



## Influence of Soaking and Sprouting on the Colour Kinetics of White Sorghum Grains

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**Abstract:** Colour is a critical sensory quality parameter reflecting the final quality of various food grain products. This study investigated the impact of different soaking durations (6, 12, 18, and 24 hours) and sprouting periods (24, 36, 48, 60, and 72 hours) on the color degradation kinetics of white sorghum grains. Colour parameters such as lightness ( $L^*$ ), redness/greenness ( $a^*$ ), yellowness/blueness ( $b^*$ ), total colour change ( $\Delta E$ ), and browning index (BI) were measured. These experimental values were fitted to four selected models (Zero-order, First-order, Power, and Quadratic) using MATLAB R2019a's non-linear regression tools. Comparing the soaked and sprouted samples, the soaked grains exhibited superior colour characteristics, with higher lightness values that are favourable for milling, whereas the sprouted samples showed increased redness and browning index values. The results indicated that the Quadratic equation best described the color changes in both soaked and sprouted sorghum samples.

**Key words:** Sorghum, color kinetics, soaking, sprouting.

Cereals are a vital food crop, providing essential daily carbohydrates. In developing countries, they are often considered staple foods. Among various cereal crops, sorghum (*Sorghum bicolor*) has gained popularity due to its diverse applications in brewing, nutraceuticals, cosmeceuticals, pharmaceuticals, and biofuel production. According to a United States Department of Agriculture (USDA) report (Williams *et al.*, 2023), the United States leads global sorghum production, with a projected 61.02 million tons for 2023-2024. In India, sorghum ranks third after wheat and rice, primarily grown in Maharashtra, Karnataka, and Rajasthan. Its resilience and drought tolerance make it ideal for arid regions, addressing food security and providing nutrition and income for communities facing environmental challenges.

Sorghum is a nutritious, gluten-free option. It is rich in protein (Duodu *et al.*, 2003), vitamins like niacin, riboflavin, and thiamine (Martino *et al.*, 2012), and minerals such as calcium, magnesium, phosphorus, iron, copper, and potassium (Anglani, 1998). Sorghum benefits those with celiac disease, wheat allergies (Pontieri *et al.*, 2013), diabetes (Hargrove *et al.*, 2011), certain cancers (Thakur and Tiwari, 2019), qualifying it

as a functional food (Frankowski *et al.*, 2022). However, sorghum contains anti-nutritional factors like polyphenolic compounds (tannins) and cyanogenic glycosides (dhurrin), which can impede nutrient absorption (Etuk *et al.*, 2012). Fortunately, processing techniques such as soaking, cooking, sprouting, and fermentation can reduce these compounds through enzyme formation (Nkhata *et al.*, 2018).

The growing concern over health issues like celiac disease, gluten intolerance, and hyperactivity allergies has led to increased interest in sorghum as a key ingredient in various food products. These include porridge (Cisse *et al.*, 2018), pasta (Palavecino *et al.*, 2019), noodles (Liu *et al.*, 2012), bread (Sharanagat and Nema, 2023), cakes (Marston *et al.*, 2015), cookies (Chiremba *et al.*, 2009), powdered drink mixes (Queiroz *et al.*, 2018), beer (Garzon *et al.*, 2019), and ready-to-drink sorghum beverages (Kiptanui *et al.*, 2022), often replacing or partially substituting wheat flour. Beyond nutritional value, food color and appearance significantly influence consumer choice. Several factors control sorghum grain color and, subsequently, the color of value-added products.

Genotype is a primary factor, and understanding sorghum kernel structure is essential to comprehend this effect. A sorghum grain comprises the embryo (germ), endosperm, and pericarp (outer cover). The pericarp is subdivided into epicarp, mesocarp, and endocarp, where color-controlling genes are concentrated, resulting in red, white, lemon-yellow, or brown color variants. Pericarp thickness, pigmented testa presence, endosperm color, and texture also contribute to grain color. Environmental factors like weathering, humidity, rain, insect attacks, and mold can alter grain color due to sunlight exposure and polyphenolic compound migration from glumes.

Processing treatments such as soaking (Moses *et al.*, 2022), germination (Wulandari *et al.*, 2020), malting (Khoddami *et al.*, 2017), fermentation (Paliwal and Sharma, 2023) can remove undesired colored compounds, mainly concentrated in the pericarp layer. However, conflicting reports exist on the impact of processing methods on color-controlling compounds. This study investigates

the effect of different processing treatments, specifically soaking and sprouting at various time combinations, on sorghum grain color characteristics, aiming to inform the development of value-added sorghum products.

## Materials and Methods

### *Collection of sorghum sample*

Sorghum grains were procured from the local market of Sarisha, South 24-Paraganas, West Bengal for conducting this experiment. The grains were cleaned manually to remove foreign particles such as; dirt, dust, broken and immature grains before starting the experiments.

### *Preparation of soaked samples*

Sorghum grains each weighing 500 g were soaked separately at different time intervals such as 6 hrs, 12 hrs, 18 hrs, and 24 hrs respectively (Chauhan *et al.*, 2022) using tap water at room temperature. After soaking, the grains were transferred to a muslin cloth to remove excess water after draining the water used for soaking. The grains were then kept for drying in a tray dryer (Narang scientific works NSW-148) at 50°C for 10 to 12 hrs until constant moisture content was achieved. Weight of the grains before and after drying was recorded using a weighing balance.

### *Preparation of sprouted samples*

For sprouting, 500 g of sorghum grains were soaked in tap water (12 hours), then drained and covered with a damp muslin cloth. The covered grains were then kept in an incubator (Bharat Scientific BOD336) at 35±2°C for different sprouting time intervals i.e. 24 hrs, 36 hrs, 48 hrs, 60 hrs, and 72 hrs respectively (Chauhan *et al.*, 2022). Water was sprayed on the grains while keeping them in the incubator for effective sprouting. The sprouted grains were then kept in the tray dryer at 50°C for 10-12h for drying until constant moisture content was achieved. The weight of the grains before and after drying was recorded using a weighing balance.

### *Determination of moisture Content*

Moisture content was determined according to the standard procedure of AOAC (1990). Weighed sample (5 g) of finely ground material

is kept in a dried and pre-weighed petridish and dried in a hot air oven at 105°C and cooled in a desiccator. The process of heating and cooling is repeated till a constant weight is obtained. Cooled petridish with dried material is then weighed. Usually, this process may take 5-6 hours depending on the moisture content in the sample.

$$\text{Moisture (\% w.b)} = \frac{\text{Loss in weight}}{\text{Weight of sample}} \times 100 \quad \dots 1$$

#### Determination of color parameters

Color measurements of different soaked and sprouted samples of sorghum grains were carried out using a Hunterlab Miniscan EZ Spectrophotometer based on  $L^*$ ,  $a^*$  and  $b^*$  values. Before using the instrument, calibration was performed against a standard black and white colored reference tile. Small transparent plastic pouches containing the sorghum samples were brought close to the viewport and the readings displayed on the display were recorded. The readings were recorded for other replications done by changing different view sides of the pouches containing samples and the average reading was taken for the analysis.  $L^*$  value gives the values of lightness and darkness ranging from 0-100 indicating dark to light,  $a^*$  value gives the redness-greenness value with a higher  $a^*$  value showing redder,  $b^*$  value gives the yellowness-blueness color, where a higher  $b^*$  value indicating more yellow. Other color change determination parameters such as the total color change ( $\Delta E$ ), and Browning index (BI) were evaluated from the  $L^*$ ,  $a^*$ , and  $b^*$  values obtained from the instrument to describe the change in color during soaking and sprouting of sorghum grains at different periods. Previous studies of hunter color parameters ( $L^*$ ,  $a^*$ , and  $b^*$ ) have found significant and useful in describing color deterioration for evaluating quality control in foods (Wulandari *et al.*, 2020).

$$\Delta E = \sqrt{(L_o - L^*)^2 + (a_o - a^*)^2 + (b_o - b^*)^2} \quad \dots 2$$

$$BI = \frac{100(x-0.31)}{0.17} \quad \dots 3$$

where;

$$x = \frac{(a^*+1.75L^*)}{(5.645L^*+a^*-0.3012b^*)} \quad \dots 4$$

$L_o$ ,  $a_o$ , and  $b_o$  are initial controlled samples without any treatment;  $L^*$ ,  $a^*$ , and  $b^*$  are the readings for the soaked and sprouted samples.

#### Colour kinetics modelling

To determine the color change parameters of sorghum grains as a function of soaking and sprouting time, zero-order, first-order, power, and quadratic model equations were used since in most food samples, the time factor dependence could be best described by these equations.

The rate of change of a quality factor C is expressed as

$$\frac{dC}{dt} = kC^n \quad \dots 5$$

where k - kinetic rate constant; C - concentration of a quality factor at any time; n - reaction order.

Integrating equation 5, the following zero order and first order reaction equation can be expressed as

$$C = C_o - k_o t \quad \dots 6$$

$$C = C_o \exp(-k_1 t) \quad \dots 7$$

where C and  $C_o$  - measured final and initial color parameters ( $L^*$ ,  $a^*$ ,  $b^*$ ,  $\Delta E$ , BI); t - time of soaking or sprouting (h);  $k_o$  and  $k_1$  - zero and first-order kinetic constant.

The power model equation is expressed as

$$C = at^b \quad \dots 8$$

The quadratic model equation is expressed as

$$C = a + bt + ct^2 \quad \dots 9$$

where t - soaking or sprouting time (h); C - color changes parameters ( $L^*$ ,  $a^*$ ,  $b^*$ ,  $\Delta E$ , BI); a, b, and c - constants of the model.

#### Statistical analysis

MATLAB R2019a software was used for performing the regression analysis. Response parameters such as the coefficient of determination ( $R^2$ ), and the roots mean square error (RMSE) were used to determine the best-fit equation that can describe the color change. Higher values of  $R^2$  (close to 1) and lower values of RMSE (close to 0) indicate better fit. These parameters can be expressed as

$$R^2 = 1 - \frac{\sum_{i=1}^N (y_{pre,i} - y_{exp,i})^2}{\sum_{i=1}^N (\bar{y}_{pre} - y_{exp,i})^2} \quad \dots 10$$

$$RMSE = \left[ \frac{1}{N} \sum_{i=1}^N (y_{exp,i} - y_{pred,i})^2 \right]^{1/2} \quad \dots 11$$

where,  $y_{exp,i}$  - experimental color change parameter;  $y_{pred,i}$  - predicted color change parameter;  $\bar{y}_{exp,i}$  - mean of the experimental color change parameter;  $\bar{y}_{pre}$  - mean of predicted color change parameter; N - number of readings.

## Results and Discussion

### *Effect of soaking and sprouting on the moisture content*

Moisture content plays an important role in determining the quality of the final products such as the appearance/color and the texture. The average moisture content (% w. b) plots of both the dehydrated soaked and sprouted sorghum samples are shown in Fig. 1. An increasing trend of moisture content was observed for the soaked samples with the increase in soaking time. Similar findings were reported with the other grains such as wheat, millet, and maize (Malik *et al.*, 2021), and sorghum (Shingote *et al.*, 2021). For the sprouted samples, moisture content was high during the initial stage but later showed a decreasing trend as the sprouting time increases. This decrease in moisture content is in agreement with the findings reported in sprouted samples of amaranth and quinoa (Beniwal *et al.*, 2019), green gram and ragi seeds (Shingote *et al.*, 2021), fenugreek (Atlaw and Kumar, 2018). Both the soaking and sprouting

process affects the moisture content of the grains as a result of absorption of water, activation of enzymes, respiration, growth, metabolism, decomposition, etc. (Ai and Ballo, 2010).

### *Effect of soaking and sprouting on the color characteristic*

The variation was observed of different color parameters for dehydrated soaked and sprouted sorghum samples at different time intervals (Fig. 2 and 3). The  $L^*$  value for soaked and sprouted samples showed a slightly decreasing trend ranging from 73.06-75.5 and 60.5-73.45 respectively. Comparing the soaked and sprouted samples, the lightness value  $L^*$  decrease was higher in the sprouted sample which may be associated with the reduction in moisture content (Batariuc *et al.*, 2023) and non-enzymatic browning reactions (Dueik *et al.*, 2010). The  $a^*$  values for the soaked sample ranged from 3.33-3.78 and 4.3-5.9 for sprouted samples which showed that more redness was noticed during sprouting treatment which may be ascribed to leaching of phenolic compounds such as tannin from the pericarp and testa of sorghum grain (Olamiti *et al.*, 2020). The  $b^*$  values for both the soaked and sprouted samples showed a decreasing trend ranging from 17.71-23.4 and 22.59-24.29, respectively as compared with the control sample but no significant difference was observed. The BI for the soaked sample showed a decreasing trend ranging from 5.66-6.67 but increased for the sprouted sample ranging from 7.48-10.67 which is in line with the findings reported by Shingare and Thorat (2014) for dehydrated sprouted wheat grains. The total color changes were found to increase for both the soaked (5.2-10.29) and sprouted samples (2.66-15.80) which may be attributed to the increased water diffusion into

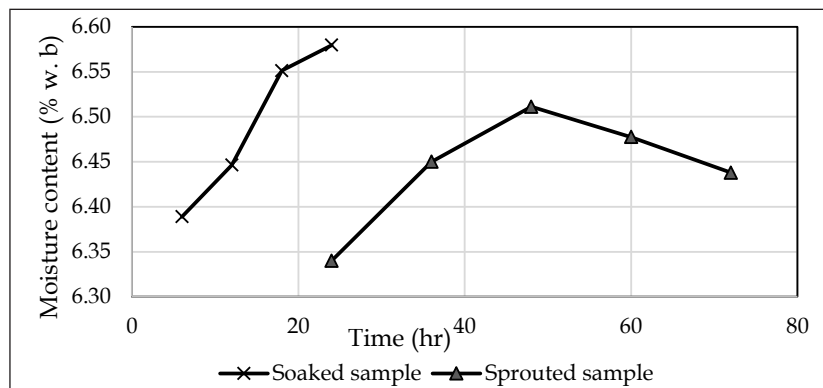


Fig. 1. Variation of moisture content of dehydrated soaked and sprouted sorghum samples at different time intervals.

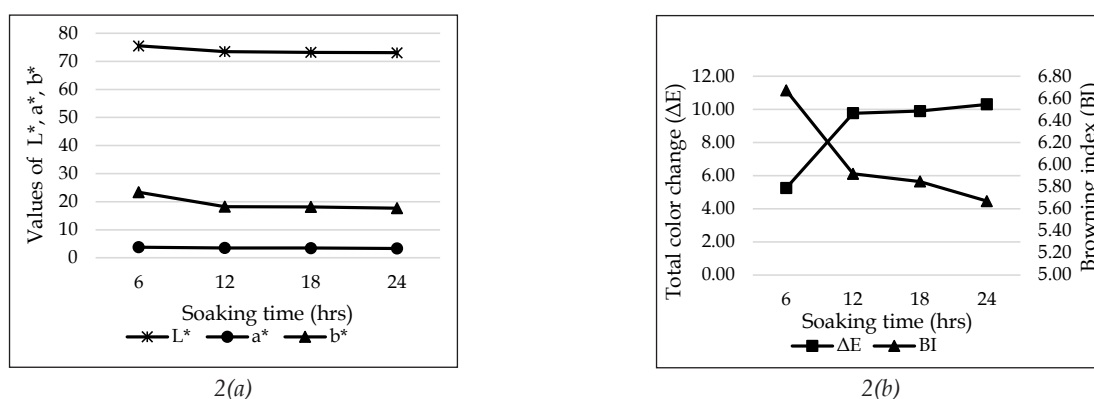


Fig. 2(a). Plot of  $L^*$ ,  $a^*$ ,  $b^*$  values for different soaking time; 2(b). Plot of total color change and browning index for different soaking time.

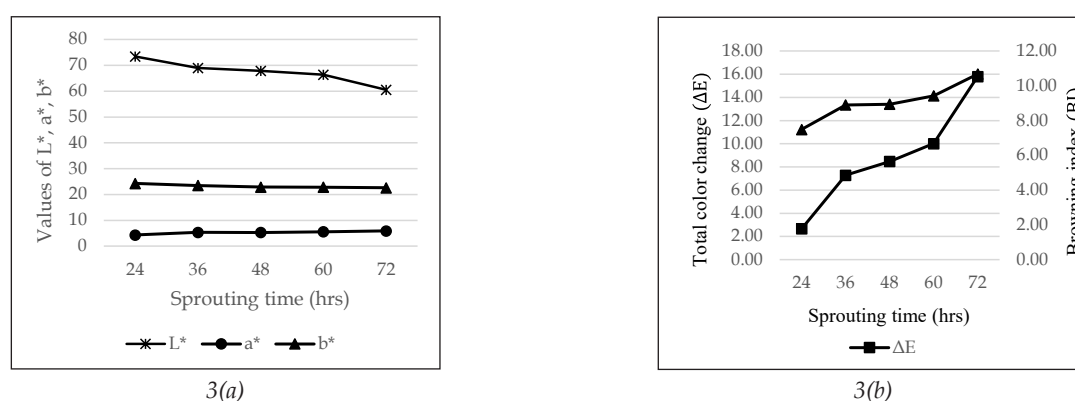


Fig. 3(a). Plot of  $L^*$ ,  $a^*$ ,  $b^*$  values for different sprouting time; 3(b). Plot of total color change and browning index for different sprouting time.

the voids, and partial gelatinization of starch. However, the color change impact was higher in the sprouted sample due to the occurrence of more biochemical process and polyphenol leaching with the increasing sprouting time (Taylor and Duodo, 2015).

#### Determination of best-fit model equation

Experimental color parameters ( $L^*$ ,  $a^*$ ,  $b^*$ ,  $\Delta E$ , BI) were fitted to the above four selected equations for describing the color intensity changes after soaking (6-24 hours) and sprouting (24-72 hours) of sorghum grains using non-linear regression curve fitting tools. Table 1 shows the different constants, coefficients, and statistical parameter values for soaked and sprouted sorghum samples. We can observe that  $R^2$  values for  $L^*$  range from 0.737-0.966,  $a^*$  ranges from 0.951-0.987,  $b^*$  ranges from 0.569-0.933,  $\Delta E$  ranges from 0.628-0.936, and BI ranges from 0.804-0.946. Higher  $R^2$  and lower values of RMSE in all the color parameters except  $a^*$  were found in the quadratic equation fitting which indicates that the color deterioration in the case of soaked

sorghum samples can be best described by it. Higher values of  $R^2$  and lower values of RMSE for  $a^*$  were found in the power equation for the soaked sample. Similarly, Table 1 shows the different constants, coefficients, and statistical parameter values for sprouted sorghum samples. We can observe that  $R^2$  values for  $L^*$  ranged from 0.879-0.933,  $a^*$  (0.813-0.893),  $b^*$  (0.873-0.981),  $\Delta E$  (0.929-0.934), and BI (0.901-0.906). Higher  $R^2$  and lower values of RMSE for all the color parameters except in  $\Delta E$  and BI were found in the quadratic equation which indicates that the color deterioration in the case of sprouted sorghum samples can be best described by it in this case also. Higher  $R^2$  and lower RMSE were found for both the  $\Delta E$  and BI in the first-order equation which shows that the browning and total color changes could be best described for sprouted samples. Given the significance of lightness value in producing various value-added products from both soaked and sprouted sorghum grains, the quadratic model appears to be the most effective equation for representing the grain's color degradation.

Table 1. Different constant and statistical parameter values for soaked and sprouted sorghum grains sample

Sample	Zero-order				First order				Power				Quadratic				
	C <sub>o</sub>	K <sub>o</sub>	R <sup>2</sup>	RMSE	C <sub>o</sub>	K <sub>1</sub>	R <sup>2</sup>	RMSE	a	b	R <sup>2</sup>	RMSE	a	b	c	R <sup>2</sup>	RMSE
Soaked																	
L*	75.7	0.12	0.73	0.71	75.7	0.001	0.74	0.71	78.5	-0.02	0.88	0.46	78.0	-0.52	0.01	0.96	0.36
a*	3.88	0.02	0.95	0.05	3.90	0.006	0.95	0.04	4.42	-0.08	<b>0.98</b>	0.02	4.02	-0.04	0.00	0.97	0.04
b*	23.6	0.28	0.67	1.86	24.32	0.01	0.56	1.76	33.39	-0.21	0.86	1.20	29.5	-1.26	0.03	0.93	1.20
ΔE	6.30	-0.3	0.68	1.86	7.30	-0.02	0.62	2.12	3.89	0.39	0.78	1.61	0.15	1.32	-0.03	0.93	1.24
BI	6.79	0.05	0.80	0.24	6.85	0.008	0.82	0.23	8.14	-0.11	0.93	0.13	7.51	-0.17	0.00	0.94	0.17
Sprouted																	
L*	78.86	0.23	0.92	1.48	79.68	0.003	0.92	1.53	118.2	-0.01	0.88	1.88	75.6	-0.08	-0.00	0.93	1.71
a*	3.90	-0.02	0.83	0.28	4.07	-0.00	0.81	0.29	2.04	0.24	0.88	0.23	2.79	0.08	-0.00	0.89	0.27
b*	24.63	0.03	0.87	0.28	24.89	0.001	0.87	0.27	29.85	-0.06	0.95	0.16	26.5	-0.11	-0.00	0.98	0.13
ΔE	-2.75	-0.24	0.92	1.46	2.09	-0.02	<b>0.93</b>	1.42	0.04	1.37	0.93	1.42	-0.0	0.11	-0.00	0.93	1.71
BI	6.32	-0.05	0.90	0.40	6.66	-0.00	<b>0.90</b>	0.40	3.07	0.28	0.90	0.41	6.44	0.05	0.00	0.90	0.49

## Conclusion

Color determination is an important step that helps in anticipating the end-product color quality of sorghum grains. For example, white sorghums are more generally preferred for porridge making, tortillas, and non-gluten-baked products whereas red-colored sorghums are generally preferred for brewing traditional beer. Experimental result shows that almost all of the color parameter values show a decreasing trend on the soaked and sprouted samples due to physio-chemical changes involved during the treatment. However, the redness and browning index values were found to increase with the sprouted sample which is not desirable as compared with the soaked sample that provides better lightness suitable for flour milling purposes which may be utilized as a composite with other refined flour for various food preparation. Moreover, both the soaking and sprouting treatments affect the moisture content of the dehydrated samples which may impact the final product quality such as the appearance/color. Out of the four model equations, the color degradation kinetics could be best described by the quadratic equation. Thus, this study could provide a basic idea about the effect of the pre-milling processes (especially soaking and sprouting) of sorghum grains on different quality end products where the incorporation of sorghum flour in food is involved.

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