Optimisation of pretreatment and drying temperature influenced the techno-functional and thermal behaviour of cassava flour from two landraces.

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Abstract

Cassava flour (CF), upon absorption of water and in the presence of heat, can be constituted into dough form for use in different food formulations. This work investigated the effect of calcium chloride and citric acid pre-treatments and their interaction with optimised drying temperatures on the functional, hydration and thermal properties of CF from red and white cassava landraces. Response surface methodology was employed in determining the linear, interactive and quadratic effect of varying concentrations of pre-treatment (0.6 – 3.4% w/v) and drying temperature (45 – 74 ºC) on CF. Differential scanning calorimetry showed an onset gelatinisation temperature of between 60.32 and 120.30 ºC, peak temperature of 71.85 - 126.84 ºC, conclusion temperature of 93.31 - 140.98 ºC and enthalpy of gelatinisation of 0.14 - 54.95 J/g in all CF samples. Citric acid pre-treatment exhibited a significantly decreasing effect on the water holding capacity when compared to calcium chloride. Drying temperatures significantly influenced the bulk densities of CF, while the loose bulk density increased with drying temperature. An increase in enthalpy of gelatinisation in CF due to pre-treatment indicates that more energy is required for gelatinisation to occur. Therefore, the processing conditions confer more stability on CF in the presence of heat and water.

Keywords: Cassava landraces, flour, Pretreatment, Drying temperatures, Thermal behaviour, Response surface methodology

1 Introduction

Cassava flour (CF) is a major product of one of the world's most important staple roots (*Manihot esculenta* Crantz), also known as tapioca or manioc [1]. The agronomic trait of the root combined with its climate resilience status guarantees low cost and all-year availability of the flour [2-4]. These advantageous characteristics have made CF currently explored as a convenient alternative to wheat in the development of gluten-free and fortified food products [5, 6]. Cassava flour can be processed into popular flour-based food products such as bread [7, 8], biscuits [9, 10], noodles [11] and other confectionery [12] both as a composite and the base flour. A common processing step in CF utilisation is the incorporation of water into the flour structure and the application of heat during dough formation for various food products. High hydration increases the swelling and gelatinisation ability of starch granules [13]. Thus, the ability of flour to take up water and its behaviour during heat application is dependent on the nature of starch in the flour as well as treatment conditions during processing [2, 14, 15].

Ayetigbo et al. [16] opined that flours exhibit water-holding capacity by their ability to bind with water at hydrophilic sites. Omowaye-Taiwo et al. [17] showed that this characteristic is highly desirable in improving the flour consistency and yield.

Although CF consists mostly of starch, its non-starch components also influence its thermal behaviour [16]. Zhu [4] noted that thermal properties such as gelatinisation are associated with the energy required for the occurrence of an irreversible change in starch structure due to the application of water and heat. Lu et al. [10], who investigated the effects of xanthan gum and inulin on the thermal properties of CF and the quality of its developed short biscuits, observed that the addition of inulin improved starch gelatinisation, resulting in decreased hardness and brittleness of biscuits processed from CF. Based on elaborating the utilisation of CF in gluten-free pasta formulation, Ramírez et al. [6] conducted a study to determine the optimal hot-air drying conditions for CF. The authors demonstrated that the operational conditions needed to minimise the hot-air drying time (57 min) to produce CF with higher water holding capacity was 57 °C at 3 m/s. In another study by Dudu et al. [2], it was reported that thermal stability and gelatinisation enthalpy of CF increased and decreased, respectively, at optimal steam-heat-moisture treatments. Furthermore, Falade et al. [18] revealed that annealing reduced the WHC of CF, while citric acid cross-linking increased its packed bulk density (PBD) and loose bulk density (LBD).

A few studies [19, 20] have reported the effect of chemical pretreatment on the functional and thermal properties of cassava starch. However, studies on the effect of chemical pre-treatment on the WHC and thermal properties of CF are scarce. Wholistically, there is a need to determine the influence of non-starch components of CF on its bulk density, WHC and thermal properties when subjected to pretreatment. Previous work by Udoro et al. [21] indicated no adverse effect on the bulk density of pre-treated CF, while Omolola et al. [22] showed no significant effect on the thermal properties of forced-air oven-dried CF.

Another study by Nemaungani et al. [23] demonstrated improved colour quality of solar-dried CF due to calcium chloride and citric acid pre-treatment. Therefore, this work comparatively determined the influence of citric acid and calcium chloride pre-treatment at varying concentrations and drying temperatures on the WHC, thermal properties and bulk densities of CF using response surface methodology.

2 Materials and Methods

2.1 Preparation of Cassava Root

Cassava roots from two landraces (white and red) were obtained from the Institute of Tropical and Subtropical Crops - Agricultural Research Council (ITSC-ARC), Levubu, Limpopo Province, South Africa (22.946º S 30.485º E) and transported to the Department of Food Science and Technology, University of Venda, South Africa for further analysis. Relevant agronomic practices and guidelines were followed during planting, harvesting and collection of the harvested cassava roots at the ITSC-ARC Levubu, South Africa farm station. Similarly, permission was obtained from the ITSC-ARC Levubu farm station before collection of the harvested cassava root samples. The harvested roots, fourteen months after planting, were sorted and washed with tap water to remove adhering dirt and avoid contamination during processing. Within 24 h after harvest, processing commenced to ascertain that the roots were in a fresh state before the onset of deterioration [24].

2.2 Experimental Design

A central composite design was used in this study as shown in Table 1. Factored into the design of this experiment are the concentration of chemical pre-treatments (citric acid and calcium chloride separately at 1-3% w/v) and drying temperature (50 – 70 ºC) as independent variables in CF processing. While WHC, onset gelatinisation temperature (To), peak gelatinisation temperature (Tp), conclusion gelatinisation temperature (Tc), gelatinisation enthalpy (ΔH), packed bulk density (PBD), loose bulk density (LBD), and difference in bulk density (ΔBD) were the dependent processing variables. Design Expert (DE) software Version 11 (Stat Ease Inc., Minneapolis, USA) was used in generating the experimental conditions for processing the flour samples The four experimental groups obtained are: citric acid pre-treated flour from the red landrace (CAR), calcium chloride pre-treated flour from the red landrace (CCR), citric acid pre-treated flour from the white landrace (CAW), and calcium chloride pre-treated flour from the white landrace (CCW).

Table 1. Levels of independent variables used for central composite design

Pretreatment	Code	DT	COP (g/100 mL)
Citric acid	-1	50	1
	0	60	2
	1	70	3
Calcium chloride	-1	50	1
	0	60	2
	1	70	3

-1- upper limit; 0- central point; 1- lower limit; DT- Drying temperature, COP- Concentration of pre-treatment. NB: Two pre-treatments (citric acid and calcium chloride) were used separately

2.3 Processing of Pre-treated Cassava Flour

The method described by the Federal Institute of Industrial Research [25] was employed, with modifications, in processing the flour. Modifications applied in this study are the application of different concentrations of chemical pre-treatment and drying temperatures for each experimental run as generated by the central composite design. The washed roots were peeled and chopped manually with a knife. Fresh cassava chips (1 ± 0.5 cm thickness) were pre-treated by steeping them in the chemical (citric acid and calcium chloride) solution for about 20 min, after which they were dried in the oven (Ecotherm, 240L digital oven, South Africa) at different temperatures (50 – 70 ºC) for 24 h. Milling of dried chips was done with the aid of a dry milling machine (Polymix PX-MFC 90D, Switzerland). Milled flour was sieved through a 500 μm sized aperture sieve and stored in sample bags made of paper material at room temperature till further analysis. Two control samples were processed, one from each landrace, using the same procedure as the experimental samples except for pre-treatment and drying temperature, which was 60 ºC. The control samples served as reference samples within a landrace during statistical analysis.

2.4 Determination of Water Holding Capacity of Cassava Flour

The method described by Onwuka and Onwuka [26] was used in determining the water holding capacity (WHC) of CF. Approximately 1 g of the flour sample was weighed into a 15 mL centrifuge tube and suspended in 10 mL of water. It was shaken on a vortex for 1 min at room temperature. The sample was allowed to stand for 30 min and centrifuged at 1200 g for 30 min. The volume of free water was read directly from the centrifuge tube. The WHC was calculated using Equation 1.

$$WHC\% = \frac{Amount\ of\ water\ added - free\ water}{weight\ of\ the\ sample\ (dry\ bases)} \times density\ of\ water \times 100 \qquad (1)$$

2.5 Determination of Thermal Properties of Cassava Flour

The onset gelatinisation temperature (To), peak gelatinisation temperature (Tp), conclusion gelatinisation temperature (Tc), and gelatinisation enthalpy (ΔH) of CF were determined with a differential scanning calorimeter (DSC 4000, Perkin-Elmer, Shelton, CT, USA) as described by Anyasi et al. [27]. To approximately 10 mg of CF, 10 μL of distilled water was added in an aluminum pan. The pan was hermetically sealed with a crimper press (Perkin-

Elmer, Shelton, CT, USA). The sample was refrigerated (4 ºC) overnight to equilibrate it. Afterwards, the sample was heated at 10 ºC/min from 20 – 140 °C, and thermal properties were extrapolated from the curves with the aid of Pyris software (Perkin Elmer Shelton, CT, USA).

2.6 Determination of Bulk Density of Cassava Flour

Bulk density was determined as described by Maninder et al. [28]. The flour samples were gently filled into 10 mL graduated cylinders. The bottom of each cylinder was tapped gently on a laboratory bench several times until the diminution of the sample level ceased after filling to the 10 mL mark to determine the packed bulk density (PBD). For the loose bulk density (LBD), the bottom of the cylinder was not tapped. Bulk density was calculated as weight per unit volume of sample (g/mL) as expressed in Equation 2. The difference in the bulk densities (ΔBD) was calculated as shown in Equation 3.

$$Bulk\ density = \frac{weight\ of\ sample\ (g)}{volume\ of\ sample\ (ml)} \quad (2)$$

$$\Delta \mathrm{BD} = PBD - LBD \quad (3)$$

2.7 Statistical Analysis and Optimisation

The DE software version 11 was employed in the analysis of variance (ANOVA) of the linear, interactive, and quadratic effects of the model parameters on CF properties. Regression models, coefficient of determination (R^2), p-values, F-values, response surface plots, contour plots, and optimisation conditions were generated by the software for all four experimental groups. Statistical data analysis was done with the IBM SPSS Statistics software Version 25 (IBM Corp., NY, USA) to ascertain the significant difference between means of the experimental and reference/control sample within each experimental group at a

95% confidence level. One-way ANOVA and separation of means were done using Duncan Multiple Range Test.

3 Results and Discussion

3.1 Effect of Pre-treatment and Drying Temperature on Water Holding Capacity of Cassava Flour

Water holding capacity is a measure of the concentration of water a material can retain, and it is dependent on the nature of the material [29]. This property is important for the reconstitution of flour to dough. It is highly desirable in food systems to improve the yield and consistency of the product [17]. Analysis of variance of the means showed significant variance between samples of the experimental runs. However, the trend of variance as a result of the processing conditions was different in all experimental groups (Table 2). This is an indication that the landrace and processing conditions have different effects on WHC of CF. The WHC of pre-treated CF processed from the red landrace ranged from 41.95% (CAR/60DT/2COP) to 99.37% (CCR/70DT/3COP). Assessment of the influence of processing on WHC of CF from the white landrace shows that the lowest (45.54%) and highest (111.45%) values were exhibited by CAW/50DT/3COP and CCW/70DT/3COP, respectively. Agunbiade et al. [7] reported a value of 62.67% for CF, which is within the range reported in this study. This value (101%) obtained by Odey and Lee [30] for non-fermented CF is similar to the values (41.95 – 111.45%) reported in this study.

It was observed that CF pre-treated with calcium chloride had higher WHC than those pre-treated with citric acid for all corresponding experimental runs. A similar trend was observed in the white landrace for most experimental runs between calcium chloride pre-treated flour (CCW) and its corresponding citric acid pre-treated flour (CAW). Higher WHC is attributed to the loose structure of the starch polymer, while a low value indicates the compactness of the molecular structure [31].

Table 2. ANOVA of water holding capacity of cassava flour

Exp Run	DT (^oC)	COP (%w/v)	WHC (%) CAR	CCR	CAW	CCW
1	50	1	$59.18^{c} \pm 6.54$	$66.71^{de} \pm 8.00$	$61.00^{cde} \pm 6.05$	$68.91^{bcd} \pm 3.43$
2	70	1	$74.50^{ab} \pm 1.46$	$75.52^{c} \pm 4.24$	$73.32^{abc} \pm 1.34$	$84.90^{b} \pm 11.72$
3	50	3	$49.59^{cd} \pm 4.43$	$57.16^{f} \pm 4.51$	$45.54^{e} \pm 4.01$	$62.07^{bcd} \pm 9.38$
4	70	3	$62.46^{bc} \pm 14.88$	$99.37^{a} \pm 5.26$	$76.64^{abc} \pm 6.86$	$111.45^{a} \pm 23.18$
5	45	2	$85.97^{a} \pm 9.48$	$94.04^{a} \pm 4.31$	$76.23^{abc} \pm 16.25$	$59.68^{cd} \pm 8.66$
6	74	2	$60.41^{c} \pm 6.32$	$67.31^{cde} \pm 3.06$	$72.93^{abc} \pm 7.19$	$69.10^{bcd} \pm 15.36$
7	60	0.6	$82.00^{a} \pm 8.96$	$98.06^{a} \pm 5.30$	$86.47^{a} \pm 12.30$	$105.75^{a} \pm 16.93$
8	60	3.4	$52.17^{cd} \pm 5.54$	$59.71^{ef} \pm 2.80$	$52.01^{de} \pm 5.85$	$56.45^{d} \pm 2.15$
9	60	2	$41.95^{d} \pm 2.45$	$72.15^{cd} \pm 3.89$	$83.69^{ab} \pm 20.13$	$80.57^{bc} \pm 5.51$
Control	60	0	$85.89^{a} \pm 2.67$	$85.89^{b} \pm 2.67$	$66.37^{bcd} \pm 6.54$	$66.37^{bcd} \pm 6.54$

Exp Run- Experimental run; DT- Drying temperature, COP- Concentration of pre-treatment; WHC- water holding capacity; CAR- cassava flour from red landrace pre-treated with citric acid; CCR- cassava flour from red landrace pre-treated with calcium chloride; CAW- cassava flour from white landrace pre-treated with citric acid; CCW- cassava flour from white landrace pre-treated with calcium chloride Means in the same column with different superscripts are significantly different ($p < 0.05$).

This implies that, in comparison with calcium chloride, citric acid may have a more compacting effect on the molecular structure of starch in the flour samples under the conditions investigated, resulting in a decrease in the WHC of the CAR samples. This observation is buttressed by the ANOVA of model parameters concerning the WHC of the samples (Table 3) generated by RSM.

Table 3. ANOVA of the effect of model parameters on water holding capacity of cassava flour

Source	CAR		CCR		CAW		CCW	
	F-value	p-value	F-value	p-value	F-value	p-value	F-value	p-value
Model	4.47	0.04*	0.50	0.77	3.69	0.06	1.56	0.29
A-DT	0.08	0.79	0.09	0.77	2.27	0.18	3.60	0.09
B-COP	4.94	0.06	0.86	0.38	5.61	0.04*	1.46	0.27
AB	0.0146	0.90	1.20	0.31	1.07	0.34	1.30	0.29
A^2	12.19	0.01*	0.27	0.62	3.56	0.10	1.10	0.33
B^2	7.28	0.03*	0.13	0.73	7.09	0.03*	0.21	0.66

*- significant at ($p > 0.05$); DT- Drying temperature; B- concentration of pre-treatment; A^2-quadractic effect of DT; B^2-quadractic effect of COP; AB- interaction effect of A and B. ER-Experimental run; DT- Drying temperature; COP- Concentration of pre-treatment; CAR-cassava flour from red landrace pre-treated with citric acid; CCR- cassava flour from red landrace pre-treated with calcium chloride; CAW- cassava flour from white landrace pre-treated with citric acid; CCW- cassava flour from white landrace pre-treated with calcium chloride.

In this study, a factor common to CAR and CAW is that the quadratic effect of B^2 (COP) is a significant model term with p-values less than 0.05. P-values less than 0.05 indicate model terms are significant. For CCR and CCW, all model parameters had p-values higher than 0.05, which implies that those model parameters did not significantly affect the WHC of the samples. The decreasing effect of citric acid and the significance of its quadratic effect on the WHC of flour from both landraces is further buttressed by the coefficient of determination (R^2) values shown in Table 4, displayed alongside regression models relating WHC and model parameters. The coefficient of determination (R^2) of the citric acid-treated experimental group (CAR – 0.7614, CAW – 0.7251) was higher than that of the calcium chloride-treated experimental group (CCR – 0.2640, CCW – 0.5274).

The WHC of flour is a useful indicator of whether the protein can be incorporated within aqueous food formulations, especially those involving dough handling [32]. Relatively high WHC allows flour to absorb water and swell for improved consistency in food.

Table 4. Regression models relating water holding capacity and model parameters for cassava flour

Experimental Group	Models	R^2
CAR	$41.95 – 0.9947A – 7.98B – 0.6125AB + 13.44A^2 + 10.39B^2$	0.7614
CCR	$72.15 + 1.65A - 4.99B + 8.35AB + 2.99A^2 + 2.10B^2$	0.2640
CAW	$83.69 + 4.84A - 7.61B + 4.70AB -6.50A^2 - 9.17B^2$	0.7251
CCW	$80.57+ 9.84A- 6.25B + 8.35AB- 5.82A^2+ 2.54B^2$	0.5274

R^2- Coefficient of determination; WHC- Water holding capacity; CAR- Cassava flour from red landrace pre-treated with citric acid; CCR- Cassava flour from red landrace pre-treated with calcium chloride; CAW- Cassava flour from white landrace pre-treated with citric acid; CCW- Cassava flour from white landrace pre-treated with calcium chloride.

It is an advantageous baking quality because it allows for increase in the moisture content of the dough during baking, resulting in improved handling characteristics and maintenance of bread freshness [33]. Conversely, lower water absorption capacity would be desirable for formulating thinner porridges for infants [34]. Hence, optimisation was conducted to minimise and maximise the WHC (Table 5) within the experimental groups.

Table 5. Processing conditions for optimal water holding capacity of cassava flour

Desired level	Predicted value (%)	COP (%w/v)	DT (ºC)	Desirability
		CAR		
Minimum	41.411	2.081	59.712	1.000
Maximum	74.144	1.000	50.000	0.731
		CCR		
Minimum	62.241	3.000	50.000	0.880
Maximum	88.925	1.000	50.000	0.753
		CAW		
Minimum	50.869	3.000	50.000	0.870
Maximum	85.624	1.648	62.457	0.979
		CCW		
Minimum	52.853	3.000	50.000	0.821
Maximum	86.721	1.100	67.500	1.000

COP- Concentration of pre-treatment; DT- Drying temperature; WHC- Water holding capacity; CAR- Cassava flour from red landrace pre-treated with citric acid; CCR- Cassava flour from red landrace pre-treated with calcium chloride; CAW- Cassava flour from white landrace pre-treated with citric acid; CCW- Cassava flour from white landrace pre-treated with calcium chloride.

3.2 Effect of Pre-treatment and Drying Temperature on Bulk Densities of Cassava Flour

Bulk density is an important property that has a direct impact on the packaging requirement of flours [34]. The loose bulk density ranged between 0.34 – 0.41 g/cm^3 (CAR), 0.37 - 0.45 g/cm^3 (CCR), 0.35 – 0.43 g/cm^3 (CAW) and 0.37 – 0.44 g/cm^3 for CCW, respectively. Chisenga et al. [35] reported a similar range of 0.40 – 0.47 g/cm^3. The flours from the red cassava landrace exhibited a similar trend as in WHC, where the LBD of all the CCR samples was higher than the CAR under the same drying temperature and concentration of pre-treatment. This was also observed for most CCW samples in comparison with their corresponding CAW samples. This suggests that the bulk density and WHC of the flours, under this processing condition, may positively correlate. It was also observed that the lowest LBD for all the experimental groups was exhibited at 45DT/2COP, while the highest LBD was exhibited at 70DT/3COP for CAR and 74DT/2COP for CCR, CAW, and CCW. It can be observed that the lowest LBD was below the lower limit at drying temperature (45 °C), while the highest LBDs were at the other extreme, 70 and 74 °C. This implies that drying temperature influences the bulk density of the flours. As seen in the response contour plots for LBD of CAR, CCR, CAW, and CCW (Fig. 1), LBD increased along the X1-DT axis for cassava flour from the red landrace irrespective of pre-treatment type. For the white landrace CCW and CAW, an initial increase and decrease were observed along the X1-DT axis. This finding is consistent with Taiwo et al. [36], who reported that fermented CF from cassava chips dried at 70 °C in the oven had higher bulk densities than fermented CF from sundried cassava chips. Taiwo et al. [36] also stated that temperatures at which the chips were dried significantly ($p < 0.05$) influenced the BD of their respective flours.

It is expected that the packed bulk density (PBD) will be higher than the LBD due to the compacting effect of tapping on the flour particles. The PBD of CF pre-treated with calcium chloride and citric acid were similar, with the lowest and highest PBD of 0.62 and 0.73 g/mL (Fig. 2). The values of LBD (0.51-0.54 g/mL) and PBD (0.58-0.71 g/mL) of CF reported by Falade et al. [18] are similar in range to the obtained results reported in this study. Atlaw [37] also recorded similar values of 0.49-0.58 g/mL for PBD of CF dried using different methods. Fig. 3 shows the contour response plots for the difference between LBD and PBD (ΔBD), which was determined to give a clue on the average particle size of the flour samples. The difference in the bulk densities for CAR (0.26-0.34 g/mL) was slightly higher than that of CCR (0.24 - 0.30 g/ml), which may suggest that CAR has smaller particle sizes than CCR. A decrease in particle size of flour results in an increase in WHC and swelling power [38]. The bulk density of a flour is important not only for packaging but also for product development. Flowability, compressibility, and fluidity of a material are directly influenced by the bulk densities [18]. High bulk density indicates higher ease of dispersibility, which is a vital parameter in dough formation and infant food formulation [37].

Contour response surface plots generated by RSM aid visualisation of changes in the response variable in relation to the experimental factors. Contour plots of the PBD and ΔBD of CF samples in the four experimental groups are shown in Figs. 2 and 3. The patterns of the contour plots were different in all experimental groups.

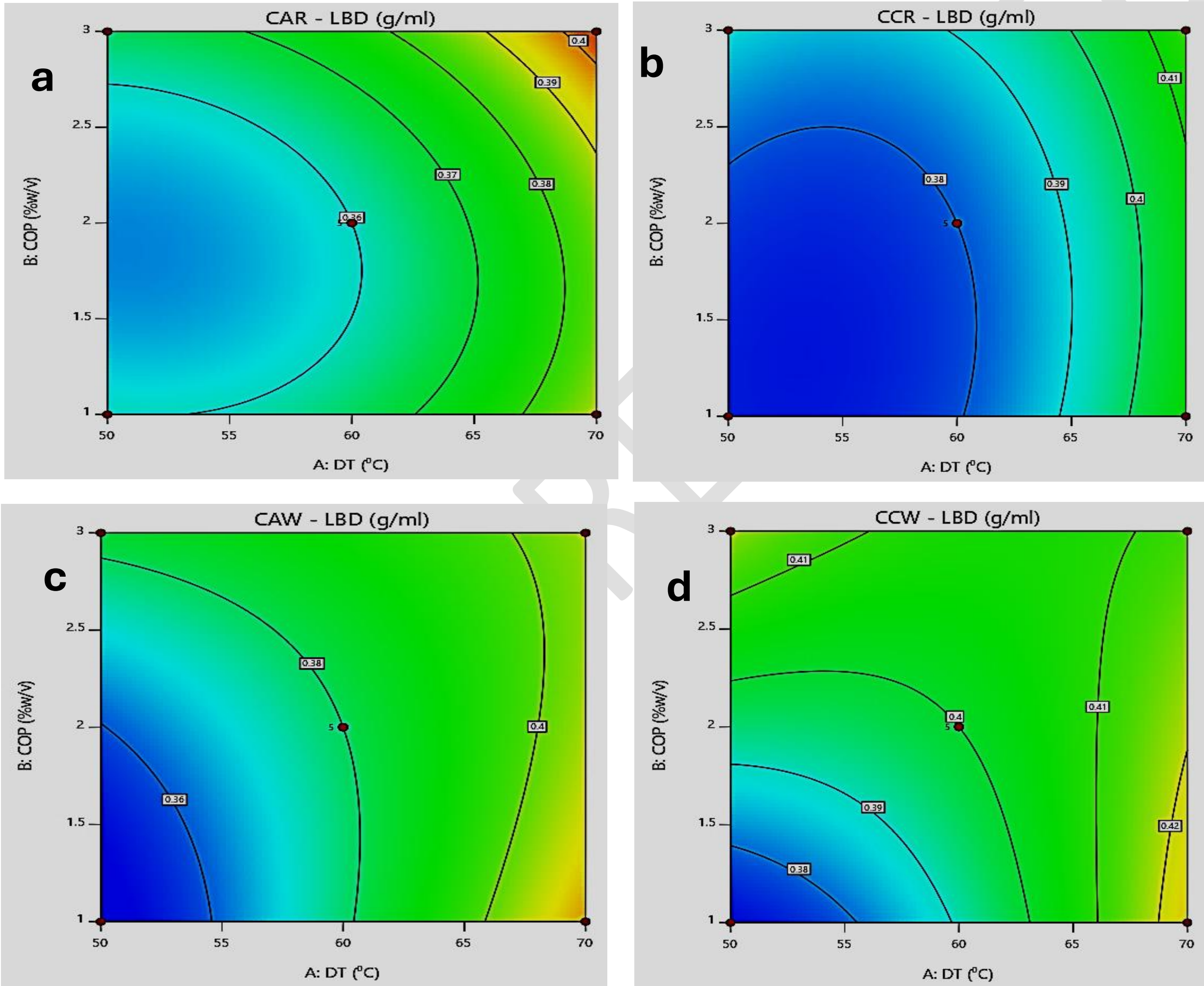

Fig. 1 Contour response surface plots for the effect of process variables on loose bulk densities of cassava flour. **a** = CAR - cassava flour from red landrace pre-treated with citric acid; **b** = CCR - cassava flour from red landrace pre-treated with calcium chloride; **c** = CAW - cassava flour from white landrace pre-treated with citric acid; **d** = CCW - cassava flour from white landrace pre-treated with calcium chloride.

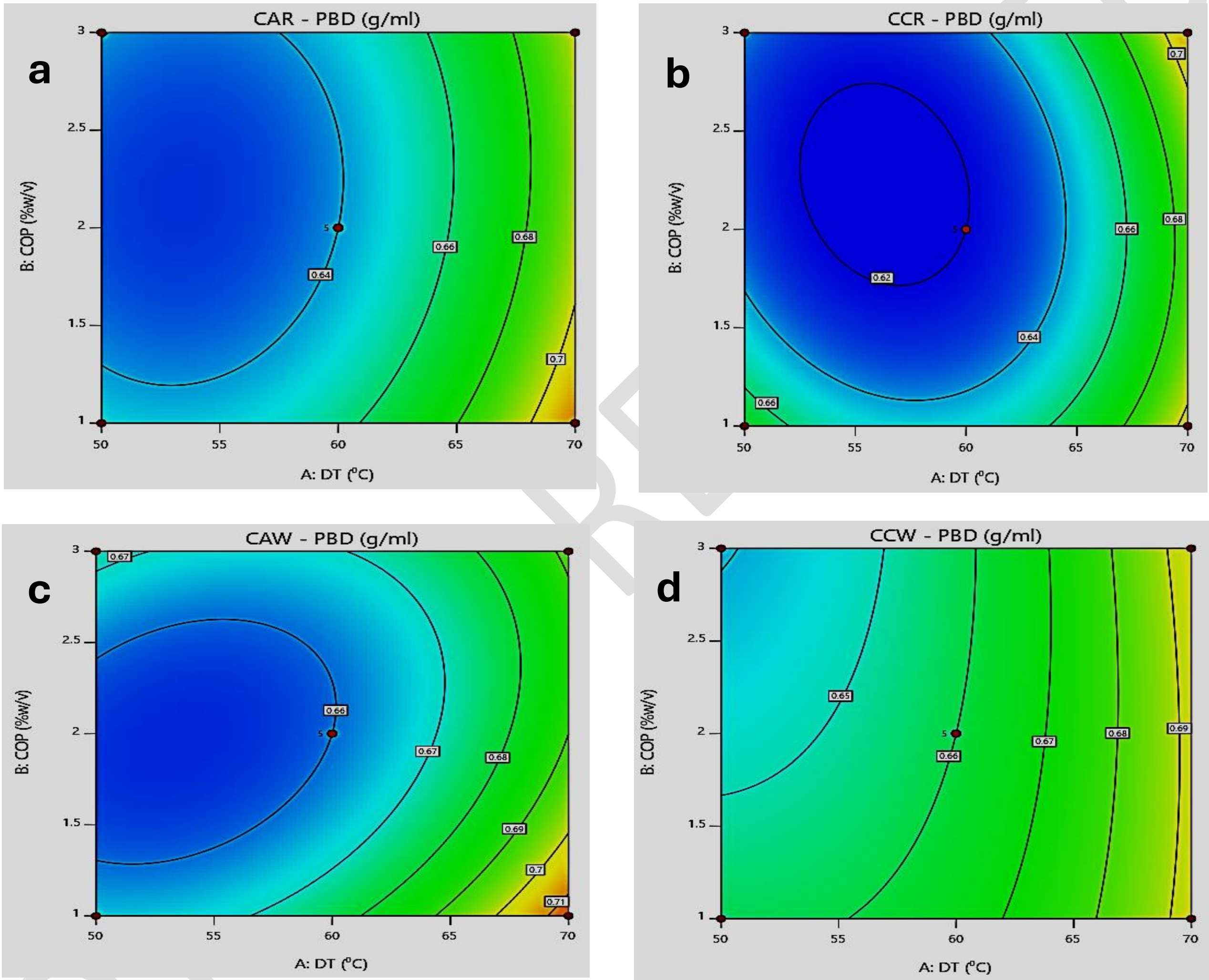

Fig. 2 Contour response surface plots for effect of process variables on packed bulk densities of cassava flour. **a** = CAR - cassava flour from red landrace pre-treated with citric acid; **b** = CCR - cassava flour from red landrace pre-treated with calcium chloride; **c** = CAW - cassava flour from white landrace pre-treated with citric acid; **d** = CCW- cassava flour from white landrace pre-treated with calcium chloride.

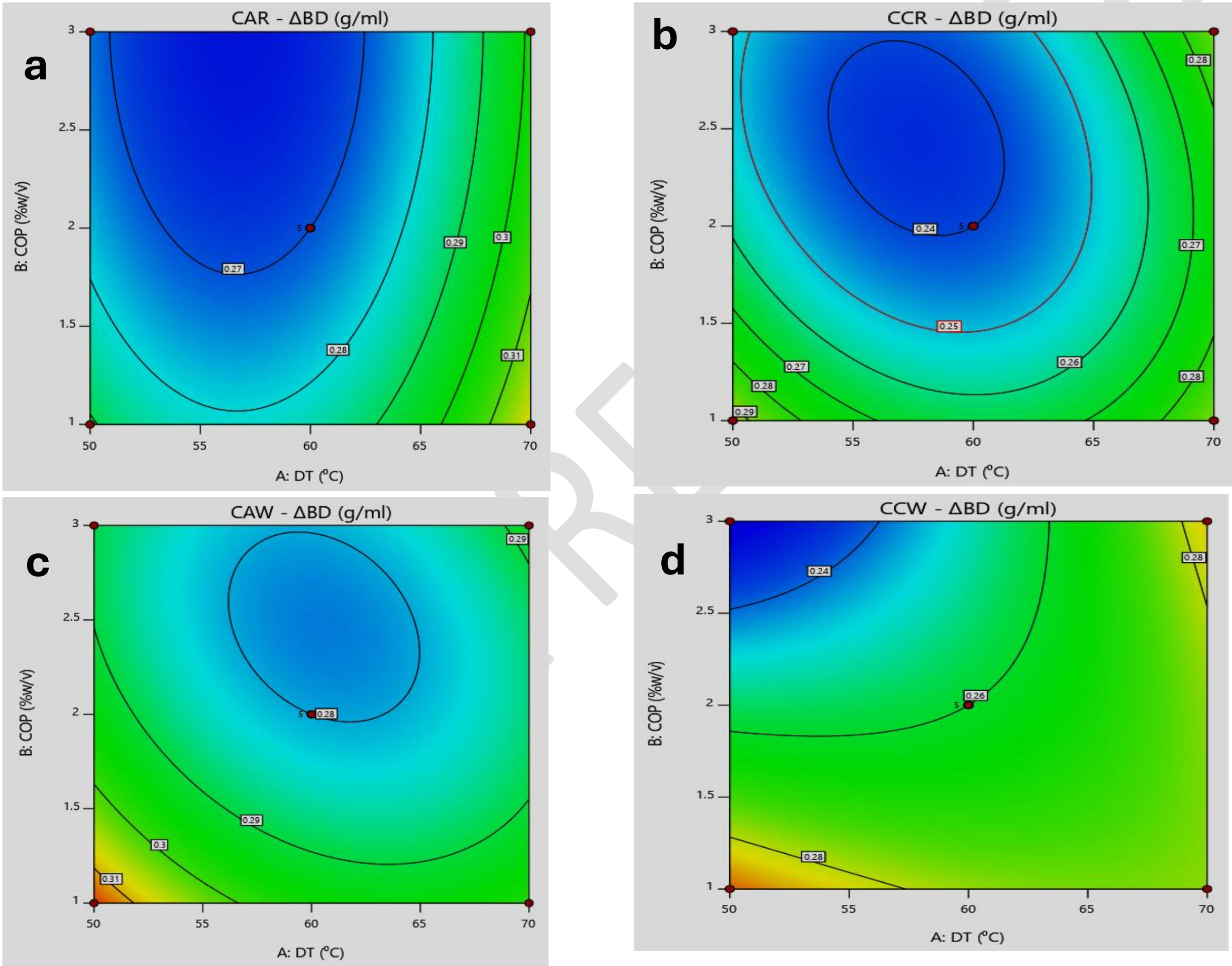

Fig. 3 Contour response surface plots for effect of process variables on difference in bulk density (ΔBD) of cassava flour; **a** = CAR - cassava flour from red landrace pre-treated with citric acid; **b** = CCR - cassava flour from red landrace pre-treated with calcium chloride; **c** = CAW - cassava flour from white landrace pre-treated with citric acid; **d** = CCW - cassava flour from white landrace pre-treated with calcium chloride.

This suggests that the landrace and pre-treatment type influenced the bulk densities of CF. This is buttressed by the variation in the ANOVA for model parameters (Table 6) and relatively high coefficient of determination values (0.6042 – 0.9458) shown in Table 7.

Table 6. ANOVA of the effect of model parameters on bulk densities of cassava flour

Source	CAR		CCR		CAW		CCW	
				LBD (g/mL)				
	F-value	p-value	F-value	p-value	F-value	p-value	F-value	p-value
Model	8.40	0.0072*	2.99	0.0930	13.30	0.0018*	2.30	0.1533
A-DT	25.13	0.0015*	8.50	0.0225*	52.03	0.0002*	4.62	0.0686
B-COP	2.69	0.1448	1.09	0.3307	4.26	0.0778	2.20	0.1816
AB	0.2763	0.6153	0.1121	0.7476	7.76	0.0271*	3.40	0.1079
A^2	5.89	0.0457*	4.87	0.0630	0.5396	0.4865	1.24	0.3021
B^2	9.73	0.0169*	0.7798	0.4065	2.16	0.1852	0.0103	0.9222
				PBD (g/mL)				
Model	6.75	0.0131*	16.14	0.0010*	11.48	0.0029*	3.66	0.0602
A-DT	21.23	0.0025*	23.91	0.0018*	27.58	0.0012*	15.14	0.0060*
B-COP	0.6545	0.4451	1.35	0.2828	1.37	0.2809	0.5361	0.4878
AB	0.0672	0.8029	1.68	0.2355	2.73	0.1424	0.3679	0.5633
A^2	10.54	0.0141*	45.41	0.0003*	14.54	0.0066*	2.25	0.1773
B^2	2.37	0.1679	14.01	0.0072*	14.54	0.0066*	0.0900	0.7729
				ΔBD (g/mL)				
Model	4.52	0.0369*	4.12	0.0460*	2.14	0.1751	24.45	0.0003*
A-DT	6.69	0.0361*	1.45	0.2679	0.5956	0.4655	28.11	0.0011*
B-COP	2.71	0.1439	2.40	0.1649	2.90	0.1323	47.41	0.0002*
AB	3.42	1.0000	1.07	0.3357	0.5956	0.4655	36.76	0.0005*
A^2	13.11	0.0085*	13.42	0.0080*	4.68	0.0673	8.15	0.0245*
B^2	0.5246	0.4924	3.84	0.0909	2.74	0.1421	2.94	0.1304

*- significant at ($p > 0.05$); DT- Drying temperature; COP- concentration of pre-treatment; A^2-quadractic effect of DT; B^2-quadractic effect of COP; AB- interactive effect of A and B; CAR- cassava flour from red landrace pre-treated with citric acid; CCR- cassava flour from red landrace pre-treated with calcium chloride; CAW- cassava flour from white landrace pre-treated with citric acid; CCW- cassava flour from white landrace pre-treated with calcium chloride.

Table 7. Regression models relating bulk densities and model parameters for cassava flour

Response	Models	R^2
	CAR	
LBD	$+0.3600 + 0.0169A + 0.0055B + 0.0025AB + 0.0087A^2 + 0.0113B^2$	0.8571
PBD	$+0.6400 + 0.0314A - 0.0055B - 0.0025AB + 0.0237A^2 + 0.0113B^2$	0.8283
ΔBD	$+0.2700 + 0.0146A – 0.0093B + 0.0000AB + 0.0219A^2 + 0.0044B^2$	0.7633
	CCR	
LBD	$+0.3800 + 0.0154A + 0.0055B – 0.0025AB + 0.0125A^2 + 0.0050B^2$	0.6809
PBD	$+0.6200 + 0.0266A – 0.0063B + 0.0100AB + 0.0394A^2 + 0.0219B^2$	0.9202
ΔBD	$+0.2400 + 0.0082A – 0.0106B + 0.0100AB + 0.0269A^2 + 0.0144B^2$	0.7462
	CAW	
LBD	$+0.3800 + 0.0229A + 0.0066B – 0.0125AB + 0.0025A^2 + 0.0050B^2$	0.9048
PBD	$+0.6600 + 0.0169A – 0.0038B – 0.0075AB + 0.0131A^2 + 0.0131B^2$	0.8913
ΔBD	$+0.2800 – 0.0035A – 0.0078B + 0.0050AB + 0.0106A^2 + 0.0081B^2$	0.6042
	CCW	
LBD	$+0.4000 +0.0124A + 0.0085B – 0.0150AB + 0.0069A^2 - 0.0006B^2$	0.6221
PBD	$+0.6600 + 0.0227A – 0.0043B + 0.0050AB + 0.0094A^2 + 0.0019B^2$	0.7233
ΔBD	$+0.2600 + 0.0108A – 0.0141B + 0.0175AB + 0.0063A^2 + 0.0038B^2$	0.9458

R^2- Coefficient of determination; LBD- loose bulk density; PBD- packed bulk density; ΔBD- difference in bulk density; CAR- cassava flour from red landrace pre-treated with citric acid; CCR- cassava flour from red landrace pre-treated with calcium chloride; CAW- cassava flour from white landrace pre-treated with citric acid; CCW- cassava flour from white landrace pre-treated with calcium chloride.

3.3 Effect of Pre-treatment and Drying Temperature on Thermal Properties of Cassava Flour

Gelatinisation is an irreversible change that occurs when starch is heated in water, characterised by gelatinisation temperatures and enthalpy, which can be determined using differential scanning calorimetry [4]. Onset gelatinisation temperature (To) of 60.32 to 120.30 ºC; peak gelatinisation temperature (Tp) of 71.85 to 126.84 ºC and conclusion gelatinisation temperature (Tc) of 93.31 to 140.98 ºC were recorded for all CF samples. The response plots for the thermal properties of CF processed from the red and white cassava landraces are shown in Figs. 4 and 5, respectively. Cassava flour from the red landrace pre-treated with citric acid (CAR) shows that gelatinisation temperatures increased with drying temperature and concentration of pre-treatment. As for CCR, the gelatinisation temperatures decrease and increase along both axes of the independent variables. The peak and conclusion gelatinisation temperatures of CF from the white landrace pre-treated with citric acid increased with an increase in COP and no increase with DT. It could be observed that the gelatinisation temperature of CCW increased with the concentration of pre-treatment. Variation in the trend displayed by the experimental groups for gelatinisation temperatures implies that the type of landrace and processing variables influenced the thermal properties of the flours. The differences in gelatinisation could be ascribed to variations in non-starch content [39].

Gelatinisation enthalpy (ΔH) is associated with the energy required for breaking of double helices in starch granules. It reflects the loss of double-helical order or overall crystallinity of starch [40] in the flour. Gelatinisation enthalpy (ΔH) within the range of 0.14 to 54.95 J/g was recorded for all the samples in this study. Wide variations were observed in the endothermic enthalpies of the samples within each experimental group (CAR, CCR, CAW, CCW). For example, ANOVA using Duncan's Multiple Test to identify the significant difference between the experimental runs in CAW (Table 8) produced 10 homogeneous subsets with each experimental run significantly different ($p < 0.05$) from the other.

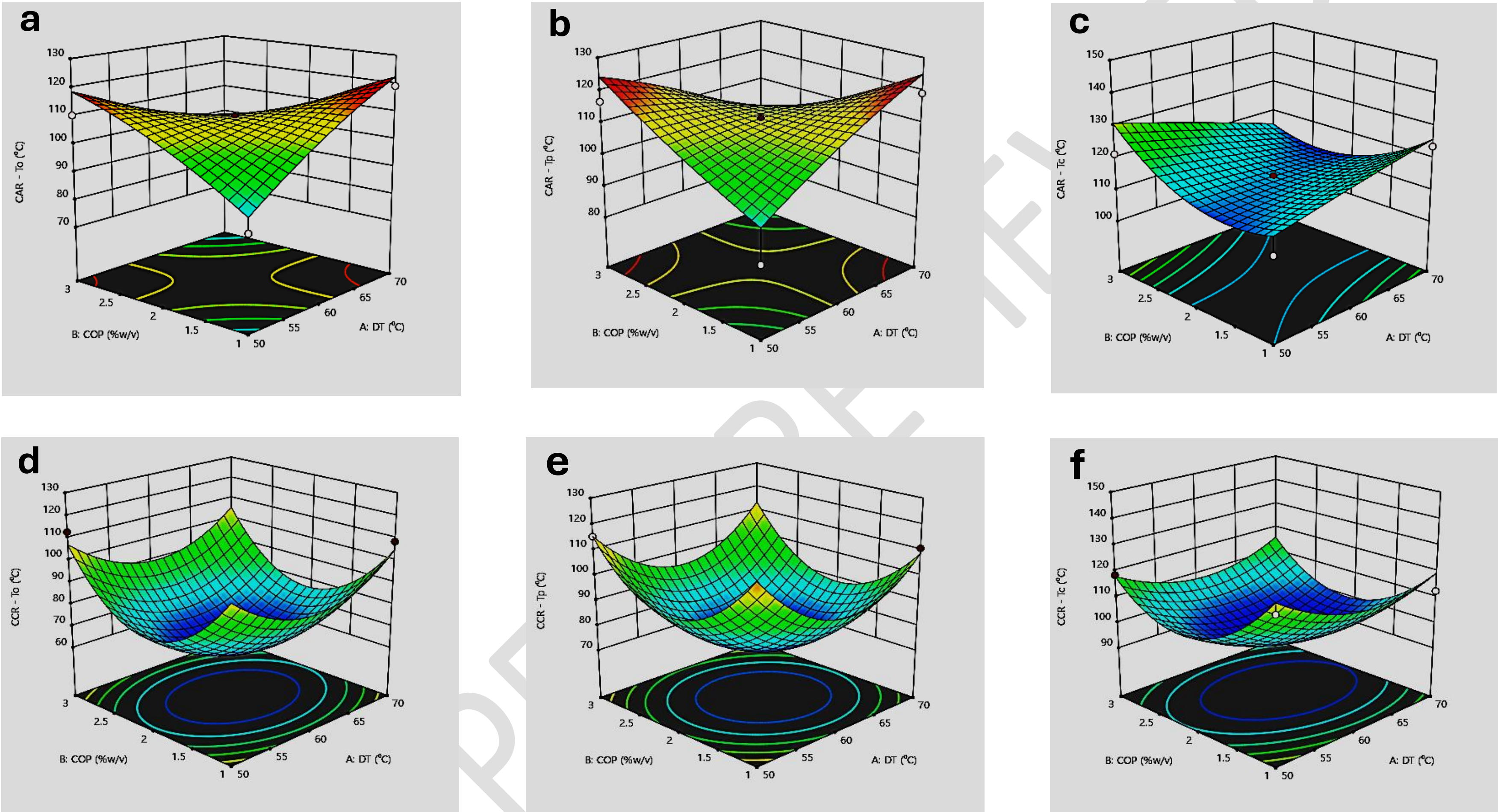

Fig. 4 Response surface plots for gelatinization temperatures of cassava flour from red landrace; DT- drying temperature, COP- Concentration of pre-treatment; CAR – Citric acid pre-treated cassava flour from red landrace; CCR – Calcium chloride pre-treated cassava flour from red landrace; **a** – Onset gelatinisation temperature of CAR; **b** – Peak gelatinisation temperature of CAR; **c** – Conclusion gelatinisation temperature of CAR; **d** - Onset gelatinisation temperature of CCR; **e** – Peak gelatinisation temperature of CCR; **f** – Conclusion gelatinisation temperature of CCR.

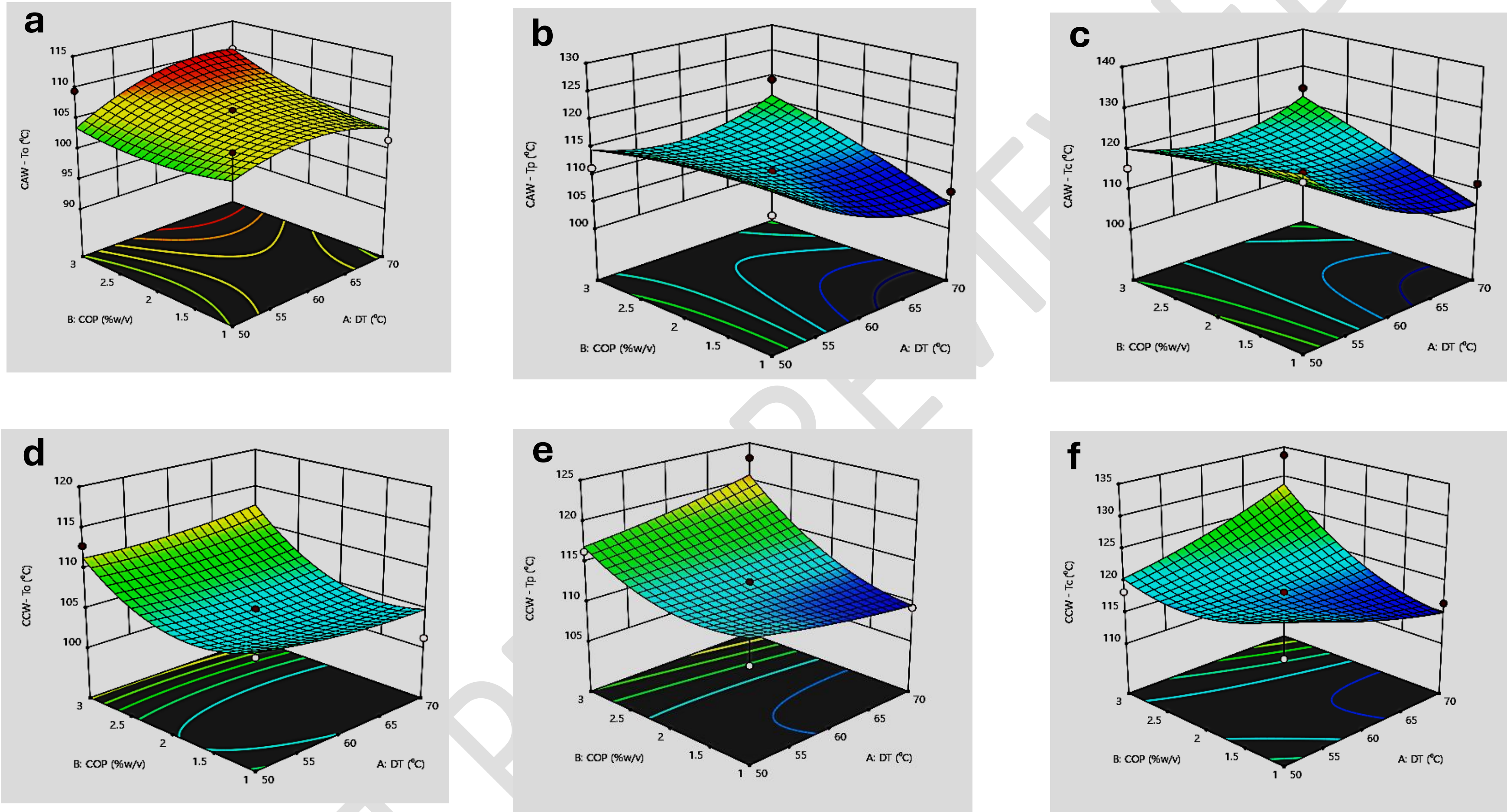

Fig. 5 Response surface plots for gelatinization temperatures of cassava flour from white landrace; DT- drying temperature, COP- Concentration of pre-treatment; CAW – Citric acid pre-treated cassava flour from white landrace; CCW – Calcium chloride pre-treated cassava flour from white landrace; **a** – Onset gelatinisation temperature of CAW; **b** – Peak gelatinisation temperature of CAW; **c** – Conclusion gelatinisation temperature of CAW; **d** - Onset gelatinisation temperature of CCW; **e** – Peak gelatinisation temperature of CCW; **f** – Conclusion gelatinisation temperature of CCW.

Table 8. Gelatinization enthalpy of pre-treated flour from red and white cassava landrace

	Independent variables		Gelatinization enthalpy (J/g)			
Exp Run	DT (ºC)	COP (%w/v)	CAR	CCR	CAW	CCW
1	50	1	$0.44^{h} \pm 0.04$	$0.94^{e} \pm 0.04$	$39.64^{d} \pm 0.55$	$0.89^{g} \pm 0.03$
2	70	1	$1.07^{g} \pm 0.03$	$1.11^{e} \pm 0.01$	$4.51^{i} \pm 0.02$	$8.35^{e} \pm 0.31$
3	50	3	$7.33^{e} \pm 0.03$	$1.95^{e} \pm 0.05$	$17.34^{g} \pm 0.29$	$1.59^{f} \pm 0.08$
4	70	3	$6.16^{f} \pm 0.14$	$1.27^{e} \pm 0.03$	$15.58^{h} \pm 0.07$	$1.18^{fg} \pm 0.02$
5	45	2	$42.12^{a} \pm 0.10$	$33.38^{b} \pm 0.33$	$32.64^{e} \pm 0.04$	$24.54^{d} \pm 0.51$
6	74	2	$17.80^{d} \pm 0.26$	$54.95^{a} \pm 2.62$	$21.14^{f} \pm 0.04$	$43.36^{c} \pm 0.53$
7	60	0.6	$23.49^{c} \pm 0.42$	$17.43^{c} \pm 0.51$	$42.35^{c} \pm 0.30$	$47.42^{b} \pm 0.36$
8	60	3.4	$38.33^{b} \pm 0.16$	$6.35^{d} \pm 0.30$	$46.88^{b} \pm 0.10$	$1.60^{f} \pm 0.10$
9	60	2	$1.01^{g} \pm 0.02$	$1.62^{e} \pm 0.11$	$52.07^{a} \pm 0.06$	$49.79^{a} \pm 0.09$
10	60	0	$1.22^{g} \pm 0.02$	$1.22^{e} \pm 0.02$	$0.14^{j} \pm 0.02$	$0.15^{h} \pm 0.01$

DT- Drying temperature; COP- concentration of pre-treatment; CAR- Cassava flour from red landrace pre-treated with citric acid; CCR- Cassava flour from red landrace pre-treated with calcium chloride; CAW- Cassava flour from white landrace pre-treated with citric acid; CCW- Cassava flour from white landrace pre-treated with calcium chloride; WHC-Water absorption capacity.

It is also interesting to note that in most groups, the control exhibited low values of enthalpy. This indicates that the processing conditions have an increasing effect on the enthalpy of the starch granules in the flour. As reported by Huang et al. [41], a high endothermic value appears around 100 ºC, the boiling point of water, and presumably reflects the vaporisation of water in the starch sample and the destruction of the crystalline structure between water and starch. Effective moisture diffusivity, thermal conductivity and heat capacity of CF increase with temperature [42]. In Table 9 are regression models relating thermal properties to drying temperature and concentration of pre-treatment. The equation in terms of coded factors (A- DT and B- COP) can be used for predictions about the response for given levels of each factor. The coded equation is useful for identifying the relative impact of the factors by comparing the factor coefficients. The coefficient of determination (R^2) obtained was relatively high. The values of gelatinisation temperatures reported [43-47] for cassava starch were below the values of this study. Although CF consists mostly of starch, its gelatinisation temperatures (onset, peak, and conclusion) were higher than those of cassava starch as reported in the work of Charoenkul et al. [48].

Table 9. Regression models relating thermal properties and model parameters of cassava flour

Response variables	Models	R^2
CAR		
To (ºC)	$+108.95+0.2174A-1.67B-16.76AB-2.40A^2-2.88B^2$	0.8079
Tp (ºC)	$+111.48+0.5867A+0.1812B-13.70AB-1.80A^2+1.24B^2$	0.6513
Tc (ºC)	$+114.36-1.11A+1.34B-6.54AB-0.3350A^2+7.89B^2$	0.6572
ΔH (J/g)	$+1.01-4.37A+4.12B-0.4508AB+7.80A^2+8.28B^2$	0.4077
CCR		
To (ºC)	$+60.32-0.5113A+0.7556B-0.4050AB+16.87A^2+28.40B^2$	0.9166
Tp (ºC)	$+71.85-2.66A+0.7340B+1.63AB+19.44A^2+22.43B^2$	0.9759
Tc (ºC)	$+93.31-2.87A-3.52B+1.72AB+9.16A^2+18.13B^2$	0.9508
ΔH (J/g)	$+1.62+3.75A-1.81B-0.2125AB+14.59A^2-1.54B^2$	0.5186
CAW		
To (ºC)	$+106.39+1.54A+1.64B+1.76AB-2.49A^2+1.32B^2$	0.5082
Tp (ºC)	$+110.61-2.94A+2.03B+3.74AB+3.48A^2-0.7146B^2$	0.7433
Tc (ºC)	$+114.23-4.87A+2.14B+5.82AB+4.84A^2-0.0408B^2$	0.8158
ΔH (J/g)	$+52.07-6.64A-0.6018B+8.34AB-16.71A^2-7.85B^2$	0.8153
CCW		
To (ºC)	$+104.95-0.2586A+2.95B+0.8725AB+0.5538A^2+3.50B^2$	0.8062
Tp (ºC)	$+112.55+0.0563A+3.73B+1.85AB+0.3300A^2+2.23B^2$	0.8355
Tc (ºC)	$+118.25+0.0776A+2.81B+4.09AB+0.7594A^2+2.80B^2$	0.6043

ΔH (J/g)	+49.79+4.21A-8.91B-1.97AB-14.48A^2-19.20B^2	0.7027

To- Onset gelatinization temperature, Tp- Peak gelatinization temperature, Tc- Conclusion gelatinization temperature, ΔH- gelatinization enthalpy, R^2- Coefficient of determination, CAR- cassava flour from red landrace pre-treated with citric acid; CCR- cassava flour from red landrace pre-treated with calcium chloride, ; CAW- cassava flour from white landrace pre-treated with citric acid; CCW- cassava flour from white landrace pre-treated with calcium chloride.

Relatively high values of To (93.68-114.21ºC), Tp (100.35-118.49 ºC), Tc (108.73-123.51 ºC), and ΔH (3.74-11.54 J/g) reported by Omolola et al. [22] for CF not chemically pre-treated are within the range obtained in this study. Omolola et al. [22], who studied the influence of optimised drying time (15-20 h) and temperature (60-70 ºC) on the thermal properties of CF, showed that the optimised drying conditions did not influence the thermal properties of CF. Non-starch components in CF such as fat could be responsible for high gelatinisation temperatures and enthalpy. Components such as lipids may affect the diffusion of water into the starch granules, and their presence on starch granules was demonstrated to retard gelatinisation. Li et al. [49] reported that defatting starch results in decreased gelatinisation temperatures, which agrees with Charles et al. [50] who postulated that the presence of amylose–lipid complex inhibits the gelatinisation of starch granules. Zhao and Saldaña [47] reported higher onset, peak, and conclusion gelatinisation temperatures for cereal and potato starches which contain a high amount of lipids, while cassava starches exhibited lower onset gelatinisation temperatures. The samples in this study contained non-starch components and were not defatted. High gelatinisation temperatures were associated with a high degree of crystallinity [51], which relates to starch granule structural stability, thus making it resistant to heating. The results of X-ray diffractometry (XRD) studies for the flours from the red and white cassava landraces indicate that they exhibit an A-type XRD pattern with a high degree of crystallinity, which could be responsible for the high gelatinisation temperatures. The highly crystalline region tends to increase the gelatinisation temperature in flour because it interrupts water penetration within starch molecules [52]. Guo et al. [53] showed that A-type starch has high gelatinisation temperatures when compared to the B- and C-type starch

Optimum processing conditions generated by the Design Expert software are displayed graphically as overlay plots (Fig. 6). The design space is used in determining the optimum variable values for the target goals. Starch of low gelatinisation temperature is cost-effective in industrial food production as it limits energy expenses [16]. Conversely, flour with higher gelatinisation temperature is more stable and resistant to heat in the presence of water, which may also be desirable in some food formulations. However, the thermal properties of samples used in this study were relatively high, compared to the literature; therefore, the optimisation goal was to minimise their thermal properties. Multi-response optimization was thus carried out for all the experimental groups and the optimum values of To, Tp, Tc, and ΔH at the various optimised drying conditions are depicted in their respective overlay plots (Fig. 6). The optimal processing conditions for minimised gelatinisation temperatures and enthalpy are 50DT/1COP with desirability of 0.805 for CAR, 57.23DT/1.75COP with 0.841 desirability for CCR, 70DT/1COP with 0.754 desirability for CAW, and 50DT/1COP with 0.732 desirability for CCW.

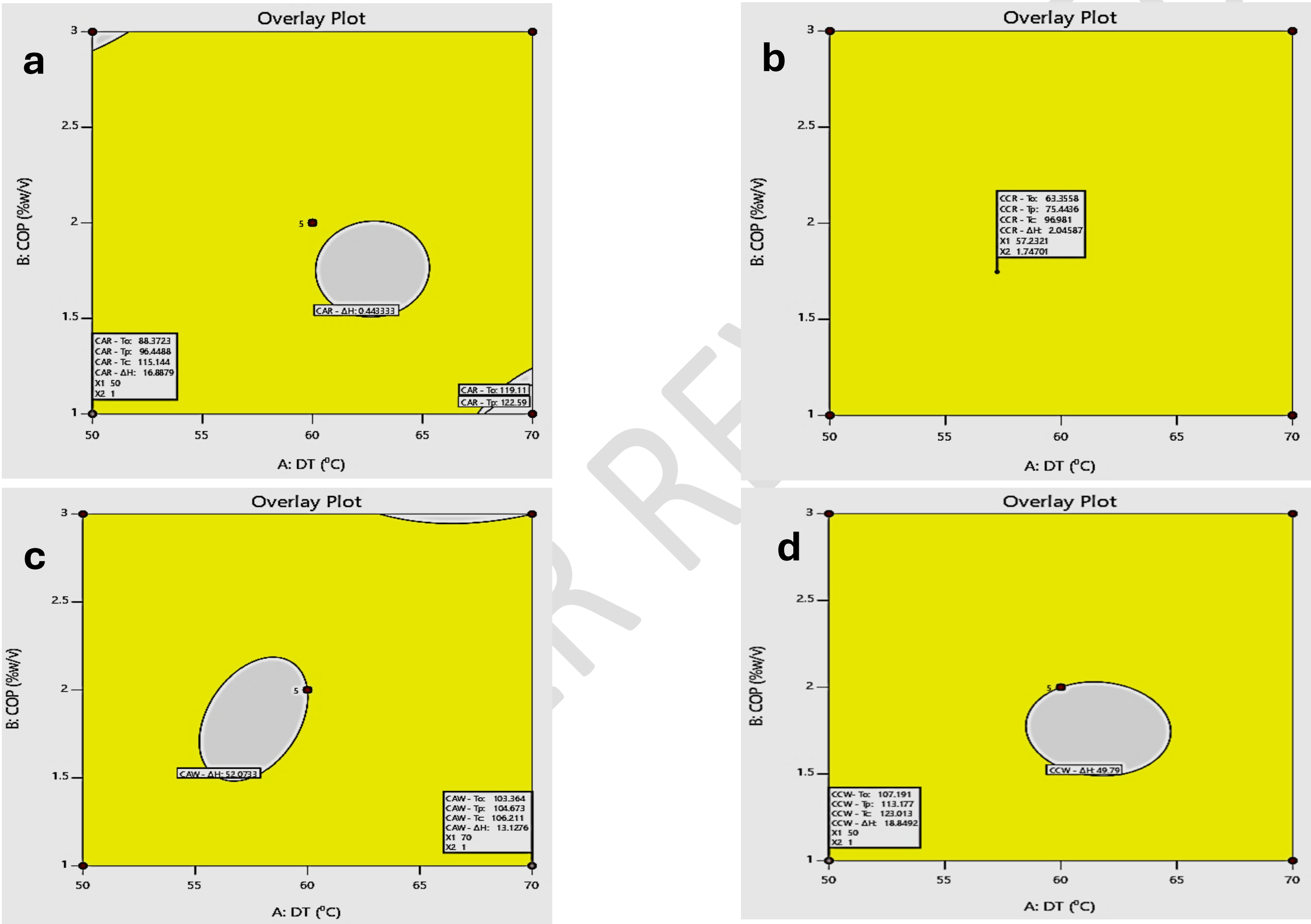

Fig. 6 Multi-response optimization overlay plots of processing conditions and thermal properties of cassava flour; DT- drying temperature, COP- Concentration of pre-treatment. **a** - cassava flour from red landrace pre-treated with citric acid; **b** - cassava flour from red landrace pre-treated with calcium chloride; **c** - cassava flour from white landrace pre-treated with citric acid; **d** - cassava flour from white landrace pre-treated with calcium chloride.

4 Conclusion

This study showed that pre-treatment of CF using citric acid and calcium chloride influenced the BD, WHC, and thermal properties of the flour. It was observed that pre-treatment led to an increase in the amount of energy required for gelatinisation to occur. Higher R^2 values were recorded for the WHC of CF pre-treated with citric acid when compared to calcium chloride. This was consistent with the significant influence of the quadratic effect of citric acid pre-treatment on WHC of CF. The optimised drying temperature influenced the BD of CF with an increase in LDB as temperature increased. Results obtained in this study, using response surface methodology, provide useful information for CF processing and product development.

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Author contributions

EOO, TAA and AIOJ conceptualised the work. EOO generated data and wrote the manuscript draft. TAA and AIOJ supervised the work. OOO, TAA and AIOJ edited the manuscript draft. EOO and AIOJ contributed to funding the research. All authors read and approved the final manuscript.

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Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

Not applicable. This study did not involve human or animal subjects.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

References

1. FAO. Save and grow cassava: A guide to sustainable production intensification. Rome, FAO. (2013). Available at: http://www.fao.org/ag/save-and-grow/cassava/index_en.html. Accessed 16 November, 2019.
2. Dudu OE, Oyedeji AB, Oyeyinka SA, Ma Y. Impact of steam-heat-moisture treatment on structural and functional properties of cassava flour and starch. Int J Biol Macromol. 2019; 126: 1056-1064.
3. El-Sharkawy MA. Global warming: causes and impacts on agroecosystems productivity and food security with emphasis on cassava comparative advantage in the tropics/subtropics. Photosynthetica. 2014; 52(2): 161-178.

4. Zhu F. Composition, structure, physicochemical properties, and modifications of cassava starch. Carbohydr Polym. 2015; 122, 456-480.

5. Aristizábal J, García JA, Ospina B. Refined cassava flour in bread making: a review. Ing Investig. 2017; 37(1): 25-33. http://dx.doi.org/10.15446/ing.investig.v37n1.57306

6. Ramírez M, Tenorio M, Ramirez C, Jaques A, Nuñez H, Simpson R, Vega O. Optimisation of hot-air drying conditions for cassava flour for its application in gluten-free pasta formulation. Food Sci Technol Int. 2019; 25(5), 414-428.

7. Agunbiade S, Ojezele O, Eze A. Maximising the incorporation of cassava flour as an adjunct in bread baking in Nigeria. Chem Int. 2017; 3(1): 92-96.

8. Alobo A, Arueya G. Physical, functional and chemical properties of Grewia venusta (ururu) mucilage extract. Int Food Res J. 2017; 24(5): 2107-2115.

9. Akingbala JO, Falade KO, Ogunjobi MA. The effect of root maturity, preprocess holding and flour storage on the quality of cassava biscuit. Food Bio Technol. 2011; 4(3): 451–457. https://doi.org/10.1007/s11947-009-0185-z

10. Lu H, Guo L, Zhang L, Xie C, Li W, Gu B, Li K. Study on quality characteristics of cassava flour and cassava flour short biscuits. Food Sci Nutr. 2020; 8(1): 521-533.

11. Abidin AZ, Devi C, Adeline A. Development of wet noodles based on cassava flour. J. Eng Technol Sci. 2013; 45(1): 97-111.

12. Falade KO, Akingbala JO. Improved nutrition and national development through the utilisation of cassava in baked foods. In: Roberts GL, Lupien JR (Eds), Using Food Science and Technology to Improve Nutrition and Promote National Development, *International Union of Food Science & Technology*, (2008). 1-12.

13. Cornejo-Ramírez YI, Martínez-Cruz O, Del Toro-Sánchez CL, Wong-Corral FJ, Borboa-Flores J, Cinco-Moroyoqui FJ. The structural characteristics of starches and their functional properties. CyTA – J Food. 2018; 16(1): 1003-1017.

14. Jyothi AN, Sajeev MS, Sreekumar JN. Hydrothermal modifications of tropical tuber starches. 1. Effect of heat-moisture treatment on the physicochemical, rheological and gelatinisation characteristics. Starch-Stärke 2010; 62(1): 28-40.

15. Jyothi AN, Sajeev MS, Sreekumar JN. Hydrothermal modifications of tropical tuber starches–Effect of ANN on the physicochemical, rheological and gelatinisation characteristics. Starch-Stärke 2011; 63(9): 536-549.

16. Ayetigbo O, Latif S, Abass A, Müller J. Comparing characteristics of root, flour and starch of biofortified yellow-flesh and white-flesh cassava variants, and sustainability considerations: a review. Sustainability 2018; 10(9): 3089.
17. Omowaye-Taiwo OA, Fagbemi TN, Ogunbusola EM, Badejo AA. Effect of germination and fermentation on the proximate composition and functional properties of full-fat and defatted *Cucumeropsis mannii* seed flours. J Food Sci Technol. 2015; 52(8): 5257-5263.
18. Falade KO, Ibanga-Bamijoko B, Ayetigbo OE. Comparing properties of starch and flour of yellow-flesh cassava cultivars and effects of modifications on properties of their starch. J. Food Meas Charact. 2019; 13(4): 2581-2593.
19. Villanueva M, Ronda F, Moschakis T, Lazaridou A, Biliaderis CG. Impact of acidification and protein fortification on thermal properties of rice, potato and tapioca starches and rheological behaviour of their gels. Food Hydrocoll. 2018; 79, 20-29.
20. Zambelli RA, Galvão AMMT, de Mendonça LG, de Souza Leão MV, Carneiro SV, Lima ACS, Melo CAL. Effect of different levels of acetic, citric and lactic acid in the cassava starch modification on physical, rheological, thermal and microstructural properties. Food Sci Technol Res. 2018; 24(4), 747-754.
21. Udoro EO, Anyasi TA, Jideani AIO. Characterisation of the root and flour of South African *Manihot esculenta* Crantz landraces and their potential end-use properties. Int J Food Prop. 2020; 23(1), 820-838. https://doi.org/10.1080/10942912.2020.1759625
22. Omolola AO, Kapila PF, Anyasi TA, Jideani AIO, Mchau GRA. Optimisation of colour and thermal properties of sweet cassava (*Manihot esculenta* Crantz Var. UVLNR 0005) flour using response surface methodology. Asian J Agric Res. 2017; 11(3): 57-65.
23. Nemaungani P, Udoro EO, Anyasi TA, Jideani AIO. Physico-functional changes associated with chemical pre-treatment of solar dried flour from South African *Manihot esculenta* Crantz landraces. Proceedings of the 43rd Conference and annual general meeting, Nigerian Institute of Food Science and Technology, Awka, Nigeria (2019). 14th -18th October, 2019. pp:199-200.
24. Coelho DG, Fonseca KS, de Mélo Neto DF, de Andrade MT, Coelho Junior LF, Ferreira-Silva SL, Simões ADN. Association of preharvest management with oxidative protection and enzymatic browning in minimally processed cassava. J Food Biochem. 2019; 43(5): e12840. https://doi.org/10.1111/jfbc.12840
25. Federal Institute of Industrial Research. Handbook on cassava processing. FIIRO, Oshodi, Lagos, Nigeria. (2005).

26. Onwuka G, Onwuka N. The effects of ripening on the functional properties of plantain and plantain-based cake. Int J Food Prop. 2005; 8(2): 347-353.
27. Anyasi TA, Jideani AIO, Mchau GRA. Effects of organic acid pretreatment on microstructure, functional and thermal properties of unripe banana flour. J Food Meas Charact. 2017; 11(1): 99-110.
28. Maninder K, Sandhu KS, Singh N. Comparative study of the functional, thermal and pasting properties of flours from different field pea (*Pisum sativum* L.) and pigeon pea (*Cajanus cajan* L.) cultivars. Food Chem. 2007; 104(1): 259-267.
29. Aremu M, Basu S, Gyar S, Goyal A, Bhowmik P, Banik SD. Proximate composition and functional properties of mushroom flours from *Ganoderma* spp., *Omphalotus olearius* (DC.) Sing. and *Hebeloma mesophaeum* (Pers.) Quél. used in Nasarawa State, Nigeria. Malaysian J Nutr. 2009; 15(2): 233-2241.
30. Odey GN, Lee WY. Evaluation of the quality characteristics of flour and pasta from fermented cassava roots. Int J Food Sci Technol. 2020; 55(2): 813-822.
31. Onitilo M, Sanni L, Oyewole O, Maziya-Dixon B. Physicochemical and functional properties of sour starches from different cassava varieties. Int J Food Prop. 2007; 10(3): 607-620.
32. Osungbaro TO, Jimoh D, Osundeyi E. Functional and pasting properties of composite cassava-sorgum flour meals. Agric. Biol. J. North Am. 2010; 1(4): 715-720.
33. Peluola-Adeyemi OA, Adepoju PA, Lawal SO. Physical properties of bread from wheat–cassava flour composite using response surface methodology (RSM). Am J Food Sci Technol. 2019; 7(4), 122-126.
34. Msheliza E, Hussein J, Ilesanmi J, Nkama I. Effect of fermentation and roasting on the physicochemical properties of weaning food produced from blends of sorghum and soybean. J Nutr Food Sci. 2018; 8(2): 1-8.
35. Chisenga SM, Workneh TS, Bultosa G, Laing M. Proximate composition, cyanide contents, and particle size distribution of cassava flour from cassava varieties in Zambia. AIMS Agric Food. 2019; 4(4): 869-891.
36. Taiwo K, Gbadamosi S, Izevbekhai E, Famuwagun A, Ajani R, Akanbi C. Influence of drying methods and soaking media on lafun processed from cassava chips. Curr J Appl Sci Technol. 2016; 1-14.
37. Atlaw TK. Influence of drying methods on flour quality and cyanide content of cassava root tuber. Int J Nutr Food Sci. 2018; 7(4): 142-147.

38. Rao BD, Anis M, Kalpana K, Sunooj K, Patil J, Ganesh T. Influence of milling methods and particle size on hydration properties of sorghum flour and quality of sorghum biscuits. LWT-Food Sci Technol. 2016; 67, 8-13.
39. Chisenga SM, Workneh TS, Bultosa G, Alimi BA. Progress in research and applications of cassava flour and starch: a review. J Food Sci Technol. 2019; 56(6): 2799–2813. https://doi.org/10.1007/s13197-019-03814-6
40. Oyeyinka S, Adeloye A, Smith S, Adesina B, Akinwande F. Physicochemical properties of flour and starch from two cassava varieties. Agrosearch. 2019; 19(1), 28-45.
41. Huang Z-Q, Xie X-l, Chen Y, Lu J-P, Tong Z-F. Ball-milling treatment effect on physicochemical properties and features for cassava and maize starches. C. R. Chim. 2008; 11(1-2): 73-79.
42. Sanni LA, Oke OO, Oladimeji FM, Ogbozomivaze KF. Thermal properties and energy utilisation of cassava meal in conductive rotary drying. Am J Food Sci Technol. 2016; 4(6), 160-167.
43. dos Santos TPR, Franco CML, Mischan MM, de Souza Fernandes D, DelBem MS, Leonel M. Production of partially gelatinised cassava starch: Effects of preheating temperature and starch concentration on physicochemical characteristics during the spray-drying process. Australian J Crop Sci. 2019; 13(9): 1486.
44. Hong J, Zeng X-A, Buckow R, Han Z, Wang M-S. Nanostructure, morphology and functionality of cassava starch after pulsed electric fields assisted acetylation. Food Hydrocoll. 2016; 54: 139-150.
45. Wongsagonsup R, Pujchakarn T, Jitrakbumrung S, Chaiwat W, Fuongfuchat A, Varavinit S, Dangtip S, Suphantharika M. Effect of cross-linking on physicochemical properties of tapioca starch and its application in soup product. Carbohydr Polym. 2014; 101, 656-665.
46. Zhang X, Tong Q, Zhu W, Ren F. Pasting, rheological properties and gelatinisation kinetics of tapioca starch with sucrose or glucose. J Food Eng. 2013; 114(2), 255-261.
47. Zhao Y, Saldaña MD. Hydrolysis of cassava starch, chitosan and their mixtures in pressurised hot water media. J Supercrit Fluids. 2019; 147, 293-301.
48. Charoenkul N, Uttapap D, Pathipanawat W, Takeda Y. Physicochemical characteristics of starches and flours from cassava varieties having different cooked root textures. LWT - Food Sci Technol 2011; 44(8): 1774-1781.

49. Li W, Gao J, Wu G, Zheng J, Ouyang S, Luo Q, Zhang G. Physicochemical and structural properties of A-and B-starch isolated from normal and waxy wheat: Effects of lipids removal. Food Hydrocoll. 2016; 60: 364-373.
50. Charles AL, Sriroth K, Huang T. Proximate composition, mineral contents, hydrogen cyanide and phytic acid of 5 cassava genotypes. Food Chem. 2005; 92(4): 615-620.
51. Singh J, Dartois A, Kaur L. Starch digestibility in food matrix: a review. Trends Food Sci Technol. 2010; 21(4), 168-180.
52. Alcázar-Alay SC, Meireles MAA. Physicochemical properties, modifications and applications of starches from different botanical sources. Food Sci Technol. 2015; 35: 215-236.
53. Guo K, Zhang L, Bian X, Cao Q, Wei C. A-, B-and C-type starch granules coexist in root tuber of sweet potato. Food Hydrocoll. 2020; 98: 105279