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Short Communication

Effect of Acid Modification on the Physico-Chemical Properties of North Sulawesi's Giant Swamp Taro (GST) Starch *(Cyrtosperma merkusii)*

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ABSTRACT

Giant swamp taro (GST, *Cyrtosperma merkusii*) is an underutilized tropical endemic crop of North Sulawesi. GST starch has the potential to be utilized in various food products such as dry noodles, artificial rice, bioplastic and various other applications. This study aimed to determine the effect of acid modification on the physico-chemical properties of GST starch. Starch was extracted from GST corms by maceration in 2% sodium chloride solution. GST starch was modified by acid hydrolysis using 0.2 M citric acid solution, followed by neutralization in 1 M sodium hydroxide solution. The starch samples (native and modified) were analysed for the following physico-chemical parameters; pasting properties, viscosity, total starch content, swelling power. The Fourier transform infrared (FTIR) spectroscopic analysis and Field emission scanning electron microscopy (FESEM) of the starch were also carried out. The results revealed that acid modification of GST starch has significant effect on its physico-chemical properties, microstructure and FTIR spectrum. Native GST starch had high amylose and amylopectin contents with a high ratio of amylopectin to amylose. Acid modification of the GST starch resulted in a starch with lower amylose and amylopectin content. Consequently, lowering the viscosity and gelling properties of the starch. Therefore, modification of GST starch by acid hydrolysis could result in starch product with better physic-chemical properties that could be employed as food thickeners. However, for applications where a more stable and stronger gel matrix is required, other modification methods such as the cross-linking and heat moisture treatment (HMT) methods should be considered.

***Keywords*:** *Acid hydrolysis, Cyrtosperma merkusii, Giant swamp taro, Starch, Modified starch*

Introduction

Giant Swamp Taro (GST) [*Cyrtosperma merkusii* (Hassk.) Schott (family Araceae)] Locally called daluga is an underutilized tropical crop endemic to Micronesia and thought to be native to Indonesia since it was found to grow only in the Sangihe and Talaud Islands, North Sulawesi. GST grows in a coastal environment and has a unique resistance to salinity. Considering the ecological habitat of GST, it may have the potential to help mitigate the impact of climate change when cultivated as a source of food starch for marginal coastal communities. GST is a paludiculture crop (a crop that can be cultivated in rewetted peat land conditions to reduce gas emissions) which may serve as a source of nutrients for iron uptake.[1]

GST tubers or corms are rich in starch with fine granules. GST are potential staple food material with a high carbohydrate composition (81-83%), with a starch content of 65.52 ± 0.02%, amylose and amylopectin composition of 29.63 ± 0.01% and 32.88 ± 0.02%, respectively and crude fibre of 18.55%.[2]

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GST is also a healthy food source with a resistant starch composition of 11.37 ± 0.03% and digestive starch content of 44.30 ± 0.26%, which is a healthier digestive character in processed foods by reducing carbohydrate loading and conversion into blood sugar and therefore has the potential as functional foods with low glycaemic index.[2,3] GST starch can be developed into advanced food products like dried noodles.[4] The pulp biomass or waste from GST starch processing contains various important components such as fibers, carbohydrates, ash, proteins, lignins, cellulose and hemicellulose which are a good source of cellulose nanocrystal (CNC) material.[5] Waste biomass of GST starch processing are good sources of carbon and inorganic nitrogen that may serve as natural feed source for fisheries to increase tilapia survival rate and a feed source that can reduce blood glucose levels in broiler chickens.[6,7]

Starch in the food industry has an important role in food products such as canned food, baked food, frozen food, salad dressings and baby food due to their good gelling and thickening property. Resistant starch is commonly used as a functional food ingredient, especially in food products that require large amounts of dietary fiber that help reduce the absorption of fat and glucose in the small intestine. Fermentation of resistant starch by some prebiotic microorganisms will produce metabolites in the form of short chain fatty acids (SCFA) which can increase the immunity of colonic cells, reduce the incidence of infection by pathogenic bacteria and help reduce the risk of colon cancer.[8-10]

Starch made from plant perimedular and plant tubers has been used for various derivative products (food and non-food), but native starches in general has properties that is limited in its application due to its low thermal resistance, low shear resistance, low solubility in cold water, high viscosity, low swelling power and the retrogradation tendency to staling and syneresis such that the final product texture and structures

can be affected, thus limiting its application in the advanced industries.[11,12]

The poor physical and chemical properties of some native starches limit their use as raw materials in the processing of advanced functional food products in the food industry. Therefore, modification efforts are needed to improve starch characteristics such as solubility, texture, and tolerance to high temperatures. Besides, starch modification has huge potential to provide functional effects to food products.[13] One way to improve the characteristics of starch is to increase the amount of short-chain amylose and the level of retrogradable starch by several modification processes such as pressure-cooling heating process, starch hydrolysis by chemical treatment using propionate esterification method,[14] acetylation,[15] cyclodextrin glycosyltransferase or specific cyclodextrinase,[16] and hydroxypropylation,[17] enzymatic treatment using amylase enzyme,[18] physical modification of starch by ultrasonic wave method,[19,20] autoclaving-cooling, heat moisture treatment (HMT),[21] cross-linking/annealing (ANN),[22] and extrusion method.[23] Starch modification generally causes changes in the polysaccharide molecules of the starch, causing changes to the properties and functions of the starch including the heat characteristics of pastes and gels as well as the digestibility of the starch.[24] Physical modification of starch can produce starch with similar characteristics as chemical modification. For example, lightly cross-linked starch is known to increase its tolerance to low pH, high temperatures, and high shear stress.[25]

A previous study has modified starch using hydroxypropyl methylcellulose and sodium tripolyphosphate which were able to increase the swelling power of corn starch.[26] Chemical modification with esterification method using octenyl succinic anhydride can increase digestible starch and improve rheological characteristics and thermal properties of potato starch,[27] cross-linking modification can increase the degree of substitution, increase the viscosity, swelling power, syneresis, and pH tolerance of canna starch,[28] chemical modification through esterification and acid alcohol can increase alterations in the physicochemical, pasting, particle size, and morphological action, lowering of the amylose content of rice bean starch and swelling power of rice bean starch.[29] This study aims to determine the changes in physicochemical characteristics of native GST *(Cyrtosperma merkusii)* starch upon modification by acid hydrolysis.

Materials and Methods

Plant Collection and Identification

GST (*Cyrtosperma merkusii*) was obtained from coastal swamps of Pokol village, Sangihe Island, North Sulawesi, Indonesia (125°9′28″E–125°56′57″E) on 11th March, 2022. The plant material was identified and authenticated by Mark W. Skinner of the National Plant Data Center, US Department of Agriculture - Natural Resources Conservation Service (USDA-NRCS). The voucher number/taxonomical 506754 reference was obtained from the Integrated Taxonomic Information System (ITIS) Report for *Cyrtosperma merkusii* (Hassk.) Schott, of the order *Alismatales* and family *Araceae*.

Starch Extraction

GST corms were cleaned by washing in running water and then pealed. The pealed GST corms were then crushed to a pulp and washed repeatedly with 10% sodium metabisulfite solution until the pulp slurry has been cleaned of the mucus. The washed pulp was macerated in 2% sodium chloride solution and left for the starch to sediment. The sedimented GST starch was then collected and dried at 25ºC under constant low airflow current in a cabinet. The dried starch sediments were then crushed using a disc miller.[4]

Starch Modification

GST starch was modified by acid hydrolysis according to the method previously described.[30] GST starch was suspended in water and heated in an autoclave at 121°C for 30 min and then cooled at 4°C for 24 h. The starch suspension was diluted in 0.2 M citric acid solution (10%, w/v) then incubated at 45°C for 24 h, the starch was then neutralized by the addition of 1 M NaOH until the pH became 7. The starch was separated from the supernatant by centrifugation at 3000 rpm for 20 min and then the starch sediment was oven dried at 50°C for 48 h.

Analyses of Starch

The starch samples were subjected to the following analyses; pasting properties, texture, total starch content, swelling power, Fourier transform infrared (FTIR) spectroscopic analysis and Field emission scanning electron microscopy (FESEM).

Determination of Total Starch Content

The total starch content was determined by gravimetric method described by the Association of Official Analytical Chemists (AOAC).

Determination of Swelling Power, Solubility and Expanding Capability

The modified GST starch samples (1.0 g) were mixed with 50 mL of distilled water and heated at 90°C for 30 min. The gelatinized samples were then cooled to room temperature and centrifuged at 1000 g for 20 min. The supernatant was dried at 110°C to a constant weight to quantify the soluble fraction. The solubility was expressed as the percentage (%) of dried solid weight based on the weight of the dry sample. The swelling power was represented as the ratio of the weight of the wet sediment to the weight of the initial dry sample (g/g), while the expanding capability was indicated as the ratio of the overflow volume of the distilled water to the constant weight of the dry supernatant (mL/g).[31]

Pasting Analysis

The pasting properties of the starch were tested using a Rapid Visco Analyzer (RVA-TecMaster, Macquarie Park, Australia) with the RVATM General Pasting Method STD2 procedure. Modified starch samples (3.5 g, 14% moisture content) were mixed with 25 g of distilled water in a disposable aluminium RVA canister. The sample was then spun at 160 rpm (500C) for 1 min, heated to 95ºC, and held at this temperature for 5 min, then allowed to cool to 50ºC in 7.5 min, and held at 50ºC for 2 min.[21]

Fourier Transform Infrared (FTIR) Spectroscopic Analysis

The FTIR spectra of the starch samples were obtained on a spectrometer (Perkin Elmer modelo Frontier, Waltham, MA) using the attenuated total reflection (ATR) accessory. Each FTIR spectrum was recorded in the wavenumber range of 4000-400 cm^{-1}, with a resolution of 4 cm^{-1} and an average of 64 scans.

Field Emission Scanning Electron Microscopy (FESEM)

FESEM system (NOVA Nano SEM 450, USA) was used to determine the surface morphology of the modified starch. An acceleration voltage of 5-30 kV was applied to obtain the best images and to minimize sample damage. The starch was transferred and fixed on the surface of a carbon tape and sputtered with a thin layer of gold for 60 s before FESEM imaging. The best magnification was selected to obtain clear and representative images.[32]

Results and Discussion

Properties of the modified starch (pasting properties, total starch content, swelling power and expanding capability) are presented in Table 1.

The modified GST starch began to swell at 90.70 ± 2.11°C, and as the temperature rises, the viscosity increased indicating the rupture of the granules and the leaching of amylose from gelatinizing starch granules, until a peak viscosity of 326.67 ± 25.40 cP was attained (Table 1), which indicates the maximum swelling power and amylose leaching point of the granules.[33,34] Low peak viscosities (<5700 cP) as observed in this study indicates that the modification of the GST starch had extensively hydrolyzed the starch and resulted in a significant loss of granule size.[35] The low values also obtained for the other viscosity parameters such as the trough, setbacks and final viscosity confirm that the native GST starch has undergone extensive hydrolysis. The appearance of very rugged aggregates in the microstructure of the starch also indicates that most of the smooth amorphous surfaces of the starch have also been hydrolysed by the acid causing defects to the denser crystalline structure of the granule.

The modification of GST starch by acid hydrolysis resulted in a GST starch with low total starch content of 46.86 ± 1.34%, low swelling power of 4.22 ± 0.58 g/g, solubility of 5.73 ± 0.42% and an expanding capacity of 15.9 ± 2.15 mL/g (Table 1). Native GST has been found to have high carbohydrates content of 89.58% in which 68.43% of it is total starch.[2] Out of the 68.43% total starch content, GST had 15.58% amylose and 52.86% amylopectin.[2] The acid hydrolysis of the GST starch has considerably reduced the total starch content to 46.86 ± 1.34% with an amylose content of 13.02 ± 0.22% and 33.84 ± 1.12% amylopectin.

The low swelling power is a reflection of the low amylose-amylopectin ratio where the amylose content has been considerably hydrolyzed by the acid leaving more branched amylopectin content, resulting in starch with more branched structure and lower swelling and expanding capabilities.[36] The solubility of the acid modified GST starch (5.73 ± 0.42%) was apparently within the normal range of solubility of starches.[37] As food starch, the proportion of amylose-amylopectin and low swelling power indicate that the modified GST starch is adequate for certain applications such as simple thickeners in liquid food products. If a denser gel matrix is desired from the GST starch such as for use as thermoplastic starch (TPS), and bioplastics,[38] then acid hydrolysis may not be the modification method of choice. In this case, other methods of modifications which might have more positive impacts on the gel strength and swelling power such as heat moisture treatment or cross-linking methods should be considered.[39,40]

The microstructure of the acid-modified GST starch as presented in Figure 1 show a rigid polygonal rock-like granule with sharp edges. After acid treatment the starch granules lost the smooth rounded granule structure. Some larger aggregates were found to retain the smooth rounded surface of the once ellipsoidal granules. The acid hydrolysis is a process where the acid donates its hydrogen ions to the oxygen atoms in the glycosidic bonds of the starch until the bonds are hydrolyzed. This process begins from the outermost surface of the amorphous granules and works its way through to the dense crystalline regions.[39] The acid modification method has the potential to excessively hydrolyze the glycosidic bonds, which could completely remove the amorphous regions, and at a high concentration of strong acids may even completely hydrolyze the dense crystalline regions causing structural defects to the granules and rapid amylopectin degradations.[39,40]

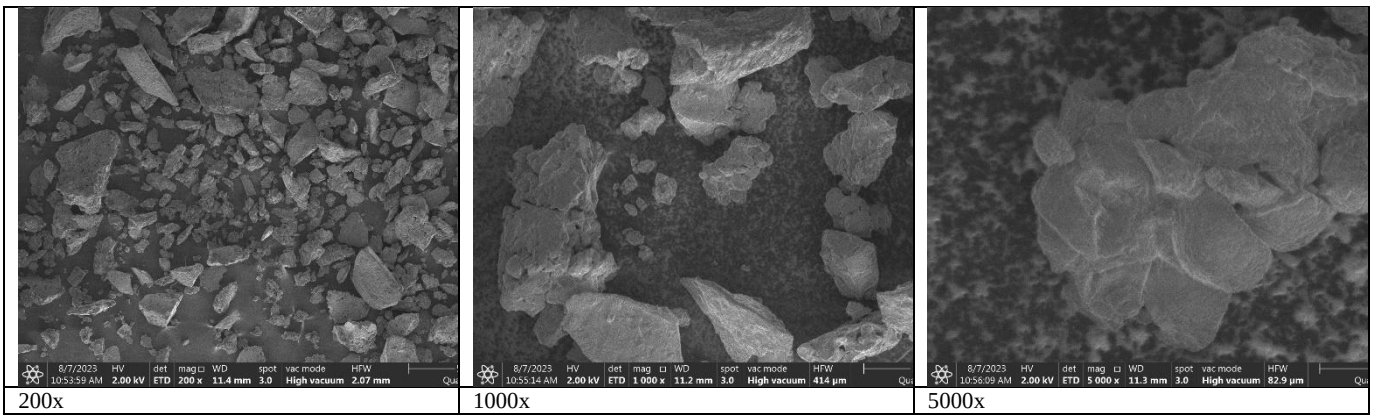
The FTIR analysis of the acid-modified GST starch showed a noticeable broad peak of hydroxyl groups of starch amylose at 3278.77 cm^{-1} (Figure 2a). This broad band peak is consistent with the findings on starch amylose spectra with vibrational frequencies of -OH groups generally below 3800 cm^{-1} in starch containing amylose and amylopectin as the main polymer backbone.[34] Another consistent occurrence is the peak at 2927.10 cm^{-1} (Figure 2a) which correspond to the C-H stretch vibrations of the hydrocarbon skeleton of the amylose and amylopectin structure. The FTIR spectrum of a study with acetylated wild cassava (*Ampelocissus Africana*) tuber starch also indicated major peaks corresponding to O-H and C-H stretching vibrations.[41] It is important to note that the acid modification of GST starch using citric acid followed by neutralization with sodium hydroxide resulted in the reduction of the GST starch construct to amylose unit $(C_6H_{10}O_5)_n$, production of sodium citrate ($C_6H_5Na_3O_7$) (Figure 2b) and sodium citrate dihydrate ($C_6H_5O_7Na_3.2H_2O$) (Figure 2c). This confirms the hydrolytic breakdown of the glycosidic bonds in the starch molecule.[34,39,40]

Conclusion

GST tuber (corm) is a rich source of carbohydrate, with a large proportion of it being starch. Native GST starch is a starch with a high amylose-amylopectin content, large smooth roundish granules and considerable gelling properties. The native GST starch that has gone through acid treatments or acid hydrolysis as shown in this study resulted in a modified GST starch with lower starch content and swelling power. The amylose and amylopectin of the modified starch was observed to be much lower than the initial amylose content and amylopectin content of the native starch. The physico-chemical performance of the modified GST starch makes it potentially suitable for use as thickeners in liquid foods. However, the utilizations of GST starch in conditions where a more rigid gel structure is required would require a different approach in modifying the native GST starch structure.

Table 1: Physico-chemical Properties of Modified Giant Swamp Taro (GST) Starch

Parameter	Value	Unit
Peak viscosity	326.67 ± 25.40	cP
Trough viscosity	217.67 ± 4.73	cP
Breakdown viscosity	401.00 ± 48.54	cP
Final viscosity	388.00 ± 142.94	cP
Set back viscosity	96.67 ± 41.68	cP
Peak time	4.91 ± 0.10	min
Pasting temperature	90.70 ± 2.11	C
Swelling power	4.22 ± 0.58	g/g
Solubility	5.73 ± 0.42	%
Expanding capability	15.9 ± 2.15	mL/gram
Amylose	13.02 ± 0.22	%
Amylopectin	33.84 ± 1.12	%
Total starch	46.86 ± 1.34	%



200x 1000x 5000x

Figure 1: Microstructure of Acid-Modified Giant Swamp Taro (GST) Starch

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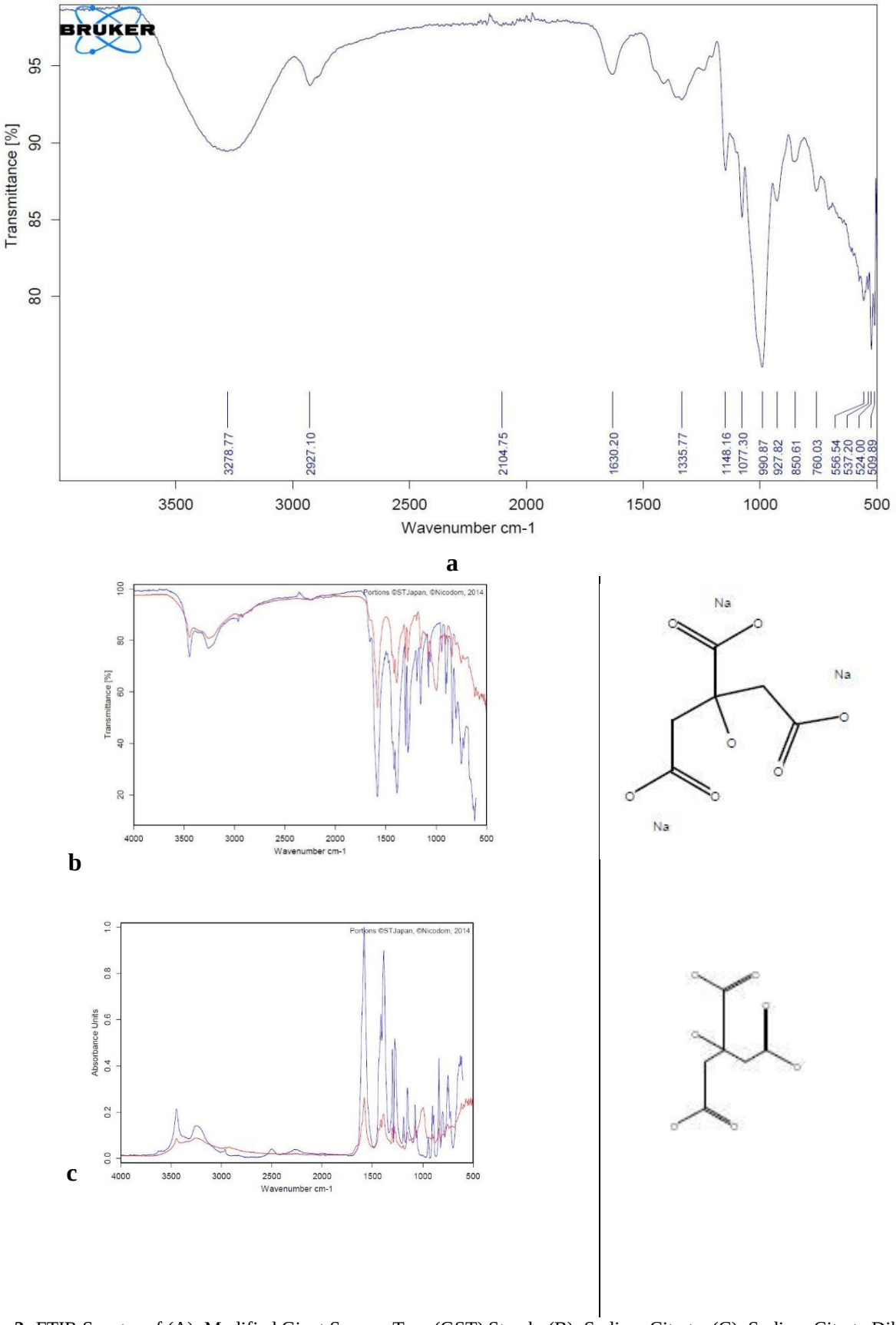

Figure 2: FTIR Spectra of (A): Modified Giant Swamp Taro (GST) Starch, (B): Sodium Citrate, (C): Sodium Citrate Dihydrate

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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