

Utilization of “Uwi” Plant (*Dioscorea* sp.) as a Renewable Bioenergy Resource

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Abstract

Many countries are currently making efforts to obtain alternative energy to reduce dependence on fossil energy resources. “Uwi” plant is a carbohydrate source that has many types and is tolerant of being planted on the upland, so it has the potential to be used as a food crop and as bioenergy. This research aims to study the potential of varieties of “uwi” (*Dioscorea* sp.) as raw material for ethanol production. The eight varieties of “uwi” were studied to assess their ethanol potential. The selected raw materials are *Dioscorea alata* L. (white yam, purple yam, and yellow yam); *Dioscorea esculenta* (Lour.) Burkill (“uwi gembili” and “uwi gembolo”); *Dioscorea bulbifera* L. (“uwi gandum”); *Dioscorea pentaphylla* L. (“uwi katak”) and *Dioscorea hispida* Dennst. (“uwi gadung”). A field experiment with Randomized Block Design was used to obtain homogenous tuber raw material, followed by laboratory methods on the assessment of ethanol production potential through fermentation. The research showed that the variety, *D. hispida* had the highest ethanol content (4.94 %), followed by *D. esculenta* (4.16 %) and *D. alata* (white yam) (3.89 %). The lowest ethanol production was obtained from *D. pentaphylla* (0.36 %) and *D. bulbifera* (1.53 %). However, in terms of productivity, *D. alata* (white-yam) is the most prospective considering its high production, ease of cultivation and tolerance of forest stands.

Keywords: Alternative energy, Bioethanol, Clean energy, Diversity, Energy plant, Root tuber crop, Upland, Yam

1. Introduction

Indonesia is the country with enormous energy consumption in Southeast Asia and fifth in the Asia Pacific, after China, India, Japan and South Korea. Increasing energy demand will increase along with the high average Gross Domestic Product (GDP) growth of 6.04 % during 2017 to 2050. Indonesia's final energy consumption (without firewood) in 2016 was still dominated by fuel oil at 47 % (Anindhita *et al.*, 2018). The dependency on fuel is a problem because, in future time, it will run out and give emissions that harm the environment (Adinurani *et al.* 2017). The threat of scarcity of fossil energy has implications for an increase in the price of itself. On the other hand, Indonesia has great potential in producing renewable energy, one of which is biofuels (Adinurani *et al.*, 2015; Tampubolon and Fauzi, 2016).

Biofuels such as bioethanol are a form of fuel that will be dominant for the future because they are renewable (Adelabu *et al.* 2018). The production of renewable fuels, especially from starchy materials such as root crops, has great potential to meet future energy

demand. The tuber group is a promising raw material for bioethanol because it has enough carbohydrates (16 % to 24 %), abundant availability and cost-effective processing (Wuryantoro *et al.*, 2020). Root crops are a better choice for bioethanol because they are cheap and less competitive as food than grains, only about 45 % of the tuber plant is consumed as food, and the rest is used for feed and industrial raw materials.

“Uwi” plant, [“uwi” is a common name in Indonesian, while in English it is called “yam” (*Dioscorea* sp.)] is a tuber group with lesser attention and has not received priority as a food source. This plant has many species and varieties, about 200 varieties with colours, shapes and flavours. Types of *Dioscorea alata* L., in particular, have colour variants and forms ranging from primary colours from white, yellow and purple, along with variations in shapes such as elongated, oblong, rounded, fingering to irregular (Wuryantoro *et al.*, 2021). The existence of this plant is increasingly marginalized from cultivation techniques and is even allowed to grow wild. This group of tubers has excellent potential as a source of carbohydrates to use as raw material for bioethanol without disturbing food needs. “Uwi” plants are very suitable for planting in various land conditions,

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especially in an upland that has not been optimally utilized. This upland area covers more than 60 % of the land area in Indonesia (Hakim et al., 2017) and more than 40 % in the world (UNESCO, 2020). This highlights the adaptability of “uwi” to the local climatic conditions and ability to thrive without significant climate impacts (Okongor et al., 2021). Some varieties can even be adapted to forest stands with higher production capacity (Wuryantoro et al., 2019). However, the product potential of this commodity for industrial use is still not well utilized (Andres and Adeoluwa, 2016). These plants are also crucial in socio-religious life in various countries. Research in Nigeria shows the importance of “uwi” plant in social, economic, religious and cultural fabric of society (Obidiegwu and Akbapio, 2017). In Ghana, these plants have even become an essential commodity being cultivated intensively involving men and women with various levels of education, which ultimately determines the level of productivity and farming efficiency (Tanko and Alidu, 2017). And in Cameroon, the commodity of yam is the primary buffer for food security and in overcoming poverty. There are seven species cultivated in this country, and about 17 wild species are feared for genetic erosion if there is no Gen Bank Institute to handle it immediately (Azeteh et al., 2019).

According to Hoover (2001) in Ulyarti (2016), the starch content of *D. alata* tubers reaches 80 % of the total dry weight (Ulyarti and Lavlinesia, 2016). The starch content in *D. alata* tubers varies greatly depending on the cultivar and growing conditions. Sahusilawane et al. (2011) stated that this plant produces an excellent source of carbohydrates in tropical and subtropical areas. In Indonesia, *Dioscorea* sp. is a type of tuber plant that has not been widely cultivated and even grows as wild plants on the edge of gardens, forests or abandoned land. This plant productivity is quite high if cultivated properly; it can reach (40 to 60) t ha⁻¹. Under certain types of forest stands, they still produce (3 to 5) kg plant⁻¹. The results of the study by Winarti and Saputro (2013) showed the starch content of *Dioscorea alata* L. (yellow yam = 83.38 %, purple yam = 86.12 %, and white yam = 86.68 %), *Dioscorea esculenta* (Lour.) Burkill (“uwi gembili” = 82.82 %), *Dioscorea pentaphylla* L. (“uwi katak” = 79.27 %), and *Dioscores bulbifera* L. (“uwi gembolo” = 84.80 %). Due to its high starch concentration, the tuber plant is considered one of the most important first-generation feedstocks for bioethanol production. Root starch can be hydrolyzed, fermented, and then refined to produce ethanol (Thatoi et al., 2016). Starch content is a good substrate for producing glucose as an intermediate product for making bioethanol. Variations in the starch content of *Dioscorea* sp. may depend on several environmental factors and agronomic practices, and the degree of maturity. The difference in starch content in *Dioscorea* sp. requires studying to determine the potential varieties as raw material for bioethanol. The research on tuber skin yielded 12.3 % bioethanol. It showed that the tubers could produce bioethanol, although not as good as cassava peels which grew 18.6 %, using the yeast *Gloeophyllum saplarium*, *Pleurotus ostreatus*, *Zymomonas mobilis* and *Saccharomyces cerevisiae* (Adiotomre, 2015). The fermentation process has been widely used in

agricultural, livestock, and fishery activities to obtain material inputs for biological productivity processes (Yasmin et al., 2020). Fermentative microbes are widely available in nature, which is accessible to isolated for various needs. It is reported that traditionally fermented vegetables are a good source of *Lactobacillus* probiotics (Mahasneh et al., 2020). Another study on fermentation resulted in ethanol yield of 13 % and 11 % from bitter yam [*Dioscorea dumetorum* (Kunth.) Pax.] and skin of water yam tubers (*Dioscorea alata* L.), respectively (Banjo et al., 2019).

2. Materials and Method

2.1. Time and Place

The research was carried out with field experiments in the Cepoko village's uplands, Ngrayun sub-district, Ponorogo district, East Java at an altitude of 700 m a.s.l., in the rainy season of November 2019 to June 2020. Subsequent experiments on ethanol production were carried out in the Faculty of Agriculture Laboratory, the Merdeka University of Madiun, from August 2020 to October 2020.

2.2. Materials and Tools

The materials used in the field experiment were eight varieties of “uwi” plants, compost and NPK “Mutiarara” fertilizer (N:P:K = 16:16:16), from PT Meroke, Indonesia. The equipment used is a hoe, sickle, bamboo for plant propagation, and measuring equipment such as meters, scales and calipers. For laboratory experiments, using tuber materials from field experiments, distilled water, well water, “tape” yeast brand “Kencana”, Indonesian product, alpha-amylase enzyme, NaOH. The equipment used is in the form of boiling equipment: gas stove, pan, peel knife and chopper, fermentation equipment in the form of closed jars, pH meters, refractometers, distillation equipment: laboratory-scale distillation, Erlenmeyer flasks, round flasks, thermometers, cooling water reservoirs, distillate containers, Bunsen heaters, gas stoves and electric stoves.

2.3. Research method

The field experiment used a Randomized Block Design consisting of eight treatments of “uwi” species (*Dioscorea* sp.), repeated three times. The treatments consisted of three sub-varieties of *D. alata* (white yam, yellow yam and purple yam), two sub-varieties of *D. esculenta* (“uwi gembili” and “uwi gembolo”), *D. hispida* (“uwi gadung”), *D. bulbifera* (“uwi gandul”) and *D. pentaphylla* (“uwi katak”). Observations were made on the production of tubers after going through homogeneous cultivation techniques. The experiment used a Randomized Block Design (Ramesh et al., 2019), and as a block, the length of fermentation time was 5 d, 7 d, and 10 d. The eight types of “uwi” tubers from field experiments as treatments were:

- U1 *D. alata* type white yam
- U2 *D. alata* type purple yam
- U3 *D. alata* type yellow yam
- U4 *D. esculenta* type “gembili”
- U5 *D. esculenta* type “gembolo”
- U6 *D. bulbifera* (“uwi gandul”)

U7 *D. pentaphylla* (“uwi katak”)

U8 *D. hispida* (“uwi gadung”)

2.4. Practices

The study has two experimental stages. The first is a field experiment using a Randomized Block Design and soil fertility as block and repeated three times. Observations variable on the production of tubers after going through homogeneous cultivation techniques. Planting was carried out on the mounds in the treatment plot with four bunds for each treatment type and repeated in three blocks. The tuber weight for seedlings ranged from 75 g to 100 g, planted at a 75 cm × 100 cm spacing. When the bulbs sprout, a double-row model of propagation poles is given (Wuryantoro, 2020).

The second stage is the laboratory experiment using a randomized block design and a grouping according to fermentation time, namely 5 d, 7 d, and 10 d. The study used eight treatments of "uwi" tubers whose productivity had been investigated from previous field experiments. The stages of ethanol production are material preparation, stripping, cutting, boiling, administering enzymes (alpha-amylase), yeast (*Saccharomyces cerevisiae*) and NPK, fermentation, filtering, refining, ending with measuring ethanol production by distillation.

Table 1. Density conversion of ethanol at various ethanol content

Ethanol content (%)	Density (30 °C)	Ethanol content (%)	Density (30 °C)	Ethanol content (%)	Density (30 °C)	Ethanol content (%)	Density (30 °C)
0	0.995 68	13	0.974 24	26	0.954 42	39	0.929 79
1	0.993 79	14	0.972 78	27	0.952 72	40	0.927 70
2	0.991 94	15	0.971 33	28	0.950 98	41	0.925 58
3	0.990 14	16	0.969 90	29	0.949 22	42	0.923 44
4	0.988 39	17	0.968 44	30	0.947 41	43	0.921 28
5	0.986 70	18	0.966 97	31	0.945 57	44	0.919 10
6	0.985 07	19	0.965 47	32	0.943 70	45	0.916 92
7	0.983 47	20	0.963 95	33	0.941 80	46	0.914 72
8	0.981 89	21	0.962 42	34	0.939 86	47	0.912 50
9	0.980 31	22	0.960 80	35	0.937 90	48	0.910 28
10	0.978 75	23	0.959 29	36	0.935 91	49	0.908 05
11	0.977 23	24	0.957 69	37	0.933 90	50	0.905 80
12	0.975 73	25	0.956 07	38	0.931 86		

3. Result and Discussion Carbohydrate and Glucose Level

From the analysis, results of carbohydrate content of fresh tubers from eight varieties of *Dioscorea* sp. were observed to be between 18.80 % and 26.60 %, sugar content before fermentation was 1.00 % to 7.00 % and sugar content after 10 d fermentation was between 0.77 % and 4.10 % (Figure 1).

Figure 1 also shows that the varieties with higher carbohydrate content in fresh tubers do not always produce high sugar content. The highest carbohydrate content in white yam (*D. alata*) was 26.60 %, with sugar content before fermentation (0 d) was 1.2 % and 10 d as 2.00 %. On the other hand, "uwi gambili" and "uwi gambolo" (*D. esculenta*) varieties with the lowest carbohydrate content (18.80 % and 20.62 %) produced the highest sugar content during the fermentation process, which indicates that the starch from these materials were more readily

hydrolyzed into sugar. This condition causes an increase in breaking the starch polymer chains in carbohydrates of "uwi gambili" and "uwi gambolo" species into sugar.

$$Ec = \frac{DE \times Vf \times 100}{Ws} \quad (1)$$

Note: Ec; Ethanol content (%)

DE: Density of alcohol at the rate of distillation (Table 1)

Vf: Volume of fermentation liquid from Ws tuber material

Ws: Sample weight of tuber material

The Density value of ethanol (DE) uses the conversion table approach in Table 1 (Perry *et al.*, 2000).

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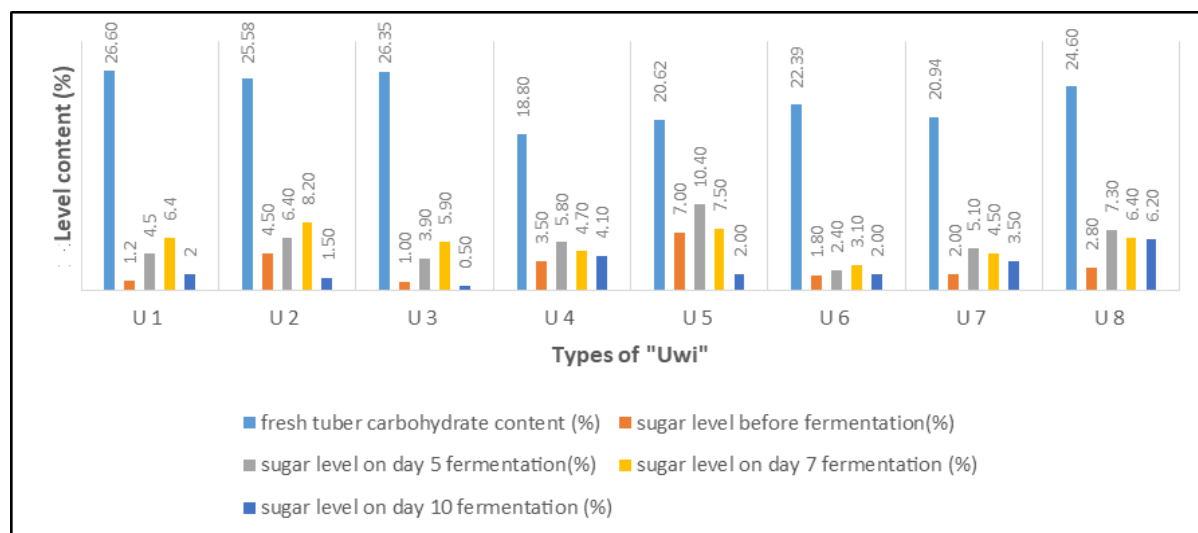


Figure 1. Carbohydrate content of fresh tubers and sugar content of various types of "uwi" at different fermentation times

The high sugar content indicates that glucose conversion to ethanol has not maximized for 10 d of fermentation. This speed of hydrolysis, in turn, yields higher ethanol (Table 2). At the same time, the enzymatic hydrolysis process of many carbohydrates in *D. alata* (white yam) needed relatively longer duration than that of "uwi gembili" to produce lower sugar levels. The α -amylase enzyme used in hydrolysis can specifically break α -1,4-glucoside bonds and quickly reduce the starch molecules size into a simple form, namely sugar (Praputri and Sundari, 2019). "uwi gadung" has a high starch content, and the amylase enzyme plays a role in the formation of high sugar until the 10th d of the fermentation process, which produces 4.94 % ethanol although not significantly different from "uwi gembili", "uwi gembolo", and white yam. This process is not the case if the hydrolysis runs faster and the hydrolyzed substrate contains enough sugar to be fermented as in the "uwi gembili" variety. High substrate concentrations can reduce energy use and overall water consumption. This is to increase the effectiveness of the fermentation process.

Based on Figure 1, the highest sugar content was in the "uwi gembolo" variety but did not show the maximum ethanol production. This may be due to the inhibition of reducing sugar fermentation (Raul *et al.* 2016). Even though the ethanol content of the "uwi gembili" variety (3.89 %) was not high, the ethanol content produced was quite a lot and not significantly different from the ethanol content of the "uwi gembolo" (4.17 %), as shown in Table 2. These results have no significant difference from the results of the research by Yuniawati *et al.* (2010), carried out for the ethanol production from white yam (3.2 %), yellow yam (2.6 %), and purple yam (2.6 %). For the "uwi gembolo" variety, the sugar content from the beginning was high (7 %), indicating that the starch was hydrolyzed into sugar, resulting in higher ethanol yield. The enzymatic and fermentation processes that run simultaneously require high energy and are generally in the early fermentation stages. In the beginning, the high amount of sugar converted a lot to ethanol during the fermentation process, so caused the amount to be lower at the end of fermentation. The amount of sugar available in

the media is still tiny. In this condition, microbial growth is slow and affects ethanol production—the level of ethanol yield is determined by the yeast activity used with fermented sugar substrate. Microbial activity in yeast enters a death phase when there are enough sugar substrates. Therefore, the microbial population does not considerably impact ethanol production, but it significantly consumes the sugar content in the fermentation substrate (Selim *et al.*, 2018). In the "uwi gembili" variety, the number of microbes grows slowly. Therefore, there is no increase in the number of microbes that can convert the substrate into bioethanol, thereby reducing bioethanol production. This moment causes the ethanol content produced to be not optimal.

The fermentation process can take a long time, depending on the yeast's ability to convert sugar into alcohol. In the fermentation process, microbes change the substrate that can turn into ethanol and produce enzymes to catalyze the conversion of complex carbohydrates to simpler sugars (Thatoi *et al.*, 2016). Enzymatic and fermentation processes that run simultaneously will reduce energy input and increase the substrate (Chen-Yeon Chu *et al.*, 2012).

Table 2. The productivity tuber of "uwi" and estimation of ethanol production

Types of "uwi"	Tuber yield (t ha ⁻¹)	Level of ethanol (%)	Estimation of ethanol product (t ha ⁻¹)
U1	47.77 c	3.56 cd	1.089 cd
U2	57.57 d	3.04 bc	1.141 d
U3	56.70 d	2.76 bc	1.049 cd
U4	29.80 ab	3.89 cd	0.517 abc
U5	33.47 b	4.17 cd	0.687 bcd
U6	25.07 ab	1.53 ab	0.162 ab
U7	23.57 a	0.36 a	0.042 a
U8	30.27 ab	4.94 d	0.885 cd

Notes: The number followed by same letters, in the same column, not significant at 5 % DMRT

The research results on ethanol content in the varieties of *Dioscorea* sp. did not show linearity in the relationship of sugar content with ethanol yield. The relationship between 10th d fermentation substrate sugar content and ethanol content follows the equation $y = 2.815 + 0.025x$.

$R^2 = 0.001$, which indicates that the effect of sugar content on the 10th d fermentation ethanol yield is insignificant. The results also showed that the ethanol yield on 5th d, 7th d and 10th d was not significant, with sugar content. The high sugar content on the 10th d fermentation indicates that the activity microbial was not maximum or the inhibition fermentation process. The addition of the enzyme glucoamylase seems necessary to increase sugar levels, given the relatively low glycemic index value of “uwi” plant tubers (Ramdath *et al.*, 2004). Braide *et al.* (2018) showed that the maximum yield of ethanol obtained at pH (4.0 to 4.5) for *Zymomonas mobilis* and pH (4.2 to 4.5) for *Saccharomyces cerevisiae*. The optimum ethanol production at 72 h fermentation process by *Z. mobilis* was 8.36 % (v/v) and 7.39 % (v/v) by *S. cerevisiae*. The sugar concentration was reduced from (11.82 to 7.69) mg g⁻¹ and (13.08 to 7.50) mg g⁻¹ in the substrate fermented by *Z. mobilis* and *S. cerevisiae*. In addition, the quality of the yeast is thought to be the cause of the low ethanol content production. Another study stated that fermentation with newly isolated yeast produced more ethanol at room temperature (11.30 %) and at 80 °C (6.15 %) than fermented with baker's yeast (Olayemia *et al.*, 2019). “Uwi” peels also had more ethanol at room temperature than at 80 °C using either of the two enzymes for fermentation (21.72 % and 27.08 %) (Olayemia *et al.*, 2019). The research results on *D. rotundata* also revealed its potential as raw material for lactic and alcohol fermentation due to the high concentration of reducing sugars after hydrolysis. However, *Saccharomyces bayanus* is not suitable to replace *S. cerevisiae* in ethanol production via SSF due to its low ethanol productivity in the first 24 h of fermentation (Villadiego-del Villar *et al.*, 2021)

3.1. Ethanol Content

The ethanol content (%) of types of “uwi” in the three fermentation stages is shown in Figure 2.

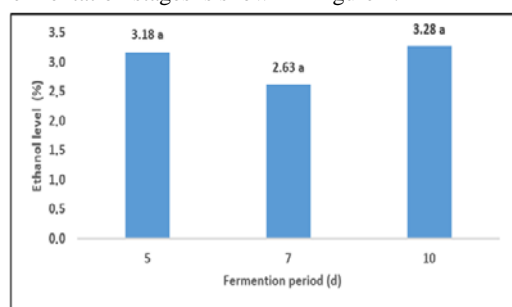


Figure 2. The average of “uwi” tuber plant ethanol content (%) in three stages of fermentation

Figure 2 shows that the fermentation time of (5 to 10) d did not show any significant changes and differences,

indicating that the fermentation process of up to 5 d is sufficient to convert carbohydrates into sugar and ethanol. The ethanol content did not increase until 10 d due to the fermentation process's delay by the high level of ethanol in the fermentation liquid, which caused the yeast to become inactive.

The amount of sugar substrate produced from the hydrolysis process seems to be an obstacle in increasing the ethanol yield. The hydrolysis process is the key to an optimum ethanol recovery from starch-based materials. The use of alpha-glucosidase enzymes and variations in hydrolysis temperature affected in increasing the rate of hydrolysis (Kusmiyati, 2010). Other studies also showed that the longer the fermentation time, the greater the bioethanol content produced until the 3th d. There was a decrease in bioethanol levels the next day (Novia and Khairunnas, 2017). The research results regarding the durian (*Durio zibethinus* L.) seed bioethanol content revealed the optimal fermentation time of 72 h. The fermentation of 20 g L⁻¹ reducing sugars for 72 h results in the highest ethanol concentration, viz., 9.85 g L⁻¹ (Purnomo *et al.*, 2016). While in the other studies on *Colocasia* Schott plants, the resulting highest bioethanol content was 19.10 % during 96 h fermentation using *S. cerevisiae* (Praputri and Sundari, 2019).

3.2. Productivity

The ethanol production process was determined by two factors, namely tuber-plant productivity and ethanol content of each type. The purple tuber of *D. alata* species produced the highest ethanol (1.141 t ha⁻¹) due to increased tuber production (57.57 t ha⁻¹), although it is not significantly different from the yellow yam. Meanwhile, “gembolo” yielded a higher ethanol yield with low tuber productivity; therefore, estimation ethanol productivity was few (0.687 t ha⁻¹). Thus, in terms of productivity, white, purple and yellow *D. alata* variety have more potential for ethanol production (Figure 3). The tuber production process determined by many factors such as climate, soil, fertilization and other cultivation techniques such as population size, seed quality, plant varieties and others. *D. alata* has more variety, differentiated based mainly on the colour and shape of the tubers. The research results of *D. alata* species revealed productivity of (5 to 7) kg plant⁻¹ with a spacing of 1.00 cm × 0.75 cm. However, the other varieties of *D. alata* had production of less than 2 kg per plant. The yellow yam is rarely used as food because of its unfavourable taste, which can be considered as a raw material for bioethanol.



Yellow yam
Figure 3. Three potential types of *Dioscorea alata*

4. Conclusion

The “uwi” plant species *D. alata*, namely white yam, purple yam and yellow yam, had the highest tuber and ethanol productivity even though the ethanol content was lower than the “uwi gadung” (*D. hispida*), “uwi gembolo” and “uwi gembili” (*D. esculenta*). High levels of tuber carbohydrates, not necessarily resulting in high ethanol yield, still depend on the speed of converting carbohydrates into sugar at the beginning and during the fermentation process. The highest percentage of ethanol yield was obtained for “uwi gadung” (*D. hispida*) at 4.94 %, followed by “uwi gembolo” and “uwi gembili” at 3.89 % and 4.17 %, and the lowest was for “uwi katak” (*D. pentaphylla*) at 0.36 %. The highest ethanol productivity was obtained by the species of “uwi kelapa” (*D. alata*) with production of (1.049 to 1.141) t ha⁻¹ of ethanol, considering higher tuber production.

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