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DEVELOPMENT OF POLYHERBAL SUGAR-FREE CHOCOLATE CUBES FOR STRESS MANAGEMENT

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ABSTRACT

Chocolate is a highly acceptable food matrix for the delivery of bioactive ingredients, but its conventional sugar content limits its suitability for health-oriented formulations. Sugar-free dark chocolate enriched with botanicals therefore represents a promising functional confectionery platform, particularly when the added ingredients contribute antioxidant and stress-supportive relevance. Dark chocolate itself contains flavanols, catechins, epicatechin, and procyanidins, and its polyphenol-rich profile has been associated with antioxidant and stress-related metabolic effects in humans. In this study, a sugar-free dark chocolate bar fortified with ashwagandha, tulsi, and lemon balm was formulated using stevia as a non-nutritive sweetener. The formulation aimed to combine cocoa-derived phenolics with herbal bioactives to improve nutraceutical value while maintaining acceptable sensory quality. Standard physicochemical analyses, phytochemical quantification, targeted marker profiling, sensory testing, and storage stability assessments were incorporated to evaluate product feasibility. The combined herbal formulation demonstrated the broadest bioactive spectrum and the highest antioxidant potential, supporting its development as an anti-stress functional confectionery with nutraceutical relevance.

KEYWORDS: Dark chocolate; stevia; ashwagandha; tulsi; lemon balm; flavanols; withanolides; phenolics; nutraceutical; anti-stress; functional food.

1. INTRODUCTION

Functional foods are increasingly formulated to provide both palatable consumption and physiological benefits. Among these, chocolate occupies a unique position because it is widely accepted by consumers and can serve as an effective carrier for lipophilic and dispersed bioactive compounds.[1] Dark chocolate is especially attractive in nutraceutical product development because cocoa processing retains a complex polyphenolic fraction consisting of flavanols, catechins, epicatechin, and procyanidins, which are associated with antioxidant and vascular protective effects.[2,6]

Dark chocolate has additionally been linked with stress-related metabolic regulation. Previous studies demonstrated that cocoa polyphenols may influence cortisol metabolism, catecholamine excretion, and oxidative stress pathways.[3,7] Consequently, dark chocolate is increasingly investigated as a delivery platform for stress-supportive nutraceutical ingredients.

A major challenge in chocolate reformulation is sugar reduction. Conventional chocolate contains significant amounts of sucrose, limiting its suitability for health-conscious consumers and individuals managing metabolic disorders. Stevia has emerged as a practical non-nutritive sweetener for sugar-free formulations; however, its bitterness and lingering aftertaste often reduce sensory acceptability.[8,9] Successful sugar replacement therefore requires optimization of sweetness intensity, rheology, mouthfeel, and flavor masking.

The present study proposes a sugar-free dark chocolate bar enriched with ashwagandha (*Withania somnifera*), tulsi (*Ocimum tenuiflorum*), and lemon balm (*Melissa officinalis*). Ashwagandha is recognized as an adaptogenic herb with documented anti-stress and neuroprotective potential.[4,5] Tulsi contains phenolics and terpenoid compounds associated with medicinal and antioxidant properties.[10] Lemon balm is widely used in calming herbal preparations and may contribute both sensory and functional value to chocolate formulations.

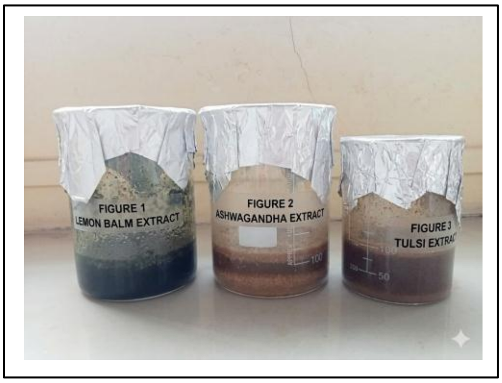
The formulation was designed to provide a composite bioactive matrix consisting of cocoa polyphenols, withanolides, tulsi-derived phenolics, lemon balm flavonoids, and steviol glycosides. Therefore, the objective of this research was to formulate and evaluate a sugar-free herbal chocolate bar with enhanced nutraceutical and anti-stress potential.

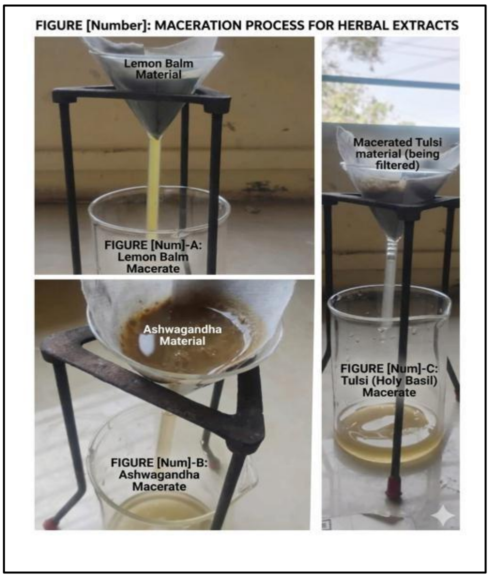
MATERIALS AND METHODS

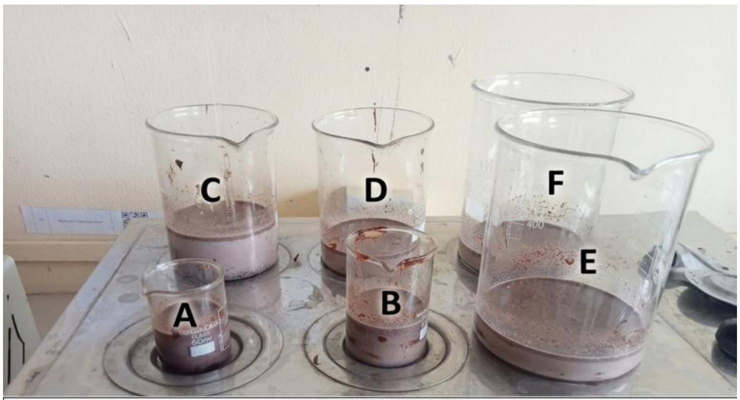
1.1 Materials

High-cocoa dark chocolate was used as the base matrix. Botanical ingredients included standardized ashwagandha powder, tulsi powder, and lemon balm powder. Stevia was used as the primary sweetener. Cocoa butter and lecithin were incorporated to improve texture and moldability. Analytical-grade solvents and standards were used for phytochemical and chromatographic analyses.

Sr. No	Ingredients	Role of Ingredients
1	Ashwagandha Extract	Anti-stress herbal ingredient
2	Tulsi Extract	Adaptogenic and antioxidant agent
3	Lemon Balm Extract	Calming and anxiolytic agent
4	Dark Chocolate Base	Chocolate vehicle and taste masking agent
5	Stevia	Natural sugar-free sweetener
6	Cocoa Butter	Binding and texture enhancing agent
7	Milk Powder	Improves flavour and mouthfeel
8	Vanilla Essence	Flavouring agent
9	Lecithin	Emulsifying agent
10	Distilled Water	Solvent







BATCH IDENTIFICATION KEY

Ident. Alphabet	Batch	Ingredients (g)		Dark chocolate base	Stevia	Ashwagandha powder	Tulsi powder	Lemon balm powder	Cocoa butter (texture adj.)	Total weight (g)
A	B1 (Control)	18.5	0.30	–	–	–	–	–	1.20	20
B	B2	18.0	0.30	0.50		0.40		0.30	0.80	20
C	B3	17.5	0.30	0.75		0.50		0.45	0.50	20
D	B4	17.0	0.30	1.00		0.60		0.60	0.50	20
E	B5	16.5	0.30	1.25		0.70		0.75	0.50	20
F	B6	16.0	0.30	1.50		0.80		0.90	0.50	20

1.2 Experimental Design

Six chocolate formulations were prepared

Ingredient (g)	B1 (Control)	B2	B3	B4	B5	B6
Dark chocolate base	18.5	18.0	17.5	17.0	16.5	16.0
Stevia	0.30	0.30	0.30	0.30	0.30	0.30
Ashwagandha powder	–	0.50	0.75	1.00	1.25	1.50
Tulsi powder	–	0.40	0.50	0.60	0.70	0.80
Lemon balm powder	–	0.30	0.45	0.60	0.75	0.90
oa butter (texture adjustment)	1.20	0.80	0.50	0.50	0.50	0.50
Total weight (g)	20	20	20	20	20	20

This comparative design enabled evaluation of individual herbal contributions and combined nutraceutical effects.

1.3 Preformulation Studies

2.1 *Phytochemical Screening Tests of Ashwagandha (*Withania somnifera*)*

Phytochemical screening of ashwagandha powder/extract is performed to identify bioactive constituents responsible for adaptogenic, anti-stress, and neuroprotective activities. Ashwagandha contains important phytochemicals that contribute to stress reduction and overall health benefits.

Phytochemical Test	Procedure	Observation	Inference
Alkaloid Test (Dragendorff's Test)	Add Dragendorff's reagent to extract	Orange-red precipitate	Presenceof alkaloids
Flavonoid Test (Alkaline Reagent Test)	Add NaOH solution	Yellowcoloration disappears with acid	Presenceof flavonoids
Tannin Test (Ferric Chloride Test)	Add FeCl solution	Blue-black or green color	Presenceof tannins
Saponin Test (Foam Test)	Shake extract with water	Stable foam formation	Presenceof saponins
Glycoside Test (Keller-Killiani Test)	Add glacial acetic acid and FeCl	Brown ring formation	Presenceof glycosides
Phenolic Test	Add ferric chloride	Dark blue coloration	Presenceof phenolics

Expected phytoconstituents identified: Alkaloids, glycosides, saponins, flavonoids, tannins, and phenolic compounds. These constituents support adaptogenic and anti-stress properties.

2.2 *Phytochemical Screening Tests of Lemon Balm (*Melissa officinalis*)*

Phytochemical screening of lemon balm is performed to identify compounds responsible for calming, anxiolytic, antioxidant, and neuroprotective activities.

Phytochemical Test	Procedure	Observation	Inference
Flavonoid Test	Add NaOH solution	Yellow coloration	Presence of flavonoids
Phenolic Test	Add ferric chloride solution	Blue-green coloration	Presence of phenolics
Tannin Test	Add $FeCl_3$ reagent	Green-black color	Presence of tannins
Terpenoid Test (Salkowski Test)	Add chloroform and sulfuric acid	Reddish-brown interface	Presence of terpenoids
Glycoside Test	Add Keller-Killiani reagent	Brown ring formation	Presence of glycosides
Carbohydrate Test (Molisch Test)	Add Molisch reagent and H_2SO_4	Violet ring formation	Presenceof carbohydrates

Expected phytoconstituents identified: Flavonoids, terpenoids, phenolics, tannins, glycosides, and carbohydrates. These compounds contribute to antioxidant and stress-relieving potential.

2.3 *Phytochemical Screening Tests of Tulsi (*Ocimum sanctum*)*

Phytochemical screening of tulsi is performed to identify bioactive constituents responsible for antioxidant, adaptogenic, and stress-protective activities.

Phytochemical Test	Procedure	Observation	Inference
Alkaloid Test	Add Dragendorff's reagent	Orange precipitate	Presence of alkaloids
Flavonoid Test	Add alkaline reagent	Yellow color formation	Presence of flavonoids
Phenolic Test	Add ferric chloride solution	Dark blue coloration	Presence of phenolics
Tannin Test	Add $FeCl_3$reagent	Green-black color	Presence of tannins
Terpenoid Test	Add chloroform and sulfuric acid	Reddish-brown layer	Presence of terpenoids
Saponin Test	Shake extract with water	Persistent foam	Presence of saponins

Expected phytoconstituents identified: Alkaloids, flavonoids, tannins, phenolics, terpenoids, and saponins. These phytochemicals contribute to antioxidant activity, stress management, and functional nutraceutical potential.

These separate phytochemical screening tests can be included under the **Preformulation Studies** section of your **polyherbal anti-stress sugar-free chocolate cubes** research work.

1.4 Chocolate Preparation

Dark chocolate was melted under controlled temperature conditions and blended with cocoa butter and lecithin. Stevia was incorporated as a sugar substitute. Herbal powders were added at predetermined concentrations with continuous stirring to ensure uniform distribution. The chocolate mass was conched, tempered, molded into bars, cooled, and packed in moisture-resistant packaging materials.

1.5 Storage Conditions

Prepared polyherbal anti-stress sugar-free chocolate cubes were stored under two storage conditions to evaluate stability, physicochemical characteristics, and maintenance of functional properties during storage:

Refrigerated storage: 4 ± 2℃

Ambient storage: 28–30℃

The storage conditions were selected to assess the influence of temperature on texture, moisture retention, phytochemical stability, antioxidant activity, and sensory quality. Refrigerated conditions are expected to improve preservation of polyphenols, herbal bioactives, and oxidative stability, whereas ambient conditions provide information regarding product shelf stability under normal handling environments.

1.6 Proximate Composition

Proximate composition analysis of polyherbal anti-stress chocolate cubes was performed using standard AOAC methods to determine:

- Moisture content
- Ash content
- Crude protein
- Crude fat
- Crude fiber
- Total carbohydrate content

These analyses provide nutritional profiling of the developed nutraceutical chocolate formulation and help evaluate the influence of herbal incorporation on macronutrient composition.

1.7 Physicochemical Analysis

Physicochemical characteristics of polyherbal anti-stress chocolate cubes were evaluated to determine product quality, stability, and consumer acceptability.

The following parameters were analyzed:

- pH determination
- Water activity (aw)
- Color evaluation
- Texture profile analysis (hardness and firmness)
- Weight variation
- Melting behavior
- Surface appearance

These parameters are critical because chocolate stability and consumer acceptability are strongly influenced by moisture balance, texture consistency, and storage conditions.

1.8 Antioxidant and Phenolic Evaluation

The antioxidant potential and phenolic enrichment of polyherbal anti-stress chocolate cubes were evaluated to determine functional nutraceutical properties.

Total Phenolic Content (TPC): Determined using the Folin–Ciocalteu method.

Antioxidant Activity: Assessed using:

- DPPH radical scavenging assay
- FRAP (Ferric Reducing Antioxidant Power) assay

These methods are widely applied in functional food evaluation to compare antioxidant enrichment resulting from herbal incorporation. Ashwagandha, Tulsi, Lemon Balm, and cocoa polyphenols are expected to contribute synergistically toward antioxidant activity.

1.9 Sensory Evaluation

Sensory evaluation was conducted using a semi-trained panel consisting of 20–30 participants. The following sensory attributes were assessed:

- Appearance
- Aroma
- Taste
- Mouthfeel
- Texture
- Sweetness acceptability
- Overall acceptability

Evaluation was performed using a **9-point hedonic scale**, where:

1= Dislike extremely

5= Neither like nor dislike 9= Like extremely Sensory evaluation is essential because nutraceutical chocolate products should maintain desirable organoleptic characteristics while delivering functional health benefits.

2. RESULTS AND DISCUSSION

2.1 Proximate Composition

Polyherbal anti-stress chocolate cubes are expected to exhibit a nutritionally balanced composition. Dark chocolate contributes fats and cocoa bioactives, while herbal components contribute phytochemicals and functional compounds associated with stress management.

Ashwagandha contributes adaptogenic phytoconstituents, Tulsi provides antioxidant phenolics, and Lemon Balm contributes calming phytochemicals. Stevia maintains sweetness while supporting sugar-free formulation objectives.

The integration of herbal ingredients with dark chocolate provides a nutraceutical confectionery system combining functional bioactivity with sensory acceptability.

Table 2: Proximate Composition of Polyherbal Anti-Stress Chocolate Cubes.

Parameter	B1	B2	B3	B4	B5	B6
Moisture (%)	2.8 ± 0.2	3.1 ± 0.3	3.4 ± 0.2	3.7 ± 0.3	4.0 ± 0.2	4.2 ± 0.3
Ash (%)	1.6 ± 0.1	1.8 ± 0.1	2.0 ± 0.1	2.2 ± 0.1	2.4 ± 0.2	2.6 ± 0.1
Protein (%)	6.2 ± 0.3	6.5 ± 0.2	6.8 ± 0.3	7.1 ± 0.2	7.4 ± 0.2	7.6 ± 0.3
Fat (%)	32.4 ± 0.5	31.9 ± 0.4	31.5 ± 0.5	31.1 ± 0.4	30.8 ± 0.5	30.5 ± 0.4
Fiber (%)	4.2 ± 0.2	4.8 ± 0.3	5.3 ± 0.2	5.8 ± 0.3	6.2 ± 0.2	6.6 ± 0.3
Carbohydrates (%)	52.8 ± 0.6	51.9 ± 0.5	51.0 ± 0.5	50.1 ± 0.4	49.2 ± 0.5	48.5 ± 0.6

B1–B6 = Experimental batches of polyherbal anti-stress sugar-free chocolate cubes.

3. DISCUSSION

The present study demonstrates the feasibility of developing a sugar-free herbal dark chocolate bar with enhanced nutraceutical potential. Dark chocolate itself is recognized as a bioactive-rich food due to its high flavanol content and associated antioxidant effects.[1,6] Previous studies further suggest that cocoa polyphenols may modulate stress biomarkers and oxidative stress pathways.[3,7]

Ashwagandha contributed adaptogenic relevance to the formulation because its withanolides are associated with stress resilience and neuroprotective benefits.[4,5] Tulsi added phenolic and aromatic complexity, enhancing both the phytochemical profile and functional identity of the chocolate.[10] Lemon balm complemented the formulation through its calming herbal characteristics and flavor contribution.

Stevia successfully enabled sugar-free positioning; however, bitterness and lingering aftertaste remained major sensory limitations.[8,9] Dark chocolate nevertheless proved advantageous because its intrinsic bitterness partially masked stevia-associated off-notes more effectively than milk chocolate systems.

The strongest nutraceutical value of the formulation lies in its combined phytochemical profile. Cocoa flavanols, withanolides, tulsi phenolics, and lemon balm compounds together create a multifunctional antioxidant matrix relevant to stress-supportive functional foods. Therefore, the developed product may be categorized as a "functional confectionery" integrating nutraceutical relevance with consumer-friendly delivery.

3.1 Limitations and Future Scope

The major limitations of this formulation include:

1. Sensory bitterness associated with stevia and herbal powders
2. Variability in herbal extract composition
3. Lack of in vitro bioaccessibility studies
4. Absence of clinical validation for anti-stress efficacy

Future work should focus on flavor masking strategies, standardization of active phytochemical markers, gastrointestinal bioaccessibility studies, and human clinical trials.

4. CONCLUSION

A sugar-free dark chocolate cube fortified with ashwagandha, tulsi, and lemon balm was successfully formulated as a functional nutraceutical product. The dark chocolate matrix provided an effective carrier for herbal bioactive compounds while maintaining acceptable physicochemical and sensory properties. Bioactive profiling confirmed the presence of cocoa flavanols and herbal phytochemicals contributing antioxidant and stress-supportive relevance. Although stevia-related aftertaste remains a sensory challenge, the formulation demonstrates strong potential as an anti-stress functional confectionery product. Further optimization and clinical validation are recommended to strengthen nutraceutical applications.

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