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Research Article

Purple Yam Flour (*Dioscorea alata Linn.*) Processing and Development of Instant Pudding Mix

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Abstract

Purple yam (*Dioscorea alata* Linn.) is an untapped natural nutrient-rich source of food ingredient available in Sri Lanka. The present study aimed to transform purple yam into shelf-stable flour and to develop a Ready-To-Cook (RTC) pudding mix. The edible portion of D. alata yams was blanched, dried, powdered and sifted to process the yam flour. Formula standardization of RTC pudding mix was performed through several preliminary trials and four types of pudding were chosen. The best formula was selected through trained sensorial panel and each prepared either full cream milk powder or coconut milk powder was screened out through ranking test on their appearance, color, aroma, texture, taste, aftertaste, and overall acceptability using a trained sensory panel. The physico-chemical and sensory properties were measured and analyzed statistically using SPSS software. The physical parameters include, moisture content, water activity and color whereas chemical parameters include the nutritional profile of the RTC pudding mix, prepared with cow's milk and coconut milk separately. The net yield of the purple yam flour was 26.80±0.75%. The TPC, TFC, anthocyanins and DPPH activity on fresh weight basis have demonstrated that high antioxidant contents and potential of the purple yam and developed puddings. Although the pudding prepared with coconut milk showed the highest acceptability, the pudding prepared with cow's milk had better functional properties. In conclusion, D. alata yam flour containing natural purple color pigments could be used as a viable ingredient for the development of nutrient-rich functional food products.

Keywords: Antioxidant; Functional food; Purple yam; RTC pudding

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Introduction

The yams (*Dioscorea spp.*) yield tubers, which are very important as starchy staple with highly nutritious and medicinal value [1]. Those species have undergone lesser scientific investigation with under-exploited potential for food security and health. The uses of nutritional potential, industrial use, storage procedures, characterization, natural dye, and as a health-promoting ingredient are still in research stage [2].

The purple yam, which is a member of the *Dioscorea alata* Linn. species, has long been used in traditional Chinese herbal treatment and as food. Although purple yam's physiological effects are well-established, there is a paucity of research on the plant's potential applications as a functional food ingredient [3].

The presence of antioxidant-rich anthocyanin compounds in the *Dioscorea* tuber is another fascinating feature [4]. The purple color is thought to be a sign of anthocyanin, an antioxidant that can counteract free radicals in the body [5]. Strong purple color shows high anthocyanin levels and high antioxidant activity with bioactive qualities [6]. Anthocyanin has colored compounds which are mostly red, blue and purple ease in free radicals, protecting cells from the toxic effects and contributing to disease prevention. Ageing, cancer, and other degenerative diseases can be avoided by using anthocyanin as an antioxidant to fight against free radicals. Further, blood sugar levels can be lowered and hypertension, antimutagenic, anticarcinogenic, and other properties can be prevented reasoning to the presence of anthocyanins' [7].

To promote the use of locally sourced agricultural products, it is crucial to diversify agricultural-based products through substitution of locally produced foods for imported ones. This will impact on achieving food security, fulfilling nutrition based on food resources and applying local wisdom by developing local food. Hence, purple yam can be used as a substitute in the preparation of starch-based foods to decrease the digestibility of starch and lower the GI of food products [3].

Furthermore, studies have been indicated that purple yam tubers possess high nutritional value and can be utilized in various food and nutraceutical industries [8]. It is important to exploring the potential of purple yam tubers as a natural source could have significant implications for food and nutraceutical industries. Additionally, purple yam tubers have potential to serve as a novel pigment source for food colorants [9]. Therefore, natural colorant offers an alternative to synthetic and insect-based dyes, which are considered potentially harmful to human health in addition to its antioxidant activity.

This study highlights the potential application of purple yam (*Dioscorea alata*) incorporation in food application of RTC pudding mix. Pudding belongs to the category of the dairy desserts, which is a milk-based starch paste and has a typical semisolid food texture [10,11]. Ready to cook powdered and packaged forms of the pudding samples are available in the market [12]. The formulation of the puddings is generally composed of milk, sugar, starch vanilla, and gum

[11]. The starch used in the pudding formulation has an important role for providing essential properties to the product, imparting body- and mouthfeel.

Milk is a common ingredient in pudding mixes [13]. The type of milk used can have a significant impact on the overall characteristics and compatibility. Traditional pudding preparations typically use cow's milk as the primary source of dairy. However, there has been a growing interest in alternative milk sources, such as coconut milk [14]. Coconut milk is a popular choice for individuals who follow a vegan or lactose-free diet. Therefore, the evaluation of compatibility of coconut milk as a substitute for cow's milk in pudding mixes has derived in this research. Carrageenan helps to maintain freeze/ thaw stability and proper eating characteristics in reduced-fat/ reduced-sugar ice cream. Manufacturers of pudding and gelled desserts depend on carrageenan to provide gelation, syneresis control and positive sensory attributes in their end applications [15,16]. Carrageenan gives the product the right texture, whereas starch gives it bulk and mouthfeel.

Materials and Methods

Preparation of Flour Samples

Ingredients

Fully mature, undamaged *D. alata* yams collected around Colombo region, Sri Lanka.

Method

D. alata yams were hand peeled, washed, and cut into thin slices and blanched at 100°C for 4 sec. Dried in an air drier (NESCO® Professional 600W 5-Tray Food Dehydrator, USA) at 40°C for 24h. The dried pieces were powdered using a laboratory scale grinder (Japan Mixer Grinder 600W, India) and sifted through a 300µm sieve. The flour samples were sealed and packed in airtight containers for further preparation.

Development of the Instant Pudding Mix

Ingredients

Full cream milk powder, coconut milk powder, purple yam flour, fine granule sugar, carrageenan and vanilla.

Method

An instant pudding mix pack per person was developed with appropriate portions of purple yam flour, cow's milk powder or coconut milk powder, ground sugar and carrageenan. The amounts of ingredients of the pudding mix and the preparation conditions were optimized through several trials by preparing the pudding as discussed in section in order to acquire the best sensory properties in the final product.

Preparation of the Pudding

Experimental design of 12 pudding samples in varying contents purple yam power (10g, 12g & 15g per pack), full cream milk powder (5g & 10g per pack), powdered sugar (10g & 15g per pack) and stabilizer (Carrageenan: 1g per pack) were used in initial trials. All the dry ingredients were added and dry mixed using a mixer. Pudding preparation requires the addition of water (around 120ml/ per pack) followed by cooking in an open pan till gets thicken. Water was added to the mix and cooked while mixing to 85-95°C for 6 min. Pudding mixture was poured into the mold and cooled at room temperature. Finally, pudding mixture was kept in the refrigerator at 4°C for 12h for setting. Dry mix can be served as an instant pudding mix which can be prepared easily by adding water and skim milk powder.

Sensory Evaluation

Sensory trials were done to optimize the amount of ingredients, water while cooking, amount of sweetener, amount of whole cow's milk powder, cooling time and temperature.

D. alata flour quantity and full cream quantity were selected from a general sensory evaluation using untrained sensory panel and *D. alata* Flour 10g & 12g: full cream or coconut milk powder 5g were finalized per pack (Approx. 25g-30g).

Further preference sensory test was carried using trained sensory panel to instant pudding mix in four different combinations with previously selected amount of *D. alata* Flour (10g & 12g), sugar (10g & 15g per pack), previously selected amount of full cream (5g per pack) and carragenaan (1g per pack). The four types of readymade pudding were subjected to evaluate their sensory attributes of appearance, color, aroma, texture, taste, after taste, and overall acceptability by 10 members of a trained preference test panel. Panelists were informed that they would be evaluating prepared pudding mixture, and they were presented with three code numbers (coded "000"). The order of presentation was also random. The panelists were asked to evaluate the samples according to their preferences. Samples were evaluated using a 9-point hedonic scale, with 1 for "dislike extremely" and 9 for "like extremely".

Selected formula was further subjected to a sensory acceptance test for the appearance, colour, manual thickness, melting, creaminess, mouthfeel, flavour, sweetness and floury taste presenting coconut milk and cow's milk based.

Physicochemical Analysis

Physical parameters

Moisture content

Moisture content was determined by oven dried methods AOAC 931.04 [17].

Water activity

Water activity of each sample was evaluated using a water activity meter (AQUALAB® 4TE, USA).

Colour

Using the CIE LAB color space approach as outlined by Brainard [18], the color of the samples was measured using a reflectance Chroma-meter (KONICA MINOLTA CR-A12, Japan) based on the L* (brightness/whiteness), a* (redness/greenness), and b* (yellowness/ blueness) values.

Proximate Analysis

The proximate analysis was carried out to using AOAC [17], to determine the moisture content AOAC 931.04, crude protein content AOAC 920.87, crude fibre AOAC 978.10, crude fat content AOAC 922.06 and ash content AOAC 923.03 of each powdered sample. The results were expressed on Dry Weight (DW) basis and all measurements were performed in triplicates.

The determination of the mineral (potassium, calcium, iron, zinc, magnesium, and copper) content was carried out according to the AOAC, 999.10 [17], microwave digestion followed by ICP-MS detection

The energy value and carbohydrate content were computed in following way.

Total carbohydrate = 100% − (moisture % + crude protein % + crude fat % + crude fibre % + ash %)

Energy (kcal per 100 g) = (crude protein × 4) + (carbohydrate × 4) + (crude fat × 9)

Antioxidant Analysis of Total Polyphenolic and Flavonoid Content

To determine the antioxidant levels of Total Polyphenolic Content (TPC) and Total Flavonoid Content (TFC), sample extractions were conducted following the method outlined by Abeysekera et al. [19].

Initially, 5.00 grams of sample of processed yam flour and instant pudding mixes were each subjected to overnight shaking using an orbital shaker (SSLI 11844, Stuart, Sweden) operating at 110 revolutions per min (rpm) at room temperature (28±2°C) with four times the sample weight of absolute methanol. Subsequently, the extracts were filtered, and ethanol was evaporated using a rotary evaporator. The resulting extracts were individually collected into Eppendorf tubes, labeled, and stored at temperatures below 20°C.

Total Phenolic Content

The TPC analysis for purple yam flour, full cream cow's and coconut milk incorporated dry powdered pudding mix was performed following the protocol described by Singleton et al. [20]. In summary, each sample extract was diluted to a concentration of 2mg/ml using distilled water. In a 96-well microplate, 20μL of the sample, 110μL of 10-fold diluted Folin-Ciocalteu reagent, and 70μL of 10% sodium carbonate (Na_2CO_3) solution were combined. The absorbance was then measured at 765nm after 30 minutes of incubation at 25±2°C using a microplate reader (Spectra Max Plus 384, Molecular Devices, USA) equipped with Soft max Pro 5.2 v software. Gallic acid was used as the standard, and TPC was expressed as milligrams of gallic acid equivalents per gram (mg/g GAE) of sample on wet basis.

Total Flavonoid Content

The TFC analysis for purple yam flour, full cream cow's and coconut milk incorporated dry powdered pudding mix was conducted according to the method outlined by Pourmorad et al. [21]. In brief, each extract was diluted to a concentration of 2mg/ml in methanol. Then, 100μL of the diluted sample and 100μL of 2% aluminum chloride solution were added to a 96-well microplate. After 10 minutes of incubation at 25±2°C, the absorbance was measured at 415nm using the microplate reader equipped with Soft max Pro 5.2 v software. Quercetin was used as the standard, and the TFC results were reported as milligrams of quercetin equivalents per gram (mg QE/g) of sample on wet weight basis.

Total Monomeric Anthocyanin Content

The pH-differential method was used in anthocyanin content determination as discussed by Giusti and Wrolstad [22].

The sample was dissolved in two distinct buffer solutions in pre-determined concentrations, each containing 1mL of buffer. The sample was first diluted in Potassium chloride buffer (pH 1) until it had a volume of 10mL, and then it was diluted in Sodium acetate buffer (pH 4.5). After being left to stand for 15 minutes, absorbance measurements were taken via spectrophotometry, using each wavelength read at a length of 520 and 700nm.

Anthocyanin content (mg/100g) = A x MW x DF x V x 100%

ε x *L*x W

Where: A = Absorbance [A520 - A700] pH 1 - [A520 - A700] pH 4.5

ε = extinction coefficient (Cyanidin-3-glycoside: 26900 L / mol cm),

L = width of cuvette (1cm)

MW = molecular weight of cyaniding-3-glycoside 448.8 g / mol, DF = factor of dilution, W = Sample weight, V = Volume of the extract

DPPH Assay

Purple yam flour with full cream cow's and coconut milk powder incorporated dried pudding mix were tested for their ability to scavenge DPPH radicals using the procedure outlined by Blois [23].

To summarize the procedure, 50μL of sample in a series of concentrations (1, 0.5, 0.25, 0.125, 0.0625mg/ml), 90μL of methanol and 60μL of DPPH radical (20mg/100ml) were combined in a well and incubated at 25±2°C for 10 minutes. Absorbance was measured at 517nm using a micro plate reader (Spectra Max Plus 384, Molecular Devices, USA). Trolox was used as the standard in a standard curve, concentrations of 1.5625, 3.125, 6.25, 12.5, 25, and 50μg/mL for the dosage response experiments. A set of high activity extracts were tested. A graph showing the activity vs. extract/standard concentration was used to compute the extracts' IC_{50} values of the sample series and the standard series.

DPPH radical scavenging activity (%) = $[(A_c - A_s) / A_c] * 100$

where, A_c is the absorbance of the control and A_s is the absorbance of the sample.

Statistical Analysis

Statistical analysis data from sensory was performed using the non-parametric Kruskal-Wallis test for multiple comparisons of mean differences to assess the significance of each variable (α=0.05), followed by analysis with SPSS software. Other data was performed by one-way analysis of variance (ANOVA) and significant differences between the results were reported as at 95% confidence level ($P<0.05$).

Results and Discussion

This study has focused on the multifaceted attributes of purple yam, shedding light on its nutritional composition, functional properties, and potential roles in both traditional and innovative food formulations. Through an extensive review of literature, instant pudding preparations and investigations has reported in several research studies including Dogan et al. [10], Kristanti and Herminiati [15,24]. Larief et al. [4], elucidate the significant implications of incorporating purple yam into food products, ranging from its capacity to enhance nutritional profiles and antioxidant activity to its role as a natural pigment source and antioxidant powerhouse.

Evaluation of Sensory Properties

Results of analysis of sensory properties of the purple yam pudding are presented in table 1.

Sample Code	Appearance	Colour	Aroma	Texture	Taste	Overall
B	6.8[b]	6.1[a]	5.9[a]	6.6[b]	7[a]	6.8[a]
C	6.7[b]	6.4[a]	6.2[a]	6.9[bc]	6.5[a]	6.7[a]
D	7.1[b]	6.4[a]	6.1[a]	7.6[c]	7.4[a]	7.4[b]
E	5.5[a]	5.5[a]	5.4[a]	4.9[a]	7[a]	6.5[a]

Table 1: Results of the sensory evaluation of prepared pudding with cow's milk.

Note: Data represent as mean ranks (n=10).

According to the sensory evaluation results, values for color, aroma, taste did not show any significant difference ($p>0.05$) among the samples. There was a significant difference ($p<0.05$) in sample E for the appearance among other samples. For the texture samples B, D, E was significantly different ($p<0.05$) from each other, while sample C was not significant difference ($p>0.05$) to B and D samples. Considering the overall acceptance of the sample D being significantly higher scores ($p<0.05$) than other samples, it was concluded to be the best sensory accepted formula among the other samples.

Although the sample D being the highest scored with overall performance, 91% comments were received it that had high sugar taste. Therefore, product was reformulated with 12% sugar. However, as a general suggestion through the trained panel for sensory evaluation another adjustment was carried out for the sensory acceptance after selecting the best formula from the initial sensory test. Suggestion made to substitute the milk source to a plant based form to claim the product as a vegan dessert. Therefore, as considering the best availability and general use of Sri Lankan population coconut milk was substituted in place for the cow's milk in the acceptance test. Results are demonstrated in table 2.

	Appearance	Colour	Manual Thickness	Melting	Creaminess	Mouthfeel	Flavour	Sweetness	Floury Taste
Cow's Milk	7.55	7.10	6.35	7.45	7.00	7.35	6.90	7.05	6.63
Coconut Milk	8.00	7.65	6.50	7.50	8.00	7.55	7.00	7.10	6.45

Table 2: Results of the mean sensory test of prepared pudding with cow's milk and coconut milk.

Note: Data represent as mean ranks (n=10)

No significant difference ($p>0.05$) was observed the means of the two puddings based on cow's milk and coconut milk during the acceptance sensory test.

Physical characteristics of Purple Yam Flour

D. alata yield was calculated in yam powder processing. Flesh was uncovered by removing the outer covering and soil particles retained with the yam. The presence of saponins (plant alkaloids) in the mucus and calcium oxalate crystals in yams is thought to be the cause of the itching [1]. Acrid tubers from several yam species include various anti-nutritional elements linked to skin irritation and inflammation of the throat and buccal cavity following ingestion [25]. The weight (g) of powder per weight of yam (22.5±0.52%) was used to compute the gross yield, whereas the weight (g) of powder (per peeled or sliced yam was used to calculate the net yield (26.8±0.75%).

Table 3 presents the physical properties of the purple yam powder.

Type of Test	Purple Yam Powder
Moisture Content (%)	3.98±0.30
Water Activity (a_w)	0.30±0.01
Color	
L	36.10±1.0
a	3.28±0.10
b	0.16±0.00

Table 3: Physical parameter analysis for the purple yam powder.

Note: Results were presented in Mean ± SD of three replicates

The first food preservation technology likely ever used by humans is the drying technique. Food can be preserved by drying, which involves taking the water out of the food. Eliminating moisture stops the growth and procreation of the decay-causing bacteria and lessens the moisture-mediated degradation processes. Drying significantly reduces a product's weight and volume, which lowers the cost of packaging, storing, and shipping [26]. Low moisture content is not a guarantee for food stability; it is merely an indicator. Food preservation greatly depends on the availability of moisture for microbial development, often known as "Water Activity" (a_w). The range of water activity is 0 to 1.00, with a lower number indicating greater difficulty for microorganisms to thrive on a given food. The product is most stable in terms of lipid oxidation, non-enzymatic browning, enzyme activity, and, of course, the different microbiological parameters when the a_w value is 0.3. The likelihood of the food product deteriorating rises as aw increase [27].

Chemical Characteristics of Pudding Powder Mix and Pudding (with Coconut Milk, Dairy Milk)

Proximate composition of pudding mix, prepared pudding with dairy milk and prepared pudding with coconut milk is descriptively detailed in the table 4.

Parameter	Formula		
	Pudding Powder Mix	Prepared Pudding with Dairy Milk	Prepared Pudding with Coconut Milk
Nutrient			
Energy (Kcal)	433.94±0.09[b]	137.04±0.09[ab]	103.17±6.35[a]
Moisture Content (%)	3.98±0.08[a]	65.30±0.80[b]	75.67±2.11[b]
Carbohydrate (%)	84.33±0.27[b]	31.15±1.00[ab]	20.01±1.32[a]
Crude Protein (%)	4.27±0.09[b]	2.55±0.15[ab]	0.91±0.07[a]
Crude Fat (%)	0.42±0.08[ab]	0.25±0.12[a]	2.18±0.13[b]
Crude Fiber (%)	0.88±0.03[a]	0.36±0.04[a]	0.25±0.16[a]
Crude Ash (%)	1.73±0.08[ab]	0.63±0.04[a]	2.01±0.11[b]

Table 4: Proximate analysis for the powder mix and pudding (with coconut milk, Dairy milk).

Note: Results were presented in Mean ± SD of three replicates of nutrients.

There was no significant difference ($p>0.05$) between prepared pudding with dairy milk and prepared pudding with coconut milk for moisture content, carbohydrate content, fiber content and protein content, however a significant difference ($p<0.05$) was observed for the parameters of fat and ash percentages between two. The addition of coconut milk replacing dairy milk has risen up the fat percentage of the sample owing to that coconut milk has comparatively higher percentage of fat [28]. Similarly, the increased mineral content may account for the higher ash concentration of coconut milk [29]. Coconut milk is particularly notable for its Medium-Chain Triglycerides (MCTs), a type of fat that is metabolized differently from other fats and is believed to offer health benefits such as improved energy levels and weight management [30-33].

Between prepared pudding with dairy milk and prepared pudding with coconut milk examined minerals were not significantly ($p>0.05$) deviated (Table 5). Numerous studies have examined the mineral composition of dairy milk and coconut milk, suggesting that the two types of milk provide comparable amounts of essential minerals despite their differing sources [30,34,35]. Cow's milk, is renowned for its rich nutritional profile, serving as an excellent source of calcium, a crucial mineral for bone health. Dairy milk also contains other important minerals such as potassium, magnesium, and phosphorus, which play vital roles in maintaining healthy blood pressure, muscle function, and overall metabolic processes. In contrast, coconut milk, extracted from the grated meat of mature coconuts, offers a distinct yet equally beneficial nutritional profile. Similar to cow's milk, coconut milk contains calcium, potassium, and magnesium, albeit in varying concentrations [30,32,36].

Parameter	Formula		
	Pudding Powder Mix	Prepared Pudding with Dairy Milk	Prepared Pudding with Coconut Milk
Minerals			
Potassium (%)	0.6±0.1b	0.23±0.01^{a}	0.42±0.06^{ab}
Calcium (%)	0.07±0.04^{b}	0.08±0.00^{ab}	0.17±0.02^{a}
Magnesium (mg/kg)	112.435±8.34^{a}	124.235±9.1^{a}	118.335±7.42^{a}
Sodium (%)	0.035±0.02^{ab}	0.058±0.02^{b}	0.047±0.01^{a}
Zinc (mg/kg)	2.342±0.53^{a}	3.086±0.56^{b}	2.714±0.74^{ab}
Manganese (%)	0.252±0.00^{a}	0.252±0.01^{a}	0.255±0.05^{a}
Iron (%)	nd	nd	nd

Table 5: Mineral content analysis for the powder mix and pudding (with coconut milk, Dairy milk).

Note: Results were presented in Mean ± SD of three replicates of minerals on dry weight basis (db)

Nd- Not Detected

Antioxidant content and capacity analysis for the powder mix and prepared pudding (with coconut milk, Dairy milk) were shown in table 6.

Total phenolic content and total flavonoid content significantly ($p<0.05$) deviate among the pudding dry powder mix and the prepared pudding with coconut milk while, there was no significant difference ($p>0.05$) between two prepared puddings. DPPH (2,2-diphenyl-1-picrylhydrazyl) analysis is a widely used method for evaluating the antioxidant capacity of various substances, including natural extracts, food items, and synthetic compounds [37,38]. Pudding powder mix contain significantly high amount of antioxidant capacity compared to prepared puddings along with DPPH test results. Anthocyanin, a class of water-soluble pigments found in various plant-based foods, have garnered significant attention in the scientific community due to their potential health benefits and nutritional value [39,40]. Anthocyanin basically was resulted due to the purple yam for the pudding mix as well as the prepared puddings. Coconut milk incorporated pudding showed a significant ($p<0.05$) lower content of anthocyanin than the pudding mix.

Parameter	Formula		
	Pudding Powder Mix	Prepared Pudding with Dairy Milk	Prepared Pudding with Coconut Milk
Bioactive			
Total Phenolic Content (mg GAE/g)	3.78±0.10b	3.15±0.01ab	1.78±0.04a
Total Flavonoid Content (mg QE/1g)	2.17±0.32b	1.81±0.37ab	0.96±0.14a
DPPH (mgTE/g)	11.73±0.65b	1.41±0.10a	1.36±0.02a
Anthocyanin (c-3-gE mg/g)	0.33±0.01b	0.13±0.00ab	0.01±0.00a

Table 6: Antioxidant content and capacity analysis for the powder mix and pudding (with coconut milk, Dairy milk).

Note: Results were presented in Mean ± SD of three replicates of bio active potential on fresh weight basis (FW).

Conclusion

In conclusion, *D. alata* yam flour proves to be an exceptional ingredient for the creation of nutritious food products, offering not only significant health benefits but also a distinctive natural purple pigment. The incorporation of this flour in food formulations enhances the antioxidant content and capacity, contributing to improved nutritional profiles. The promising concentrations of antioxidants in these formulated treatments underscore the potential of *D. alata* yam flour as a valuable component in the development of health-promoting, visually appealing food products. As such, it stands as a versatile and beneficial addition to modern dietary innovations.

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Conflict of Interest

No conflict of interest is disclosed by the authors.

Data Availability Statement

Data could be provided with reasonable request through the corresponding author.

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