86
28.010	2-Hydrazino-4 methyl-6-methylthiopyrimidine	3.53
28.093	2,5-Piperazinedione, 3,6-bis (2-methylpropyl)	21.44
29.879	10-Chlorotricyclo [4.2.1.1 (2,5) deca-3,7-dien-9-ol	4.25
30.044	3-benzyl-6isopropyl-2,5-piperazinedione	7.65
30.320	3-benzyl-6isopropyl-2,5-piperazinedione	3.69
30.368	3-benzyl-6isopropyl-2,5-piperazinedione	8.24
30.458	Cyclo-(1-leucyl-1-phenylalanyl	24.11
32.133	Oleamide	1.71

Table-8: The Composition of Bioactive Compounds as Anticancer Properties in the Extract and Encapsulations Formula

No.	Name of Bioactive Compound	Abundance (M:WPC:SPI) (%)				
		Extract	(2:1:1)	(2:0:1)	(2:1:0)	(3:0:0)
1	Hexadecanoic acid/ Palmitic acid[22,27]	1.50	-6.23	6.87	1.19	31.89
2	Octadecadienoic acid/ Stearic acid[27]	NI	5.24	5.59	85.16	57.61
3	Oleic acid[22,27]	NI	2.15	NI	-NI	NI
4	Acetic acid[33]	NI	NI	2.25	NI	NI
5	Linoleic acid[22]	NI	NI	NI	NI	NI
6	Tridecanoic acid[21,20]	NI	NI	NI	NI	NI
7	Oleamide[32,57]	-1.71	NI	-NI	NI	NI
8	Dipalmitin[8]	NI	NI	2.65	NI	NI
9	Tricosane[39]	NI	NI	NI	NI	1.40
10	Eicosane[39]	NI	NI	NI	NI	1.45
11	Heptacosane[39]		NI	NI	NI	-1.71
12	Nonacosane[39]	NI	NI	NI	NI	-0.83
13	Stigmasterol[26]	NI	NI	NI	NI	1.07

Hexadecanoic acid is proven to possess anticancer activity as a fatty acid by Al-Asmari *et al.*[2] This compound has been found to be beneficial in treating colorectal cancer and breast cancer. Researchers have found that KB 6-3-4 plant extracts of rodent tuber mutants showed cytotoxic activity against MCF-7 breast cancer cells using hexadecanoic acid compounds.[43] Metabolites of octadecadienoic acid and stearic acid have shown inhibitory effects on human breast cancer cells *in vitro*[49,16] and *in vivo.*[11,56] Apoptosis in breast cancer cells has been shown to be induced by octadecadienoic acid or stearic acid.[18,25]

Encapsulated Rodent Tuber Mutant Plant Extract Characterization Based on Morphology

The morphological structure of encapsulated rodent tuber mutant extract was analyzed using a Scanning Electron Microscopy (SEM) tool. The coatings were used maltodextrin, whey protein concentrate (WPC), and soy protein isolate (SPI) with a certain ratio. In Fig.-2A, they were showing an encapsulation with maltodextrin, WPC, and SPI coating ratio of 2:1:1 having a smooth particle surface structure and appearing round without wrinkles. The spheres are attached to each other so that it looks clustered, and the resulting particle size ranges from 10-20 µm. This can also be seen in Fig.-2B with a ratio of maltodextrin and SPI encapsulation ratio of 2:1 showing a smooth surface structure of particles and having a particle size range of 10-20 µm. Particle sizes are very diverse, making it difficult to predict the average size of encapsulated powders.The results of observations with SEM in this study indicate that the rodent tuber mutant extracts

have been encapsulated by the capsule materials, shown by the smooth surface of the capsule wall (Fig.-2A & Fig.-2B). The spheres formed do not show any wrinkles or cracks due to heating during spray drying. According to Etzbach *et al.*[57], good encapsulate results are round without wrinkles, which means the active ingredients are well encapsulated. Some particles attached to the surface of the wall can be caused by the distance of spray drying and SEM analysis that is long enough so that the powder particles coagulate with each other.

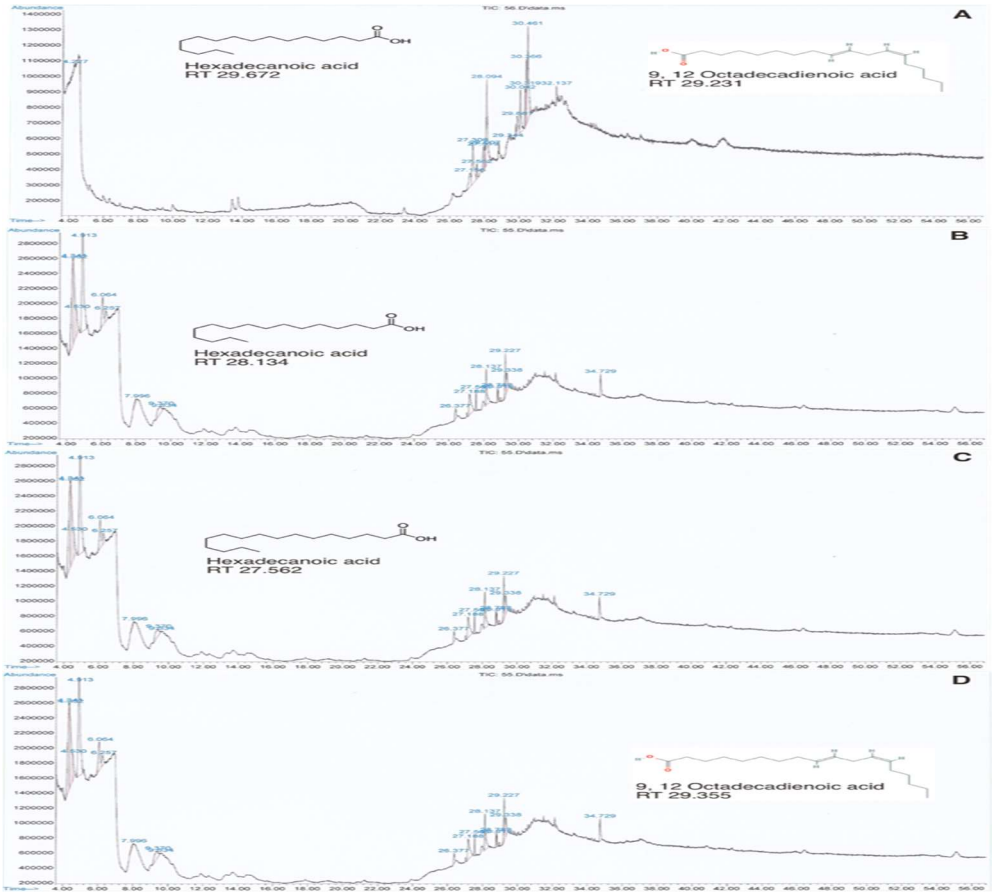

Fig.-1: The GCMS Chromatogram of Encapsulation Rodent Tuber Mutant Plant Extract, which X-axis is representing the Retention Times and the Y-axis is Representative of Relative Abundances. Note: A = The Encapsulated Ratio with Maltodextrin, Whey Protein Concentrate (WPC), Soy Protein Isolate (SPI) (2:1:1); B = The Encapsulated Ratio with Maltodextrin, and SPI (2:1); C = The Encapsulated Ratio with Maltodextrin, and WPC (2:1); D = The Encapsulated Ratio with Maltodextrin (3).

Based on Fig.-2C, the composition of maltodextrin and WPC (2:1) has a round, clustered structure, and some particles appear wrinkled. This indicates that some of the powder particles produced are still not well encapsulated. In Fig.-2D, we can see that the encapsulated powder particle structure of the encapsulated material only maltodextrin much wrinkled and deflated. This can be indicated that the deflation of particles is caused by the ballooning event, which the rupture of the capsule wall due to not being able to withstand inflation, which could have been caused by too high spray drying temperatures.[55] Huang *et al.*[17] was described the results of SEM characteristics in salicyclic acid in microcapsules producing morphological images of spherical-like spheres with dense shells should suggested as successful encapsulation. Rasyid *et al.*[36] stated that encapsulation in particles that have wrinkled and deflated shapes, chitosan is not filled with ketoprofen or empty, whereas chitosan filled with ketoprofen has an intact round shape.

The microscopic structure of the encapsulate is influenced by the characteristics of the coating material used. WPC and SPI have hygroscopic properties, which affect the microscopic structure of the encapsulate. When compared between the microscopic structure of maltodextrin (3) with maltodextrin, WPC, and SPI (2:1:1), the structure looks more attached to the composition of maltodextrin, WPC, and SPI. In the

composition of maltodextrin, WPC, and SPI are seen as sticking together because there are WPC and SPI which have hygroscopic properties.

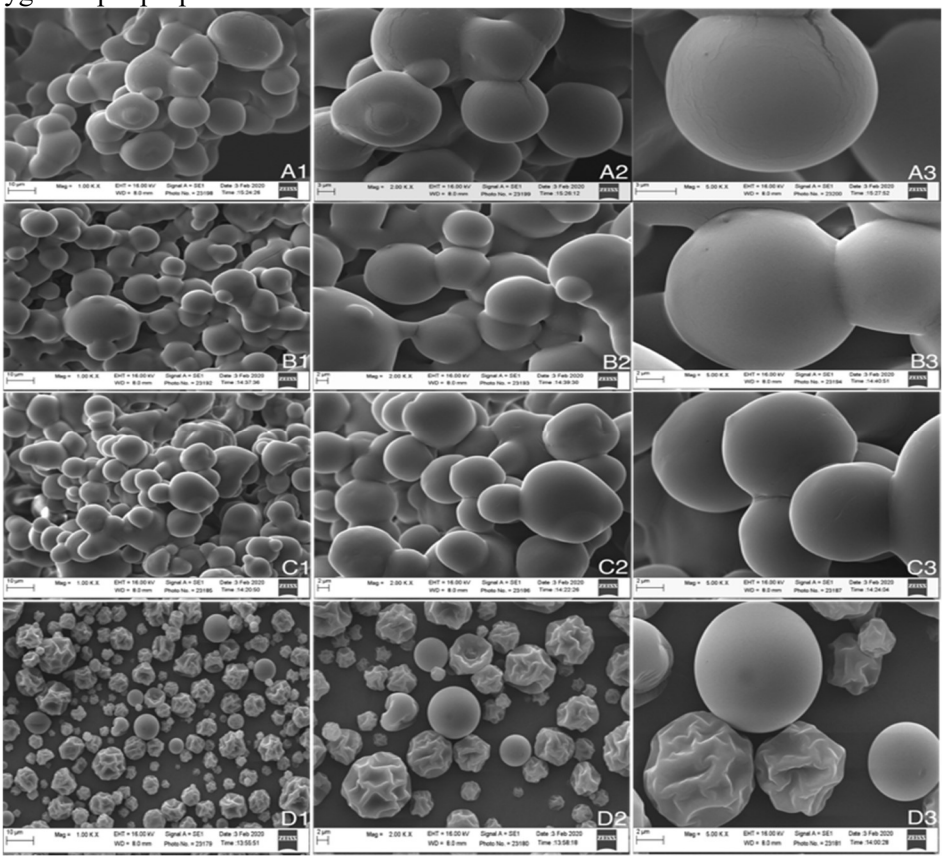

Fig.-2: Scanning electron microscope image of encapsulated ratio (A) maltodextrin, whey protein concentrate (WPC), soy protein isolate (SPI) (2:1:1) (B) maltodextrin & SPI (2:1) (C) maltodextrin & WPC (2:1) (D) maltodextrin (3*).* The magnifications used were 1000 – 5000 times.

The hygroscopic nature will cause the encapsulation to absorb the water from the environment. Molecular mobility is increased by the presence of absorbed water. Glassy molecules can become rubbery upon absorption of water. In the rubbery state, the molecules will stick to each other. Morphological differences in the results of encapsulation powders can be caused by the rate of evaporation of water during the spray drying process. The higher the temperature (faster evaporation), the smoother and clearer the surface. These results are similar to those reported by Alamilla-Beltran *et al.*[3], who found that lower inlet air temperatures produce irregular shapes. According to Young *et al.*[51], for materials using spray drying or spray drying methods, the encapsulated material must exhibit high solubility and emulsification ability, can form film layers, dry ability, and produce concentrated solutions with low viscosity. According to Stewart & Swaisgood[45], the use of encapsulation materials is usually in the form of hydrocolloids, which are long-chain polymers with high molecular weight, can dissolve, or are dispersed in water, and function as a thickener and gel-forming effect.

CONCLUSION

The powder characterization showed that all formulations still contained a bioactive compound that has potential as an anticancer. The best formula composition is the ratio of maltodextrin encapsulate material, whey protein concentrate (WPC), and soy protein isolate (SPI) 2:1:1. The bioactive compounds detected in the best formula composition using GC-MS analysis were hexadecanoic acid, octadecadienoic acid, tridecenoic acid, and dipalmitin. The best encapsulation composition has suitable clusters of physical characteristics. This indicates that the encapsulated material used can protect bioactive compounds in rodent tuber mutant extracts. Advanced analysis stages such as the percentage of encapsulation efficiency and bioavailability test can be carried out in the development of encapsulation powder products.

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REFERENCES

1. H.H. Ahmed, A.A. Abd-Rabou, A.Z. Hassan, S.E. Kotob, *Asian Pacific Journal of Cancer Prevention*, **16**, 7179(2015), https://doi.org/10.7314/apjcp.2015.16.16.7179
2. A.K. Al-Asmari, S.M. Albalawi, M.T Athar, A.Q. Khan, H. Al-Shahrani, M. Islam, *PLoS ONE*, **10**, 1(2015), https://doi.org/10.1371/journal.pone.0135814
3. L. Alamilla-Beltran, J.J. Chanona-Pe´rez, A.R. Jime´nez-Aparicio, G.F. Guite´rrez-Lopez, *Journal of Food Engineering*, **67**, 179(2005), https://doi.org/10.1016/j.jfoodeng.2004.05.063
4. L.F. Ballesteros, M.J. Ramirez, C.E. Orrego, J.A. Teixera, S.I. Mussato, *Food Chemistry,* **237***,* 623(2017), https://doi.org/10.1016/j.foodchem.2017.05.142
5. J. Bhagat, R. Lobo, N. Kumar, *BMC Complement Alternative Medicine,* **14***,* 421(2014), https://doi.org/10.1186/1472-6882-14-421
6. C.G. da Rosa, C.D. Borges, R.C. Zambiazi, M.R. Nunes, E.V. Benvenutti, S.R. da Luz, F.D. Krumreich, *LWT-Food Science & Technology,* **58**, 527(2014), https://doi.org/10.1016/j.lwt.2014.03.042
7. S. Drusch & K. Schwarz, *Europe Food Research Technology*, **222**, 155(2006), https://doi.org/10.1007/s00217-005-0020-3
8. P. Elumalai, D.N. Gunadharini, K. Senthilkumar, S. Banudevi, R. Arunkumar, C.S. Benson, G. Sharmila, J. Arunakaran, *Toxicology Letters,* **215**, 131(2012), https://doi.org/10.1016/j.toxlet.2012.10.008
9. S. Ersus, U. Yurdagel, *Journal of Food Engineering*, **80**, 805(2007), https://doi.org/10.1016/j.jfoodeng.2006.07.009
10. M. Estevez, C. Guell, S.D. Lamo-Castellvi, M. Ferrando, *Food Chemistry*, **272**, 478(2019), https://doi.org/10.1016/j.foodchem.2018.07.217
11. L.M. Evans, S.L. Cowey, G.P. Siegal, R.W. Hardy, *Nutrition & Cancer*, **61**, 746(2009), https://doi.org/10.1080/01635580902825597
12. Z. Fang, B. Bhandari, *Trends Food Science Technology,* **21**, 510(2010), https://doi.org/10.1016/j.tifs.2010.08.003
13. A. Farrag, T. Mohammed El-Messery, M. Mohamed El-Said, T.N. Soliman, H. Mohamed Fakhr El-Din, *Journal of Biological Sciences*, **18**, 373(2018), https://doi.org/10.3923/jbs.2018.373.380
14. M. Gibis, K. Thellman, C. Thongkaew, J. Weiss, *Food Hydrocolloids*, **41**, 119(2014), https://doi.org/10.1016/j.foodhyd.2014.03.024
15. H. Harada, U. Yamashita, H. Kurihara, E. Fukushi, J. Kawabata, Y. Kamei, *Anticancer Research* , **22**, 2587(2002)
16. R.W. Hardy, N.S. Wickramasinghe, S.C. Ke, A. Wells, *Advances in Experimental Medicine & Biology,* **422**, 57(1997)
17. Y. Huang, Y. Li, H. Fan, Q. Xia, *Integrated Ferroelectrics*, **152**, 22(2014), https://doi.org/10.1080/10584587.2014.901160
18. V.V. Iyer, G.W. Griesgraber, M.R. Radmer, E.J. McIntee, C.R. Wagner, *Journal of Medicinal Chemistry*, **43**, 226(2000), https://doi.org/10.1021/jm000110g
19. G. Javir, G. Joshi, S. Rojatkar, *Journal of Pharmacognosy & Phytochemistry*, **8**, 1(2019)
20. S.M. Jafari, E. Assadpoor, Y. He, B. Bhandari, *Drying Technology*, **26**, 816(2008), https://doi.org/10.1080/07373930802135972
21. E.C. Jinfeng, M.I. Mohammad Rafi, K. Chai Hoon, H. Kok Lian, C. Yoke Kqueen, *World Journal of Microbiology & Biotechnology,* **33**, 1(2016), https://doi.org/10.1007/s11274-016-2175-4

22. T. Karan, R. Erenler, *Tropical Journal of Pharmaceutical Research*, **17**, 1977(2018), https://doi.org/10.4314/tjpr.v17i10.12
23. M. Kesente, E. Kavetsou, M. Rousakki, S. Blidi, S. Loupassaki, S. Chanioti, P. Siamandoura, C. Stamatogianni, E. Philippou, C. Papaspyrides, S. Vouyiouka, A. Detsi, *Bioengineering*, **4**, 1(2017), https://doi.org/10.3390/bioengineering4030075
24. C.S. Lai, R.H. Mas, N.K. Nair, M.I. Majid, S.M. Mansor, V. Navaratnam, *Journal of Ethnopharmacology*, **118**, 14(2008), https://doi.org/10.1016/j.jep.2008.02.034
25. C. Li, X. Zhao, E.C. Toline, G.P. Siegal, L.M. Evans, A. Ibrahim-Hashim, *et al.*, *Carcinogenesis*, **2**, 1251(2011), https://doi.org.10.1093/carcin/bgr092
26. H.M.F. Madkour, M.A. Ghareeb, M.S. Abdel-Aziz, O.M. Khalaf, A.M. Saad, A.K. El-Ziaty, M. Abdel-Mogib, *Journal of Applied Pharmaceutical Science,* **7**, 023(2017), https://doi.org/10.7324/JAPS.2017.70604
27. F. Mericli, E. Becer, H. Kabadayi, A. Hanoglu, D. Yigit Hanoglu, Ozkum, D.O. Yavuz, T. Ozek, S. Vatansever, *Pharmaceutical Biology*, **55**, 1239(2017), https://doi.org/10.1080/13880209.2017.1296003
28. S. Mohan, A.B. Abdul, S.I. Abdelwahab, A.S. Al-Zubairi, Aspollah, M. Sukari, R. Abdullah. *Leukemia Research*, **34**,1483(2010), https://doi.org/10.1093/ecam/neq010
29. S. Mohan, A. Bustamam, S. Ibrahim, A.S. Al-Zubairi, M. Aspollah, *Journal of Toxicology Pharmacology*, **3**, 449(2008), https://doi.org/10.3923/jpt.2008.449.456
30. V. Nedovic, A. Kalusevic, V. Manojlovic, S. Levic, B. Bugarski, *Procedia Food Science*, **1,** 1806(2011)
31. A. Nurrochmad, E. Lukitaningsih, E. Meiyanto, *International Journal of Phytomedicine*, **2**, 138(2011).
32. Y. Ohba, Y. Kanao, N. Morita, E. Fuji, M. Hohrai, D. Miura, A. Watari, M, Yutsudo, H. Zhao, A. Ito, Y. Kita, H. Nojima, *International Journal of Cancer*, **121**, 4 (2007).
33. S. Okabe, T. Okamoto, C. Zhao, D. Chen, H. Matsui, *Journal of Gastroenterology & Hepatology*, **29,** 65(2014), https://doi.org/10.1111/jgh.12775
34. S. Paul,& R. Kundu, *International Journal of Pharmacy and Pharmaceutical Sciences*, **9**, 199(2017), https://doi.org/10.22159/ijpps.2017v9i11.21628
35. M.T. Postek, A.E. Vladar, J.S. Villarrubia, A. Muto, *Microscopy Microanalysis*, **22**, 768(2016), https://doi.org/10.1017/S1431927616011430
36. N.Q. Rasyid, P. Sugita, L. Ambarsari, G. Syahbirin, *Makara Journal Science*, **18**, 86(2014), https://doi.org/10.7454/mss.v18i3.3720
37. S. Ray, U. Raychaudhuri, R. Chakraborty, *Food Bioscience,* **13**, 76(2016), https://doi.org/10.1016/j.fbio.2015.12.009
38. M. Riche, T.N. Williams, *Aquaculture Nutrition*, **16**, 223(2010), https://doi.org/10.1111/j.1365-2095.2009.00654.x
39. R.M. Romeilah, *Research Journal of Medicine & Medical Sciences*, **4**, 332(2009).
40. R. Santiago-Adame, L. Medina-Torres, J.A. Gallegos-Infante, F. Calderas, R.F. Gonzalez-Laredo, N.E. Rocha-Guzman, L.A. Ochoa-Martinez, M.J. Bernad-Bernad, *LWT-Food Science & Technology*, **64**, 571(2015), https://doi.org/10.1016/j.lwt.2015.06.020
41. N.F. Sianipar, R. Purnamaningsih, D.L. Gumanti, Rosaria, M. Vidianty, *Pertanika Journal of Tropical Agricultural Science*, **40**, 185(2017)
42. N.F. Sianipar, R. Purnamaningsih, *Pertanika Journal of Tropical Agricultural Science*, **41**, 305(2018).
43. N.F. Sianipar, K. Assidqi, R. Purnamaningsih, T. Herlina, *Asian Journal of Pharmaceutical Clinical Research*, **12**, 185(2019)
44. R. Sivasubramanian, P. Brindha, *International Journal of Pharmacy & Pharmaceutical Sciences*, **5**, 364(2013)
45. W.W. Stewart, H.E. Swaisgood, *Enzyme & Microbial Technology,* **15**, 922(1993).
46. Z. Teng, Y.C. Luo, Q. Wang, *Food Chemistry*, **141**, 524(2013), https://doi.org/10.1016/j.foodchem.2013.03.043
47. P. Voz, M.M. Faas, M. Spasojevic, J. Sikkema, *International Dairy Journal,* **20**, 292(2010). https://doi.org/10.1016/j.idairyj.2009.11.008

48. C. Wandrey, A. Bartkowiak, S.E. Harding, 2010, Materials for Encapsulation In: Zuidam N.J., Nedovic, V.A. (Eds.) Encapsulation Technologies for Food Active Ingredients and Food Processing, Springer: Dordrecht, The Netherlands. pp 31-100.
49. N.S. Wickramasinghe, H. Jo, J.M. McDonald, R.W. Hardy, *The American Journal of Pathology*, **48**, 987(1996)
50. C.Y. Xie, H. Zhu, L.P. Lin, Z.H. Miao, M.Y. Geng, Y.J. Cai, Y. Chen, H.J. Zhao, H.B. Luo, X.W. Zhang, *Molecular Cancer Therapeutics*, **6**, 3059(2007).
51. S.L. Young, X. Sarda, M. Rosenberg, *Journal of Dairy Science*, **76**, 10(1993).
52. F.R. Yu, X.Z. Lian, H.Y. Guo, P.M. McGuire, R.D. Li, R. Wang, F.H. Yu, *Journal of Pharmacy & Pharmaceutical Sciences*, **8**, 528(2005)
53. T. Yu, F. Guo, Y. Yu, T. Sun, D. Ma, J. Han, Y. Qian, I. Kryczek, D. Sun, N. Nagarsheth, Y. Chen, H. Chen, J. Hong, W. Zou, J.Y. Fang, *Cell*, **170**, 548(2017), https://doi.org/10.1016/j.cell.2017.07.008
54. J. Yuan, W. Zhao, M. Pan, L. Zhu, *Macromolecular Rapid Communications,* **37**, 1229(2016), https://doi.org/10.1002/marc.201670059
55. R.V.D. Fernandes, S.V. Borges, D.A. Botrel, *Carbohydrate Polymers,* **101**, 524(2014), https://doi.org/10.1016/j.carbpol.2013.09.083
56. K. Zibara, K. Awada, L. Dib, J. El-Saghir, S. Al-Ghadban, A. Ibrik, N. El-Zein, M. El-Sabban, *Scientific Reports*, **5**, 1(2015), https://doi.org/10.1038/srep12598
57. L. Etzbach, M. Meinert, T. Faber, C. Klein, A. Scheiber, F. Weber, *Current Research in Food Science*, **3**, 73(2020), https://doi.org/10.1016/j.crfs.2020.03.002
58. S. Silaban, B. Nainggolan, M. Simorangkir, T.S. Zega, P.M. Pakpahan, K. Gurning, *Rasayan Journal of Chemistry*, **15**, 745(2022), https://doi.org/10.31788/RJC.2022.1526911
59. F.J. Sami, N.H. Soekamto, Firdaus, J. Latip, *Rasayan Journal of Chemistry*, **14**, 676(2021), https://doi.org/10.31788/RJC.2021.1425976
60. A. Setyawati, N.C. Rizqi, C.A. Putri, *Rasayan Journal of Chemistry*, **14**, 981(2021), https://doi.org/10.31788/RJC.2021.1426224

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