



# Sweetpotato Research Front

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## Acceleration of Genome Sequencing and its Utilization

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General-Director of Kyushu Okinawa Agricultural Research Center, NARO



In 2004 whole genome sequencing of crops was completed in rice for the first time, after seven years of research using large-scale research funding by an international consortium of ten countries led by Japan.

The progress of rice research after the completion of whole genome sequencing has been remarkable. A large number of physiologically and agriculturally important genes have been identified and functionally characterized, resulting in a drastic increase in publication of rice-related articles in such leading scientific journals as *Nature* and *Science*. It has also promoted utilization of DNA markers closely linked to agricultural traits in the selection process of rice breeding. Recently, development of new technologies, such as genomic selection and genome-wide association study, has been underway to improve complicated traits controlled by many quantitative trait loci (QTLs), including yield and taste.

Furthermore, technical innovations and cost reduction in genome sequencing have dramatically advanced since the completion of rice genome sequencing. As a result, whole genome sequencing of other crops (e.g., barley,

wheat, soybean, Chinese cabbage, tomato, carnation, and eggplant) have also been completed by consortiums or research groups in which the National Agriculture and Food Research Organization (NARO) or National Institute of Agrobiological Sciences (consolidated to NARO in 2016) participated.

In addition, draft genome sequencing of *I. tirifda*, a diploid wild relative of sweetpotato, was reported in 2015 (<https://doi.org/10.1093/dnares/dsv002>). A high-density genetic linkage map of the cultivated sweetpotato genome was constructed through identification of differences in the genome sequences of a cultivated sweetpotato and the diploid wild relative (<https://doi.org/10.1038/srep44207>). These achievements are important bases for the whole genome sequencing project of cultivated sweetpotato that was started in 2014. Also, they may accelerate sweetpotato breeding by facilitating development of DNA markers to improve starch yield and quality, or resistance to important pests and diseases (e.g., southern root-knot nematode and stem rot).

The progress of genome studies and their application in sweetpotato has been behind other major crops because of its complicated autohexaploid genome; however, it is now time to accelerate these research activities.

# Research Paper

## Effect of Sweetpotato (*Ipomoea batatas* L.) Leaf Consumption on Rat Lipid Metabolism

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The nutrient content of the leaves of sweetpotato cultivar Koganesengan was determined to be 35.0kcal energy, 89.0g water, 3.9g protein, 0.8g fat, 1.0g carbohydrates, 1.2g minerals, and 4.1g dietary fiber/100g fresh weight. The polyphenol content present in the leaves was 11.5g chlorogenic acid equivalents per 100g dry weight. Some reports indicate that the consumption of dietary fiber and polyphenols is effective in treating dyslipidemia. Therefore, we investigated the influence of sweetpotato leaves, which represent an abundant untapped biomass resource that is rich in both dietary fiber and polyphenols, on lipid metabolism in rats fed with a high-fat diet (HFD). We conducted tests using an HFD supplemented with freeze-dried sweetpotato powder (SPLP). After 35 days of rearing, the expected obesity was observed in the control group, whereas weight gain was suppressed in all the groups that consumed the SPLP-supplemented HFD in a dose-dependent manner (Table 1). Statistically significant suppression was observed for the 5% SPLP group

compared with the control group. Furthermore, a clear dose-dependent decrease in adipose tissue weight was observed for the 1%, 3%, and 5% SPLP groups. The triglyceride and total cholesterol levels in plasma, and the total cholesterol level in the liver were significantly lower in rats who fed on SPLP than in rats on an HFD. These results suggested that the simultaneous consumption of sweetpotato leaf and an HFD inhibited excessive accumulation of adipose tissue in a rat. Collectively, our data suggest that sweetpotato leaves can be a novel leafy vegetable with high dietary fiber content, and might have anti-obesity effects through improved lipid metabolism when consumed with high fat foods.<sup>1)</sup> Further studies are necessary to confirm the anti-obesity effect of SPLP and to identify the SPLP component that directly contributes to improved lipid metabolism.

### REFERENCES

1) Kurata, R. et al., Food Sci. Technol. Res. 23, 57-62 (2017).

Table 1. Effects of sweetpotato leaf consumption on body weight, feed intake, tissue weight, plasma, and liver lipids in SD rats

	HFD	HFD+SPLP		
		1%	3%	5%
Body weight (g)				
Initial	147.3 ± 5.16	147.3 ± 4.80	147.3 ± 5.09	147.3 ± 4.84
Final	425.0 ± 24.97 <sup>a</sup>	409.2 ± 10.53 <sup>ab</sup>	403.2 ± 8.11 <sup>ab</sup>	390.6 ± 15.69 <sup>b</sup>
Feed intake (g/rat/35d)	588.4 ± 25.91 <sup>a</sup>	567.8 ± 23.23 <sup>ab</sup>	548.2 ± 9.80 <sup>b</sup>	581.5 ± 20.94 <sup>ab</sup>
Energy intake (kcal/rat/35d)	3020.93±133.01 <sup>a</sup>	2903.64±118.81 <sup>ab</sup>	2782.01±49.74 <sup>b</sup>	2928.27±105.45 <sup>ab</sup>
Tissue weight (g/100g of body weight)				
Liver	21.75 ± 1.70	20.58 ± 2.91	20.70 ± 2.38	20.88 ± 2.05
Kidney	3.19 ± 0.33	3.08 ± 0.20	3.11 ± 0.17	2.85 ± 0.17
Adipose tissue	8.85 ± 1.32 <sup>a</sup>	7.23 ± 1.01 <sup>b</sup>	7.01 ± 0.51 <sup>b</sup>	6.25 ± 0.95 <sup>b</sup>
Plasma lipids				
Triglyceride (mg/dl)	178.63 ± 44.76 <sup>a</sup>	134.08 ± 44.41 <sup>ab</sup>	110.67 ± 61.23 <sup>ab</sup>	66.96 ± 35.29 <sup>b</sup>
Total cholesterol (mg/dl)	93.64 ± 17.29 <sup>a</sup>	74.31 ± 6.31 <sup>b</sup>	76.43 ± 6.66 <sup>b</sup>	79.64 ± 17.29 <sup>ab</sup>
HDL- cholesterol (mg/dl)	63.46 ± 10.37	57.48 ± 6.90	60.92 ± 8.90	61.85 ± 8.70
Glucose (mg/dl)	139.53 ± 19.30 <sup>a</sup>	120.99 ± 8.41 <sup>b</sup>	127.55 ± 7.31 <sup>a</sup>	124.74 ± 8.19 <sup>a</sup>
Liver lipids				
Triglyceride (mg/g)	95.27 ± 10.51	118.94 ± 14.12	110.68 ± 15.08	111.14 ± 28.32
Total cholesterol (mg/g)	8.57 ± 0.39 <sup>a</sup>	7.99 ± 0.94 <sup>a</sup>	7.49 ± 0.37 <sup>a</sup>	7.07 ± 1.03 <sup>b</sup>

Values are means ± SD (n = 7).

Means in the same row and with different letters are significantly different (p<0.05).

HFD: High fat diet group. HFD+SPLP: High fat diet + sweetpotato leaf powder.

# Research Paper

## Color and Anthocyanin Content in Drum-Dried Powder of Anthocyanin-Enriched Purple-Fleshed Sweetpotato Breeding Line Kyushu 166

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A new purple-fleshed sweetpotato breeding line, Kyushu 166, was developed at the Miyakonojo Research Station of the NARO Kyushu Okinawa Agricultural Research Center on Kyushu Island. This breeding line contained a higher level of anthocyanin than the three cultivars already released (Ayamurasaki, Murasakimasari, and Akemurasaki). This breeding line was expected to be utilized for processing. In recent years, the most dominant cultivar of purple-fleshed sweet potato on Okinawa Island, southern Japan, has been Churakoibeni. In this study, we examined the color and anthocyanin content in drum-dried powder made of Kyushu 166 tubers cultivated on Okinawa Island, compared with that made of Churakoibeni.

Both purple-fleshed sweetpotatoes were harvested in farmlands of Itoman City on Okinawa Island in September of 2014 and 2015. The tubers of purple-fleshed sweet potatoes were peeled and steamed. The steamed tubers were mashed, and then dried using a drum dryer. Table 1 indicates the color values ( $L^*$ ,  $a^*$ , and  $b^*$ ) for both powders. The  $L^*$  value for Kyushu 166 powder was lower than that for Churakoibeni powder, implying that Kyushu 166 powder is darker than Churakoibeni powder. The  $a^*$  and  $b^*$  values for Kyushu 166 powder were also lower than those of Churakoibeni powder, implying that Kyushu 166 powder is less red and is bluer than Churakoibeni powder. The distance between two colors,  $\Delta E^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$ , was 11.1, indicating

Table 1. Color values ( $L^*$ ,  $a^*$ , and  $b^*$ ) for drum-dried powders made of Kyushu 166 and Churakoibeni

Cultivar/line	year	$L^*$	$a^*$	$b^*$
Kyushu 166	2014	47.6	17.5	-6.3
	2015	43.6	17.4	-5.3
Churakoibeni	2014	58.2	19.2	-3.1
	2015	54.0	21.1	-3.7

that the color difference was appreciable, as illustrated in Photo. 1. As depicted in Fig. 1, peonidin-group anthocyanins were dominant over cyanidin-group ones in Kyushu 166 powder, like Churakoibeni powder. The total content of anthocyanin in the powder made of Kyushu 166 was 12.5mg/g on average, which was 2.6 times higher than that from Churakoibeni (4.7mg/g on average). Due to high anthocyanin content, the color of Kyushu 166 powder is darker; however, we confirmed that it is just as good for mixing with flour to make confectionery.

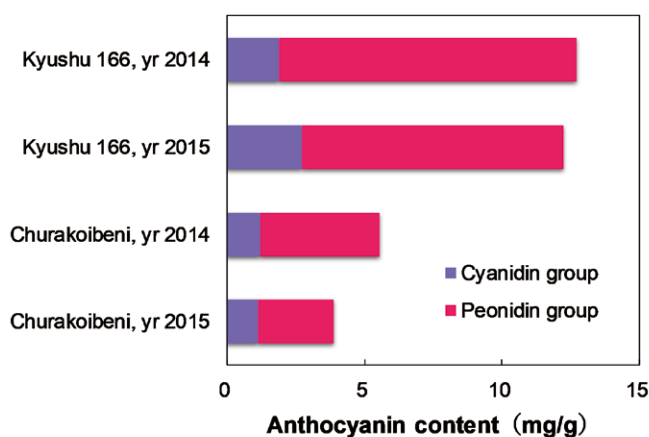


Fig. 1. Anthocyanin content in drum-dried powder made of Kyushu 166 and Churakoibeni tubers



Photo 1. Drum-dried powder made of Kyushu 166 (left) and Churakoibeni (right)

# Research News

## Report of the 7<sup>th</sup> China-Japan-Korea International Sweetpotato Workshop

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The 7<sup>th</sup> China-Japan-Korea sweetpotato workshop was held at Jinan city, Shangdong Province, China, from October 10 to 14, 2016. More than 200 sweetpotato researchers attended the workshop from China, Japan and Korea, as well as several participants from Cornell University (USA) and the International Potato Center (China, India and Peru). Fifteen Japanese researchers attended from Kazusa DNA research Institute, Okayama University, Kirishima Shuzo Co., Ltd., and NARO.

During the workshop, 27 oral presentations were made in five sessions: 1. Keynote lectures. 2. Sweetpotato biotechnology, genetic resources, and breeding. 3. Sweetpotato cultivation and physiology. 4. Sweetpotato pest-disease control. 5. Sweet potato processing, quality and functionality. The present status of sweetpotato production and research in each country was reported in the keynote lectures and recent progress in sweetpotato research was introduced in the following sessions. The steering committee meeting of the trilateral research association of sweetpotato (TRAS) and the business meeting on the sweetpotato genome sequencing project of TRAS were also held during the workshop.

At the end of the workshop, the general assembly of TRAS was held, in which Prof. Makoto Tahara of Okayama University was selected as the next president. Also, the 8<sup>th</sup>

workshop was decided to be held in September 2018, at Jeonju, Korea. The best presentation awards were selected from the oral and poster presentations. Japanese participants Mr. Kishimoto and Mr. Aikawa from Okayama University and Dr. Kurata from NARO won the best presentation awards.

On the afternoon of October 12, the participants visited the Shandong Academy of Agricultural Sciences and looked over their facilities for sweetpotato research. Also, on October 13, the participants visited the Sishui Lifeng Food Co., Ltd., the largest sweetpotato processing company in China. The company processes 300 to 500 thousand tonnes of sweetpotatoes for starch and noodle production per year and also produces flour and confectionaries from sweetpotatoes with different flesh colors.



# Research News

## Current Status of the Sweetpotato Genome Sequencing Project under TRAS Collaboration

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Sweetpotato is an autohexaploid species having 90 chromosomes ( $2n = 6x = 90$ ), and heritage manners of agronomically important traits are extraordinarily complex due to the complex genome structure. In an effort to overcome the difficulty of genetic and genomic analyses of sweetpotato, an international genome sequencing project was launched in 2012 by the Trilateral Research Association of Sweetpotato (TRAS) genome sequencing consortium consisting of six organizations: the Institute of Sweetpotato Research, the Chinese Academy of Agricultural Sciences (China), China Agricultural University (China), the Rural Development Administration (Korea), the Korea Research Institute of Bioscience and Biotechnology (Korea), the National Agriculture and Food Research Organization (NARO) (Japan), and the Kazusa DNA Research Institute (Japan).

First, *de novo* genome assembly of the wild relative diploid species *I. trifida* was performed in order to construct a reference genome of sweetpotato. Genome assembly with Illumina reads were constructed in two lines of *I. trifida* developed by NARO: the selfed line Mx23Hm and the highly heterozygous line 0431-1. The genome of Mx23Hm was further sequenced by Pacbio, and assembled together with Illumina scaffolds. As a result, 650 scaffolds were constructed with a total length of 496Mb and a N50 length of 5.3Mb. Fifteen pseudomolecules with a total length of 333Mb were also developed based on an SNP linkage map developed using the dd-RAD-Seq method.

Meanwhile sweetpotato genome sequencing of the Chinese variety Xushu 18 was performed by Illumina and PacBio platforms. PacBio reads with a total length of 181.5Gb were assembled into 9,179 primary contigs and 8,806 haplotigs by FALCON unzip. The total length of the primary contigs was 1.8Gb (N50 = 325.5Mb), and that of the haplotigs was 336kb (N50 = 44.9kb). Pseudomolecules are under development based on an SNP linkage map consisting of 28,087 SNPs mapped across 96 linkage groups. The genome sequencing of sweetpotato is expected to be published in 2018.



The Chinese variety Xushu 18 was selected for the genome sequencing project.



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