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**Chirag Prajapati**Dairy Engineering Division,  
ICAR-National Dairy  
Research Institute, Karnal,  
Haryana, India**Amandeep Singh**Dairy Technology Division,  
ICAR-National Dairy  
Research Institute, Karnal,  
Haryana, India**Vikram**Dairy Technology Division,  
ICAR-National Dairy  
Research Institute, Karnal,  
Haryana, India**Anup Mishra**Department of Food  
Technology, Parul Institute of  
Applied Sciences, Parul  
University, Vadodara,  
Gujarat, India**Amrita Tigga**Department of Food and  
Nutritional Sciences,  
University of Reading,  
Reading, United Kingdom**Corresponding Author:****Amrita Tigga**Department of Food and  
Nutritional Sciences,  
University of Reading,  
Reading, United Kingdom

## Development and characterisation of sunflower-lotus seed enriched dark chocolate

**Chirag Prajapati, Amandeep Singh, Vikram, Anup Mishra and Amrita Tigga**

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### Abstract

Dark compound chocolate is widely consumed due to its desirable sensory attributes and enhanced processing robustness resulting from the partial or complete replacement of cocoa butter with vegetable fats. However, conventional chocolate products remain energy-dense and provide limited dietary fibre and high-quality plant protein. In this study, a functional dark compound chocolate enriched with sunflower (*Helianthus annuus*) and lotus (*Nelumbo nucifera*) seed powders was developed using a ratio-based formulation strategy, wherein the total seed solids were held constant to isolate the effect of ingredient balance. Sunflower and lotus seeds were roasted (120–130 °C, 8–10 min), milled, and sieved (100 µm) prior to incorporation into molten dark compound chocolate. Three formulations (T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>) were prepared by varying the sunflower-to-lotus seed ratio while maintaining a fixed total seed addition of 24 g per 100 g product and a constant chocolate base of 76 g. Sensory evaluation was conducted using a 9-point hedonic scale (n = 45 panellists, three sessions). Proximate and micronutrient composition of the optimised formulation was analysed in triplicate, along with microbiological quality and production cost estimation. Among the formulations, T<sub>2</sub> (12 g sunflower: 12 g lotus per 100 g) achieved significantly higher scores (p<0.05) for appearance, aroma, taste, texture, and overall acceptability. The optimised chocolate exhibited enhanced nutritional value (per 100 g): moisture 3.31%, protein 5.97 g, carbohydrates 65.00 g (sugars 21.19 g), fat 25.17 g, dietary fibre 3.96 g, potassium 385.45 mg, calcium 90.25 mg, and vitamin E 3.70 mg. Microbiological analysis confirmed acceptable hygienic quality (TPC 780 cfu/g; yeast and mould not detected). The estimated production cost was ₹ 32.22 per 100 g, dominated by raw material costs. Overall, maintaining constant total seed solids demonstrated that optimisation of ingredient ratio, rather than increased enrichment level, is critical for improving nutritional quality while preserving sensory acceptability and processing feasibility in seed-enriched dark compound chocolate systems.

**Keywords:** Dark compound chocolate, sunflower seed, lotus seed, sensory evaluation, nutritional enrichment

### 1. Introduction

Dark chocolate and cocoa-based products have attracted considerable scientific and consumer interest owing to their characteristic flavour and association with bioactive compounds such as flavonoids and methylxanthines. These constituents, particularly epicatechin and catechin, have been linked to antioxidant, anti-inflammatory, cardiovascular, and cognitive health benefits in clinical and epidemiological studies (Jalil & Ismail, 2008; Hooper *et al.*, 2012; Desideri *et al.*, 2012; Magrone *et al.*, 2017) <sup>[10, 9, 5, 15]</sup>. Despite these attributes, conventional chocolate products remain energy-dense and typically provide limited dietary fibre and high-quality plant protein, restricting their contribution to improved diet quality. Dark compound chocolate differs from conventional chocolate in that cocoa butter is partially or fully replaced with compatible vegetable fats, resulting in reduced tempering sensitivity and improved processing robustness. These characteristics make compound chocolate particularly suitable for experimental formulation studies and small-scale production, where minor compositional changes can otherwise lead to fat crystallisation defects and sensory inconsistency (Beckett, 2019) <sup>[2]</sup>.

In response to growing demand for “healthier indulgence,” recent research has focused on enriching chocolate matrices with nutrient-dense plant ingredients while preserving consumer acceptability (Latif, 2013; Di Mattia *et al.*, 2013) <sup>[14, 7]</sup>.

From a food engineering perspective, chocolate is a fat-continuous suspension system in which solid particles are dispersed within a continuous lipid phase. Its rheology, melting behaviour, and mouthfeel are highly sensitive to particle size distribution, solid volume fraction, and fat-solid interactions (Beckett, 2019; Goya *et al.*, 2022) [2, 8]. The incorporation of non-cocoa solids can therefore increase viscosity, disrupt melt smoothness, and induce sensory defects such as grittiness if not carefully controlled.

Sunflower seeds (*Helianthus annuus*) represent a promising functional ingredient due to their high content of unsaturated fatty acids, dietary fibre, minerals, and tocopherols, particularly vitamin E, along with phenolic compounds and phytosterols associated with antioxidant and cardiometabolic benefits (Kasote *et al.*, 2015; Rehman *et al.*, 2021) [12, 18]. In addition, sunflower seed proteins exhibit favourable techno-functional properties, including emulsification and water-holding capacity, which are relevant in composite food systems (Karamać *et al.*, 2012; Vidosavljević *et al.*, 2025) [11, 24]. Lotus seeds (*Nelumbo nucifera*) are nutritionally rich and traditionally consumed in Asian diets, providing carbohydrates, plant protein, dietary fibre, and minerals such as potassium and calcium (Mehta *et al.*, 2013; Saeed *et al.*, 2025) [16, 19]. Their bioactive phytochemicals have been associated with antioxidant, anti-inflammatory, and neuroprotective effects, while their functional properties may influence texture and stability in formulated foods. Although the individual nutritional attributes of sunflower seeds, lotus seeds, and cocoa products are well documented, their combined application in chocolate systems has received limited attention. Moreover, many functional chocolate studies vary both total enrichment level and ingredient ratio simultaneously, making it difficult to isolate the effect of compositional balance. Addressing this gap, the present study applies a ratio-based formulation strategy in which total seed solids are held constant while the sunflower-to-lotus ratio is systematically varied. This approach enables attribution of changes in sensory and nutritional characteristics to ingredient balance rather than to increased solid loading. Accordingly, this study aimed to develop and characterise a sunflower-lotus seed enriched functional dark compound chocolate with improved nutritional value and consumer acceptability, while maintaining processing feasibility through controlled formulation design.

## 2. Materials and Methods

### 2.1 Raw Materials

Sunflower seeds (*Helianthus annuus*) and lotus seeds (*Nelumbo nucifera*) were procured from local agricultural markets in Bharuch, Gujarat, India. Prior to processing, seeds were manually sorted to remove broken kernels, foreign matter, and visibly defective grains in order to ensure uniform raw material quality. Commercial-grade dark compound chocolate was obtained from a reputed manufacturer to ensure consistent fat composition, melting behaviour, and flavour profile across all formulations. The use of dark compound chocolate (containing cocoa solids and vegetable fat substitutes) was selected to improve process robustness and reduce sensitivity to minor formulation-induced changes in fat crystallisation during laboratory-scale processing. As a compound chocolate was used, tempering was not applied; controlled cooling was used to ensure uniform setting. No external binding agents,

emulsifiers, stabilisers, or preservatives were added in any formulation. All raw materials were stored in airtight containers under ambient laboratory conditions ( $25 \pm 2$  °C) until further use to prevent moisture uptake and oxidative deterioration.

### 2.2 Preparation of Seed Powders

Sunflower and lotus seeds were thoroughly washed with potable water to remove adhering dust and impurities, followed by air drying at ambient temperature ( $28 \pm 2$  °C) to eliminate surface moisture. The cleaned seeds were then subjected to light dry roasting at 120-130 °C for 8-10 min in a preheated hot-air oven to reduce moisture and develop roasted flavour notes, following standard practices reported for edible oilseeds and starch-rich seeds (Kasote *et al.*, 2015; Saeed *et al.*, 2025) [12, 19]. Roasting conditions were carefully controlled to prevent excessive browning and to minimise degradation of heat-sensitive bioactive components, particularly tocopherols and phenolic compounds.

After roasting, the seeds were cooled to room temperature (25 °C) and ground using a laboratory-scale grinder to obtain fine powders (as shown in Fig. 1 and Fig. 2). The ground material was passed through a 100 µm sieve to standardise particle size and reduce perceived grittiness. The prepared seed powders were stored in airtight, light-resistant containers at ambient temperature (25-30 °C) under dry conditions and used within 48 h to minimise oxidative changes prior to chocolate formulation.



Fig 1: Sunflower seed powder (100-µm sieved)



Fig 2: Lotus seed powder (100-µm sieved)

### 2.3 Formulation of Dark Compound Chocolate Enriched with Sunflower and Lotus Seeds

Dark compound chocolate was melted using the double-boiling method at 48-50 °C, ensuring indirect heating to prevent thermal degradation, fat separation, or scorching of

the cocoa solids, in accordance with standard chocolate processing guidelines (Beckett *et al.*, 2019) [2]. The temperature was continuously monitored using a calibrated digital thermometer and maintained below 50 °C to preserve flavour integrity and ensure uniform melting of the fat phase. Once the chocolate was completely melted and achieved a smooth, homogeneous consistency, pre-weighed quantities of sunflower seed powder and lotus seed powder were gradually incorporated into the molten chocolate under continuous manual stirring at 45-48 °C. Mixing was continued for 5-7 min to facilitate uniform dispersion of seed particles within the fat continuous chocolate matrix and to avoid particle agglomeration or sedimentation. Maintaining the mixture temperature above 45 °C during incorporation ensured adequate flowability and prevented premature solidification.

The homogenised chocolate mixture was immediately poured into pre-cleaned moulds maintained at ambient temperature (25-28 °C) and gently tapped to remove entrapped air bubbles and promote uniform surface finish. The filled moulds were then allowed to pre-set at room temperature for 10-15 min, followed by controlled setting under refrigeration at 8-10 °C for 30-40 min to achieve complete solidification and structural stability. After refrigeration, samples were equilibrated to room temperature before wrapping to minimise condensation related surface defects. After setting, the chocolates were demoulded and wrapped in aluminium foil to protect against light and oxidative deterioration. The finished products were stored in a cool, dry environment (25 ± 2 °C), protected from light, to minimise fat bloom and oxidative deterioration until analyses.

#### 2.4 Formulation Design

A ratio-based formulation strategy was employed to isolate the effect of sunflower-to-lotus seed balance on product quality. Three experimental formulations (T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub>) were developed by varying the relative proportions of sunflower and lotus seed powders while maintaining a fixed total seed powder content of 24 g per 100 g sample. The dark compound chocolate content was kept constant at 76 g per formulation, resulting in a total sample weight of 100 g for each treatment.

This approach ensured that changes in sensory, nutritional, and processing characteristics could be attributed to ingredient ratio rather than to differences in total solid loading or chocolate base composition.

**Table 1:** Ingredient composition of experimental formulations (per 100 g)

Ingredient	T <sub>1</sub> (g)	T <sub>2</sub> (g)	T <sub>3</sub> (g)
Sunflower seed powder	16	12	8
Lotus seed powder	8	12	16
Dark compound chocolate	76	76	76
Total	100	100	100

Preliminary bench-scale trials conducted prior to final formulation selection indicated that increasing total seed powder content beyond 24 g per 100 g sample resulted in excessive viscosity, reduced melt smoothness, and inferior mouthfeel due to increased solid volume fraction. Conversely, lower seed incorporation levels produced minimal nutritional enhancement. Therefore, a fixed seed addition level was selected, and only the sunflower-to-lotus

ratio was varied to identify an optimal balance between functional enrichment and sensory quality. This formulation logic is consistent with reported strategies for composite and functional chocolate systems, where control of total solid content is critical for maintaining desirable rheological and sensory properties (Latif, 2013) [14].

#### 2.5 Sensory Evaluation

Sensory evaluation was conducted using a 9-point hedonic scale, where 9 corresponded to “like extremely” and 1 corresponded to “dislike extremely” (Stone *et al.*, 2020). A semi-trained sensory panel comprising 45 panellists (aged 20-45 years), including faculty members, postgraduate students, and food industry professionals familiar with chocolate products, participated in the evaluation. Samples (5 g) were served at 25 ± 2 °C, and panellists provided informed consent and reported no known ingredient allergies.

The attributes assessed included appearance, aroma, taste, texture, and overall acceptability. Chocolate samples from each formulation were coded with random three-digit numbers and presented in a randomised order to minimise positional and order bias. Evaluations were conducted under controlled ambient conditions with uniform lighting and temperature. Panellists were instructed to rinse their mouths with potable water between samples to avoid carryover effects. Each formulation was evaluated across three independent sessions. For statistical analysis, individual panellist scores were retained (n = 45) and session-wise data were pooled after confirming that session did not significantly influence scores.

#### 2.6 Proximate and Nutritional Analysis

The proximate nutritional composition of the optimised formulation (T<sub>2</sub>) was determined in triplicate using standard analytical procedures to ensure analytical accuracy and reproducibility. Moisture content of the chocolate samples was determined by the hot-air oven drying method, wherein approximately 2.0 ± 0.1 g of sample was weighed into pre-dried, tared aluminium dishes and dried at 105 ± 2 °C to constant weight, following the procedures described by Kreiser and Martin Jr. (1977) [13] and AOAC International (2000) [1]. Constant weight was confirmed when two successive weighings, after cooling in a desiccator, differed by less than 1 mg.

Ash content was determined by incinerating 2.0 ± 0.1 g of the dried chocolate sample in a pre-weighed crucible at 550 °C in a muffle furnace for 4-6 h, until a light grey residue and constant weight were obtained, in accordance with AOAC-recommended procedures (AOAC International, 2000) [1]. Crude fat content was extracted using a Soxhlet apparatus with petroleum ether as the solvent. Protein content was determined by the Kjeldahl method, applying a nitrogen-to-protein conversion factor of 6.25. Total carbohydrate content was calculated by difference using the formula: Carbohydrate (%) = 100 - (moisture + protein + fat + ash). Total sugars were estimated using Fehling's solution titration method following standard protocols for confectionery products. Dietary fibre content was analysed by the enzymatic-gravimetric method in accordance with IS 11062:1984 (Bureau of Indian Standards).

For micronutrient analysis, calcium was quantified by AAS, and potassium by flame photometry after wet digestion. Vitamin E (α-tocopherol) was quantified using HPLC and

expressed as mg per 100 g of sample (Dandin *et al.*, 2023) [4].

## 2.7 Microbiological Analysis

To assess the microbiological safety of the optimised dark compound chocolate formulation (T<sub>2</sub>), total plate count (TPC) and yeast and mould count were evaluated. Chocolate samples were aseptically homogenised, and appropriate serial dilutions were prepared using sterile diluent following standard microbiological practices. Total plate count was determined by plating serial dilutions on nutrient agar using the pour plate method, followed by incubation at 37 °C for 48 h. Yeast and mould counts were carried out by inoculating diluted samples onto potato dextrose agar (PDA) and incubating at 28 °C for 72 h. After incubation, colonies were enumerated and results were expressed as colony-forming units per gram (cfu/g) of sample.

These analyses were performed to confirm the hygienic quality and microbiological safety of the developed functional chocolate. The inherently low moisture content and fat-rich nature of dark compound chocolate are known to restrict microbial growth; nevertheless, microbiological evaluation was essential to verify that seed incorporation did not compromise product safety (Beckett *et al.*, 2019) [2].

## 2.8 Cost Analysis

The economic feasibility of the sunflower-lotus seed enriched dark compound chocolate was evaluated using a bottom-up cost estimation approach, representative of laboratory-scale to small-batch production. The analysis incorporated the cost of raw materials, including dark compound chocolate, sunflower seeds, and lotus seeds, along with packaging costs (aluminium foil), overheads, GST (as applicable), and a profit margin of 10%, based on prevailing local market prices at the time of the study. GST was considered as applicable, and the final cost shares are presented in Fig. 8.

Since both the dark compound chocolate content (76 g per 100 g sample) and the total seed powder content (24 g per 100 g sample) were kept constant across all formulations, raw material costs remained comparable among treatments. The cost analysis was conducted to assess the commercial viability of the optimised formulation (T<sub>2</sub>) relative to premium and functional chocolate products currently available in the market.

## 2.9 Statistical Analysis

Sensory evaluation data were expressed as mean scores. For each attribute, individual panellist scores (n = 45) were used for ANOVA, with formulation as the fixed factor; session was treated as a repeat assessment and pooled after confirming no session effect ( $p>0.05$ ). Statistical analysis was performed using R statistical software (R Foundation for Statistical Computing, Vienna, Austria) through the R-Studio integrated development environment. Prior to inferential analysis, assumptions of parametric testing were verified. Normality of residuals was assessed using the Shapiro-Wilk test, and homogeneity of variances among formulations was evaluated using Levene's test in R. As all datasets satisfied these assumptions, parametric analysis was considered appropriate. A one-way analysis of variance (ANOVA) was applied to evaluate the effect of sunflower-to-lotus seed ratio on individual sensory attributes, including appearance, aroma, taste, texture, and overall acceptability,

at a significance level of  $p<0.05$ . When significant differences were detected, Tukey's Honestly Significant Difference (HSD) test was employed for post hoc pairwise comparisons among formulation means.

Nutritional and microbiological analyses were performed in triplicate, and results are reported as mean values to ensure analytical reliability and reproducibility. Session-to-session variability was addressed by conducting three sessions and pooling session-wise data after confirming no significant session effect ( $p>0.05$ ).

## 3. Results and Discussion

### 3.1 Product Development and Formulation Strategy

Formulation T<sub>2</sub> (as shown in Fig. 3) was identified as the optimised formulation, as it consistently exhibited the highest mean sensory scores across all evaluated attributes, namely appearance, aroma, taste, texture, and overall acceptability (Fig. 4). Compared with the sunflower-dominant (T<sub>1</sub>) and lotus-dominant (T<sub>3</sub>) formulations, T<sub>2</sub> demonstrated superior consumer perception, together with enhanced nutritional value, satisfactory microbiological quality, and a favourable cost structure. This combination of attributes establishes T<sub>2</sub> as the most practically viable formulation for further development.

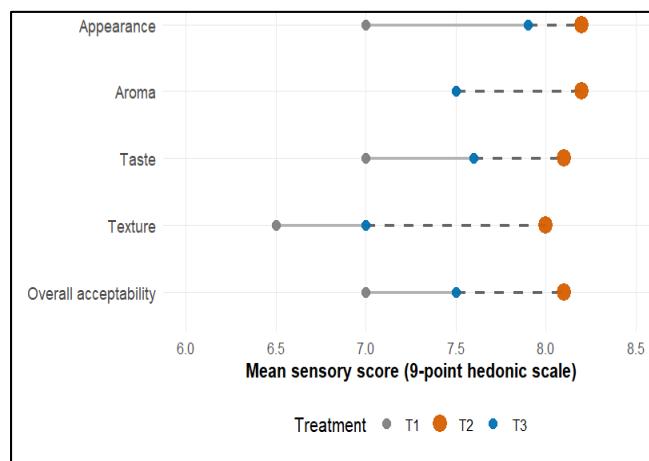


**Fig 3:** Optimised sunflower-lotus seed enriched dark compound chocolate (T<sub>2</sub>)

Three formulations (T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>) were developed using a ratio-based formulation design, wherein the total seed powder incorporation level was fixed at 24 g per 100 g of product and the dark compound chocolate content was maintained constant at 76 g per formulation. Only the sunflower-to-lotus seed ratio was systematically varied to isolate compositional effects on product quality while eliminating confounding variations in total solid loading and chocolate base composition. This controlled formulation approach is particularly critical in composite chocolate systems, as non-cocoa solids can strongly influence the fat-continuous microstructure, apparent viscosity, particle-fat interactions, and sensory perception when total solids are altered simultaneously (Latif, 2013; Goya *et al.*, 2022) [14, 8]. During processing, all formulations were successfully prepared using conventional melting and moulding techniques, confirming the feasibility of seed incorporation at the selected enrichment level. However, clear formulation-dependent differences were observed in mixing behaviour and mould filling characteristics. The sunflower-dominant formulation (T<sub>1</sub>) exhibited relatively higher apparent viscosity and reduced flowability, consistent with stronger solid-fat structuring effects associated with higher

levels of sunflower seed fibre and protein fractions. In contrast, the lotus-dominant formulation (T<sub>3</sub>) showed comparatively smoother handling behaviour, which may be attributed to the higher carbohydrate and starch content of lotus seeds exerting a weaker structuring effect on the fat phase.

Notably, the balanced formulation (T<sub>2</sub>) exhibited the most favourable processing behaviour, characterised by uniform particle dispersion, acceptable flowability, and smooth surface finish after setting. These processing advantages were reflected in the sensory outcomes (Fig. 4), where T<sub>2</sub> consistently outperformed both T<sub>1</sub> and T<sub>3</sub> across all attributes. The final optimised dark compound chocolate product (T<sub>2</sub>) is therefore presented as the preferred formulation.



**Fig 4:** Mean sensory scores of formulations (T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>) on a 9-point hedonic scale

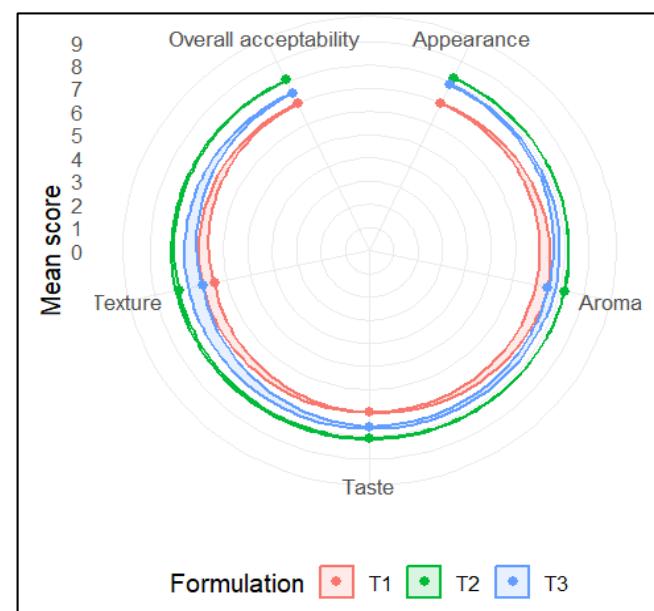
### 3.2 Sensory Evaluation of Chocolate Formulations

The sensory attributes of the sunflower-lotus seed enriched dark compound chocolate formulations (T<sub>1</sub>, T<sub>2</sub>, and T<sub>3</sub>) were evaluated using a 9-point hedonic scale for appearance, aroma, taste, texture, and overall acceptability. Sensory scores were analysed using individual panellist responses pooled across sessions, ensuring a robust representation of panel consensus. As illustrated in the sensory radar plot (Fig. 5), formulation T<sub>2</sub> recorded the highest mean scores across all evaluated attributes, followed by T<sub>3</sub>, while T<sub>1</sub> exhibited comparatively lower scores.

One-way analysis of variance (ANOVA) indicated that formulation type, defined by the sunflower-to-lotus seed ratio, had a statistically significant effect ( $p<0.05$ ) on each sensory attribute evaluated. Post hoc comparisons using Tukey's Honestly Significant Difference (HSD) test confirmed that T<sub>2</sub> scored significantly higher than T<sub>1</sub> for key consumer-relevant attributes, particularly taste, texture, and overall acceptability ( $p<0.05$ ). Although T<sub>3</sub> generally showed intermediate scores between T<sub>1</sub> and T<sub>2</sub>, its sensory performance remained consistently lower than that of the optimised formulation. The superior sensory performance of T<sub>2</sub> can be attributed to its optimised ingredient balance, which resulted in a harmonised nutty-cocoa flavour profile, controlled solid loading, and improved melt smoothness. The radar plot (Fig. 5) highlights this balance through the broader and more uniform sensory profile of T<sub>2</sub> compared with the other formulations.

In contrast, the sunflower-dominant formulation (T<sub>1</sub>) exhibited relatively reduced texture and overall acceptability

scores, consistent with increased perceived grittiness or melt resistance that may arise when higher levels of insoluble non-cocoa solids disrupt the fat-continuous chocolate matrix. The lotus-dominant formulation (T<sub>3</sub>) was perceived as comparatively smoother, but slightly lower scores for aroma and taste suggest that excessive lotus seed incorporation may attenuate the roasted and nutty flavour notes that contribute positively to chocolate sensory complexity. These observed sensory trends reinforce previous findings that functional ingredient incorporation in chocolate must be carefully balanced, as disproportionate enrichment can adversely affect rheology, mouthfeel, and flavour perception (Latif, 2013; Velciov *et al.*, 2021)<sup>[14, 23]</sup>.



**Fig 5:** Radar profile of sensory attributes (T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>)

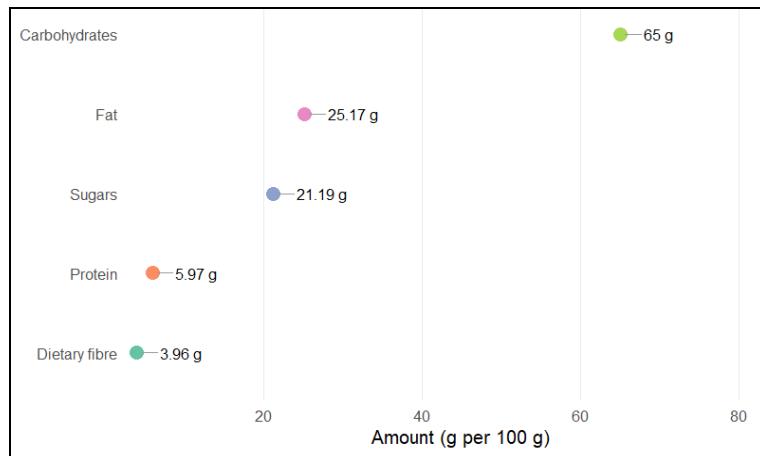
### 3.3 Nutritional Composition of the Optimised Formulation (T<sub>2</sub>)

Nutritional values are expressed per 100 g of product for the optimised formulation (T<sub>2</sub>). The proximate composition and selected micronutrient profile are summarised in Table 2 and visually illustrated in the macronutrient plot (Fig. 6) and micronutrient chart (Fig. 7). As shown in Fig. 6, formulation T<sub>2</sub> exhibited a nutritionally enhanced profile, containing 5.97 g protein, 65.00 g carbohydrates (including 21.19 g sugars), 25.17 g fat, and 3.96 g dietary fibre per 100 g. Sugars are included within total carbohydrates; dietary fibre is reported separately.

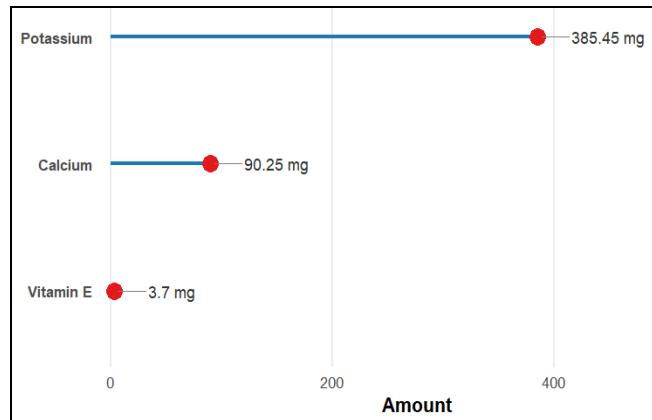
The dot plot representation highlights the dominance of carbohydrates as the major macronutrient, followed by fat, while protein and dietary fibre are present at moderate but nutritionally meaningful levels for a chocolate-based product. The observed protein and fibre contents reflect successful nutritional enrichment through sunflower and lotus seed incorporation, consistent with the inherent nutrient density of these seeds (Kasote *et al.*, 2015; Saeed *et al.*, 2025)<sup>[12, 19]</sup>. The moisture content of T<sub>2</sub> (3.31%) remained within the desirable range for chocolate products, supporting product stability by limiting microbial growth and quality deterioration. This observation is consistent with the inherently low water activity of fat-rich chocolate matrices (Beckett *et al.*, 2019)<sup>[2]</sup>.

Micronutrient composition of the optimised formulation is illustrated in Fig. 7. Potassium was present at 385.45 mg per

100 g, representing the most abundant micronutrient measured, followed by calcium at 90.25 mg per 100 g, while vitamin E ( $\alpha$ -tocopherol) was detected at 3.70 mg per 100 g. The lollipop chart clearly demonstrates the relative dominance of potassium over calcium and vitamin E, emphasising the mineral contribution of lotus seeds and the antioxidant associated tocopherol contribution of sunflower seeds (Dandin *et al.*, 2023)<sup>[4]</sup>.



**Fig 6:** Macronutrient composition of T<sub>2</sub> (per 100 g)



**Fig 7:** Micronutrient content of T<sub>2</sub> (per 100 g)

**Table 2:** Nutritional composition of the optimised chocolate formulation (T<sub>2</sub>) per 100 g

Parameter	Value (per 100 g)
Moisture (%)	3.31
Ash (%)	2.47
Protein (g/100 g)	5.97
Carbohydrates (g/100 g)	65.00
Sugars (g/100 g)	21.19
Fat (g/100 g)	25.17
Dietary fibre (g/100 g)	3.96
Potassium (mg/100 g)	385.45
Calcium (mg/100 g)	90.25
Vitamin E (mg/100 g)	3.70

### 3.4 Microbiological Analysis

The microbiological assessment of the optimised formulation (T<sub>2</sub>) indicated satisfactory microbial quality. The total plate count (TPC) was 780 cfu/g, while yeast and mould were reported as not detected when counts were below the detection limit of the method, indicating good hygienic quality and microbiological safety. These findings demonstrate that the product maintained acceptable hygiene standards during preparation and storage.

Overall, the nutritional data confirm that the ratio-optimised seed incorporation strategy enhances both macro and micronutrient density without excessive fat dilution or moisture increase. When considered alongside the favourable sensory outcomes, these results demonstrate that nutritional enrichment can be achieved while maintaining the technological and sensory integrity of dark compound chocolate systems.

The low microbial load can be attributed primarily to the low moisture content of the formulation, combined with effective processing steps such as seed roasting and chocolate melting, along with hygienic handling during production. Chocolate is inherently characterised by low water activity and high fat content, conditions that restrict microbial proliferation and contribute to shelf stability (Beckett *et al.*, 2019)<sup>[2]</sup>. Therefore, the present findings align with established knowledge on the microbiological safety of chocolate products, indicating that seed enrichment at the selected level does not compromise safety.

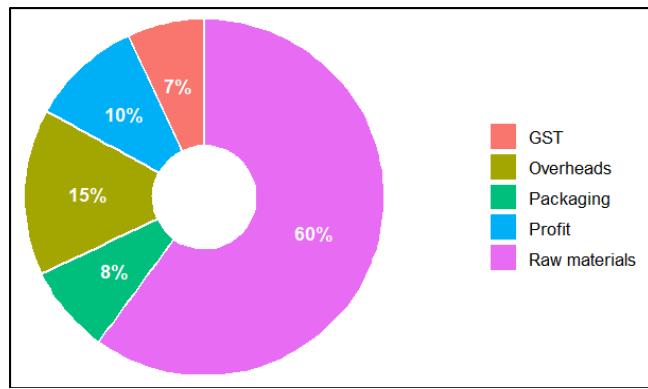
### 3.5 Economic Feasibility and Cost Structure

The economic feasibility of the optimised sunflower-lotus seed enriched dark compound chocolate formulation (T<sub>2</sub>) was evaluated by analysing the contribution of major cost components, including raw materials, packaging, overheads, statutory taxes, and profit margin, based on prevailing local market prices. Since the dark compound chocolate content (76 g per 100 g sample) and the total seed powder content (24 g per 100 g sample) were kept constant across all formulations, cost variation among treatments was negligible. Consequently, the present analysis primarily reflects the baseline cost structure and commercial feasibility of seed-enriched dark compound chocolate.

The estimated production cost of the optimised formulation was ₹ 32.22 per 100 g, inclusive of packaging costs, applicable taxes, overheads, and a standard profit margin. As illustrated in the cost distribution chart (Fig. 8), raw materials constituted the largest cost component (60%), followed by overheads (15%), profit margin (10%), packaging (8%), and goods and services tax (GST) (7%). The dominance of raw material costs is characteristic of chocolate-based products, where cocoa-and fat-based ingredients represent a major share of total production expenditure.

Notably, the incorporation of sunflower and lotus seed powders contributed only a modest fraction of the total raw

material cost, while delivering measurable improvements in nutritional quality. This indicates that functional enrichment through seed incorporation can be achieved without substantial cost escalation, thereby maintaining economic competitiveness. Overall, the cost structure supports the commercial viability of the optimised formulation and suggests potential for market positioning alongside premium and functional chocolate products.



**Fig 8:** Cost share of T<sub>2</sub> (per 100 g)

### 3.6 Integrated Evaluation and Commercial Relevance

An integrated evaluation of sensory performance (Fig. 4 and Fig. 5), nutritional enhancement (Figs. 6 and 7; Table 2), microbiological safety, and economic feasibility clearly supports T<sub>2</sub> as the optimised formulation for sunflower-lotus seed enriched dark compound chocolate. The balanced seed ratio (12 g sunflower: 12 g lotus per 100 g) achieved the best overall outcome by combining strong consumer acceptability with improved nutrient density, safe microbiological status, and a competitive cost profile.

From a product development perspective, the findings confirm that ratio optimisation at fixed total seed inclusion is a practical strategy for functional chocolate innovation. This approach enhances nutritional value without the common negative trade-offs associated with higher total non-cocoa solids, such as excessive viscosity, grittiness, and impaired melt behaviour (Goya *et al.*, 2022) [8]. The use of dark compound chocolate further supports scalability and process robustness, making the formulation suitable for small-scale commercial translation and potential broader functional confectionery applications.

## 4. Conclusion

This study developed a sunflower-lotus seed enriched dark compound chocolate using a ratio-based formulation strategy at a fixed total seed inclusion of 24 g per 100 g, enabling isolation of ingredient balance effects on product quality. Among the formulations tested, T<sub>2</sub> (12 g sunflower: 12 g lotus per 100 g) showed significantly higher sensory acceptability ( $p<0.05$ ) across appearance, aroma, taste, texture, and overall acceptability. The optimised formulation exhibited enhanced nutritional value, providing protein (5.97 g/100 g), dietary fibre (3.96 g/100 g), potassium (385.45 mg/100 g), calcium (90.25 mg/100 g), and vitamin E (3.70 mg/100 g), while maintaining low moisture content (3.31%), acceptable fat levels (25.17 g/100 g), and satisfactory microbiological quality (TPC 780 cfu/g; yeast and mould not detected). The estimated production cost of ₹ 32.22 per 100 g supports economic feasibility. The findings demonstrate that optimisation of ingredient ratio, rather than

increased total seed addition, is critical for achieving nutritional enhancement without compromising sensory quality or processing robustness in chocolate systems. Limitations of the study include the absence of detailed rheological, fat crystallisation, antioxidant, and shelf-life analyses. Future work should therefore focus on these aspects, along with bioactive retention, particle size optimisation, and broader consumer validation, to further support industrial scalability of seed-enriched functional chocolates.

## Statements and declarations

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### Competing Interests

The authors declare that they have no competing interests.

### Author's Contributions

Chirag Prajapati: Conceptualization, Methodology, Writing-original draft

Anup Mishra: Methodology, Writing-editing

Amandeep Singh: Data curation, Review & editing

Vikram: Review & editing

Amrita Tingga: Supervision & Resources

### Consent

We, the authors of this manuscript, give our consent to publish the data and results reported in this manuscript.

### Ethical Approval

Not applicable

### Disclaimer (Artificial intelligence)

Author(s) hereby declare that generative AI technologies were used only for language editing and grammatical refinement during manuscript preparation. The AI tool used was ChatGPT (OpenAI). All scientific content, including study design, experimental work, data analysis, interpretation of results, and conclusions, was entirely generated by the authors, who take full responsibility for the accuracy, originality, and integrity of the manuscript.

### Details of the AI usage are given below

1. AI-assisted language editing and sentence refinement

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