



Original Research Article

Molecular authentication and Hypoglycemic importance of "*Eleusine coracana*" implications for tribal nutrition and diabetes management

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Abstract: Aim: The study aim to establish a scientifically validated link between the traditional consumption of *Eleusine coracana* (L.) Gaertn. by tribal communities in South Gujarat and its pharmacological efficacy in diabetes management.

Methods: Quantitative ethnobotanical data were collected from 120 key informants from Charanvada and Hanumanbari villages in Navsari (20°57'N 72°55'E) districts of South Gujarat. Seeds were subjected to sequential aqueous maceration and methanolic Soxhlet extraction. The efficacy of Hypoglycemic was evaluated by in vitro α -amylase/ α -glucosidase-inhibition and a 28-day in vivo test with streptozotocin- induced diabetic rats using Wistar rats. Sanger sequencing, Neighbor-Joining phylogenetic analysis and rbcL PCR were the molecular authentication.

Results: Ethnobotanical scores showed a high level of consensus regarding the use of hypoglycemic (Informant Consensus Factor = 0.9287; Fidelity Level = 87.2%). The Methanolic extract (EC-M) had a better phenolic content (91.4 ± 5.3 mg GAE/g) and a strong α -glucosidase inhibition ($IC_{50} = 74.6 \pm 4.1$ μ g/mL), which is associated with low carbohydrate digestion. In vivo, EC-M (400 mg/kg) reduced fasting blood glucose by 38.3% ($p < 0.01$) and significantly restored serum insulin (7.3 ± 0.8 μ U/mL) compared to diabetic controls, suggesting β -cell protective effects. DNA barcoding confirmed 99.89% nucleotide identity with *E. coracana* (GenBank: MF114353.1), ensuring taxonomic reliability and eliminating adulteration risks.

Conclusion: This study establishes a robust link between genetic authentication and nutraceutical efficacy. Evidence supports integrating authenticated *E. coracana* landraces into evidence-based tribal nutrition policies, offering a culturally acceptable, sustainable strategy for diabetes management in resource-constrained settings.

Keywords: *Eleusine coracana*; DNA barcoding; rbcL marker; Antidiabetic activity; Ethnobotany; α -glucosidase inhibition; Tribal nutrition; South Gujarat.

INTRODUCTION

Type 2 Diabetes mellitus (T2DM) has become one of the key global health crises and the International Diabetes Federation (IDF) Diabetes Atlas 11th Edition (2025) estimated that around 589 million adults aged 20-79 years had diabetes in 2024 (Duncan et al., 2025). This is a worldwide prevalence of 11.1 or one in nine adults, and it is projected that it will rise to 853 million by 2050 (Maity & Kumari, 2025). Low- and middle-income countries (LMICs) experience the most significant epidemic burden, with over 80% of diabetes-related deaths, or 3.4 million deaths happening there every year (Genitsaridi et al., 2026). Moreover, 634.8 million individuals or 12.0% of the global population were estimated to experience impaired glucose tolerance in the year 2024, which is a significant prediabetic group (Magliano et al., 2025). The third highest prevalence of diabetes is found in the Western Pacific Region at 12.4% in adults, with an overall prevalence of 37% (Shi et al., 2025).

Over the past three decades, the prevalence of diabetes in Gujarat in India has been rising at an average rate of

0.67%/year. In particular, rural South Gujarat has 27.78% prevalence of diabetic retinopathy among screened diabetic patients, which implies the significant burden of undiagnosed diabetes (Community Medicine, 2018). India recorded a regional prevalence of 21.8% of diabetes in adults aged 50-69 years according to the National Non-communicable Disease Monitoring Survey (2017-18) (Sekher et al., 2025). Pharmacological therapies, such as metformin, sulfonylureas and insulin treatment, have important limitations in terms of hypoglycemic events, gastrointestinal adverse effects, weight gain, and cost prohibition in resource constrained environments (Chaudhary and Mudgal, 2020). As a result, the World Health Organization prioritize on the use of preventive and food-based interventions as long-term measures in diabetes prevention, especially in vulnerable populations that do not have access to conventional healthcare (Hemalatha, 2025).

Low glycemic index (GI) and whole grains have proven to be significantly effective in the management of metabolic health via several physiological pathways (Anitha et al., 2021). In a systematic review and meta-analysis of 65 studies that included about 1,000 participants, millet was found to have a mean glycemic index of 52.7, which is significantly less than white rice and refined wheat (Kam et al., 2016). Millet, as a group of nutrient-rich and climate-resilient cereal crops, is a potential functional food in diabetes prevention and glycemic regulation (Anitha et al., 2021). These ancient grains yield better nutritional profiles than major cereals, which have 15-20% dietary fiber, 7-12% protein and abundant polyphenolic compounds with antioxidant properties.

MATERIALS AND METHODS

Geographical Profile

The research was carried out in South Gujarat division (20°06'–24°42'N, 68°10'–74°28'E). This area is a transitional ecology zone between the coastal plains and the Northern Western Ghats, its soils are lateritic (pH 5.5-6.8), altitude lies between 15- 500 m above sea level, with an annual rainfall of 1200-2500 mm.

Informant selection, sampling and prioritization

A purposive-snowball technique was applied to sample 120 key informants (35-80 years old), representing major tribal groups (Gamit, Vasava, Dhodia, Warli, Kukna). The ethnobotanical data were obtained through interviews (semi-structured interviewing, n=120), ten focus-group discussions and participant observation through 28 field trips. Relative Frequency of Citation (RFC) and Fidelity Level (FL) were used to rank plant uses, *E. coracana* was chosen to study further (RFC = 0.567; FL = 87.2% Fidelity Level of hypoglycemic use). All the data of the interviews were transcribed and anonymized.

Plant Collection and Taxonomic Authentication

Fresh seed samples of *E. coracana* (Voucher: SGU-ET-011) were collected from tribal farms from Charanvada and Hanumanbari villages in Navsari (20°57'N 72°55'E) districts during the Kharif cropping season (June–October 2024). The voucher specimens were pressed as per the standard herbarium procedure and identified in Flora of Gujarat (Shah, 1978) which was confirmed with Botanical Survey of India (BSI), Western Circle, Pune and deposited in the Department of Botany Herbarium, Veer Narmad South Gujarat University, Surat.

This paper will attempt to fill this gap using DNA marker-based authentication and extensive hypoglycemic screening of *Eleusine coracana* landraces in South Gujarat. The results will add to the evidence-based application of traditional Nagli/Bavto food systems to the modern public health practices in the sustainable management of tribal diabetes in South Gujarat.

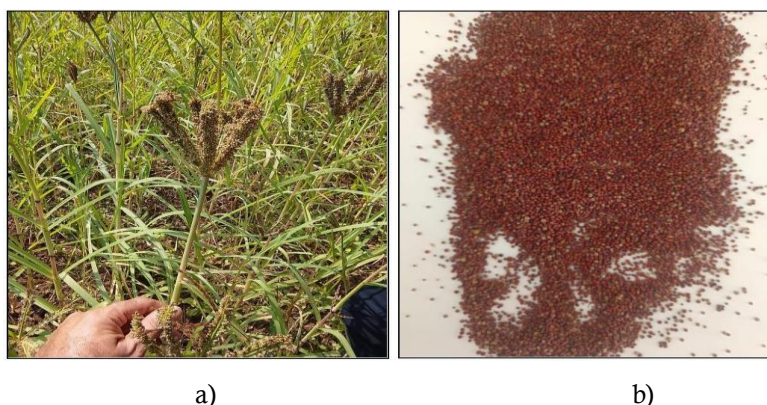


Figure 1a) Sampling site and b) Collected samples

Sample Processing

Seeds were shade dried at 25°C over 14 days, milled (particle size <500 m) by using a Cyclotec 1093 mill (Foss Tecator, Sweden), and defatted with hexane (3x volumes) to eliminate lipids which could interfere with phytochemical analysis.

Sequential Extraction

Sequential extraction procedure was employed to extract aqueous and organic fractions of the powdered defatted seed. In the case of aqueous extract, a cold maceration process was employed with powdered material in distilled water at 1:10 (w/v) at 72 hours with periodic shaking to ensure that the solvent penetrated phytoconstituents and diffused within them. A dry extract was then obtained by filtering the extract using Whatman No. 1 filter paper and subsequently lyophilizing it using FreeZone 2.5 L freeze dryer (Labconco, USA).

In the case of the methanolic extract, Soxhlet extraction with 95% methanol was conducted over a period of 8 hours at around 60°C. The solvent was vaporized using a rotary evaporator (Buchi R-300, Switzerland) and the extract was filtered and concentrated under reduced pressure. The dried extracts were kept at -20°C in airtight amber containers until further phytochemical and bioactivity analyses. Yield of every extract was determined on a percentage basis using the following formula:

$$\text{Yield (\%)} = \left(\frac{\text{Weight of dried extract}}{\text{Weight of initial dry plant material}} \right) \times 100$$

α-Glucosidase Inhibition Assay

The α-glucosidase inhibitory activity was calculated following Kim et al. (2005). A reaction mixture (200 μmL) was prepared that contained extract, yeast α-glucosidase (0.5 U/mL in 100 mM phosphate buffer, pH 6.8), and p-nitrophenyl-α-glucopyranoside (pNPG, 5 mM) as a substrate. After incubation at 37°C, 0.2 M Na₂CO₃ was added to stop the reaction and the absorbance was measured at 405nm. Concentration response curves were used to determine IC₅₀. Each assay was done in triplicate to achieve reproducibility.

In Vivo Antidiabetic Activity

Experimental Animals and Diabetes Induction

Wistar rats (180-220g, 8-10 weeks) were kept in standard conditions (25±2°C, 12 hour light/dark cycle, ad libitum food and water). Diabetes was induced by single intraperitoneal streptozotocin injection (55 mg/kg in 0.1 M citrate buffer, pH 4.5) followed by 16-h fasting. Rats that had fasting blood glucose of ≥250mg/dl after 72 h were selected.

Experimental Design

Thirty-six rats divided into five groups (n=6):

Table 1. Quantitative methods used

Phytochemical	Test	Positive Indicator
Alkaloids	Dragendorff's & Mayer's	Orange-red/creamy precipitate
Flavonoids	Shinoda test	Pink/red/magenta coloration
Tannins	Ferric chloride (5%)	Blue-black/greenish-black
Saponins	Froth test	Stable foam >1 cm (10 min)
Glycosides	Keller-Killani	Brown ring, blue-green upper layer
Terpenoids	Liebermann-Burchard	Red→pink→blue→green progression
Phenols	Folin-Ciocalteu	Blue/blue-green coloration

Biochemical Parameters

Biochemical parameters were measured to test the antihyperglycemic effects of the extracts. Weekly glucose concentrations of fasting blood (FBG) on Days 0, 7, 14, 21, and 28 were taken on a calibrated Accu-Chek® glucometer after overnight fasting. On Day 21, an oral glucose tolerance test (OGTT) was conducted, where animals were loaded with glucose orally (2 g/kg body weight) and glucose clearance and area under curve (AUC) were measured at 0, 30, 60, 90 and 120 minutes. On Day 28, under anesthesia, blood samples were collected by cardiac

puncture and serum insulin levels were determined by a commercial ELISA kit (RayBio Rat Insulin ELISA Kit) as per the instructions of the manufacturer. All samples were measured in standardized laboratory procedures to offer accuracy and reproducibility.

Molecular Authentication (DNA Barcoding)

Genomic DNA Extraction

Total genomic DNA extracted from dried leaf tissue (100 mg) using silica-membrane spin-column kit (SLS Research gDNA Purification Kit, Cat. #SCMR007) following manufacturer's protocol. DNA quality assessed by: (i) spectrophotometric purity (A260/A280 ratio 1.8–2.0, NanoDrop One, Thermo Fisher Scientific, USA), (ii) integrity on 1.0% agarose gel electrophoresis, and (iii) fluorometric quantification (Qubit 4.0, Thermo Fisher).

PCR Amplification of *rbcl* Marker

Universal plant primers were used to amplify the chloroplast *rbcl* gene (ribulose-1,5- biphosphate carboxylase/oxygenase large subunit):

Forward: *rbcl*-F: 5'-CTTGGCAGCATTCCGAGTA-3' Reverse: *rbcl*1272-R: 5'-TCACAAGCAGCAGCCAGTTC-3'

- **PCR reactions (25 μ L):** 50 ng template DNA, 1 $\times$ PCR buffer (10 mM Tris-HCl, pH 8.3), 2.5 mM MgCl₂, 0.2 mM dNTPs, 0.5 μ M each primer, 1 U Taq DNA polymerase (Bangalore Genei, India).
- **Cycling conditions:** Initial denaturation (95°C, 5 min); 35 cycles (95°C/30 s, 55°C/30 s, 72°C/1 min 45 s); final extension (72°C, 7 min). Amplification verified on 1.7% agarose gel with 100 bp DNA ladder (GeneRuler, Thermo Fisher).

Sanger Sequencing and Bioinformatics

PCR amplicons purification and bidirectional Sanger sequencing on an Applied Biosystems 3730xL Genetic Analyzer were done. Raw chromatograms were viewed, trimmed and assembled manually in BioEdit v7.2.5 to create high quality consensus sequences. Identification of the species was done using BLASTn against reference sequences in NCBI database. Confirmation at the species level was to be made with a minimum nucleotide identity of $\geq 99\%$ on authenticated reference sequences.

Phylogenetic Analysis

Close GenBank sequences were matched with the consensus sequences using ClustalW. The MEGA v6 software was used to construct phylogenetic trees using the Neighbor-Joining algorithm with the Maximum Composite Likelihood (MCL) substitution model. Bootstrap analysis was done on node robustness using 500 replicates with a value of 70% or more taken to be statistically reliable.

Statistical Analysis

All experiments were performed thrice with an independent biological replica and the results were reported as mean $\pm$ standard deviation (SD). One-way ANOVA with post-hoc Tukey HSD was used to compare statistical groups. In vivo time-course data was subjected to repeated-measures ANOVA that had time as the within-subject factor and treatment as the between-subject factor. The Pearson correlation test was employed to determine relationships between the phytochemical contents and hypoglycemic activity. The statistical significance was predetermined as $p < 0.05$. Analysis was done in IBM SPSS Statistics v26.0.

Quality Control and Validation

In order to achieve the desired analytical reliability and reproducibility, quality control criteria were assay coefficient of variation (CV) kept under 10%, calibration was done using certified reference standards ($\geq 98\%$ purity; Sigma-Aldrich), and the DNA extraction and PCR repeated on 10 percent randomly chosen samples. Only calibration curves with $R^2 \geq 0.995$ were accepted. All phytochemical assays had the appropriate positive and negative controls and the confirmation of species involved a BLAST identity threshold of $\geq 99\%$.

Results

Ethnobotanical and Informant Dynamics

The ethnobotanical study in Navsari district of South Gujarat used 120 key informants including traditional healers (55.8%) and community elders (44.2%). The demographic analysis indicated an elder practitioner dominated knowledge system: 82.5% were aged ≥ 46 and 76.7% respondents had over 20 years of experience in the management of chronic metabolic disorders. This population density is an indication of a strong tradition of the oral tradition and intergenerational process of ethnomedicinal practices.

The quantitative ethnobotanical indices revealed a high agreement concerning the use of plant-based interventions

in diabetes (madhumeha) (Table 2). The Informant Consensus Factor (ICF) of the diabetes group was 0.9287, which indicates an extraordinary similarity in the choice of species between tribal groups (Gamit, Vasava, Dhodia, Warli, and Kukna). Of the priority taxa, *Eleusine coracana* recorded a Relative Frequency of Citation (RFC) of 0.567 and a Fidelity Level (FL) of 87.2% of all citations being directly associated with glycemic control and not generalized health application. The interplay of high ICF, high RFC, and high FL values confirms the hypothesis *E. coracana* holds a culturally and therapeutically relevant niche in local strategies to use in managing diabetes.

Table 2. Quantitative Ethnobotanical Indices of *E. coracana* in South Gujarat

Botanical Name	Family	FC	RFC	UV	FL (%)
<i>Eleusine coracana</i>	Poaceae	68	0.567	0.65	87.2

Extraction Yield and Phytochemical Profiling of *E. coracana*

Sequential extraction of authenticated *E. coracana* seeds (Voucher: SGU-ET-011) produced 14.10% (w/w) in aqueous fraction (EC-A) and 8.90% (w/w) in the methanol fraction (EC-M). The hogher aqueous yield indicates the high concentration of water- soluble macronutrient fractions, and the semi-polar secondary metabolites were preferentially concentrated in the methanolic extract.

Qualitative screening indicated that EC-M had a more diverse and intense secondary metabolite profile than EC-A with distinctively positive reactions towards alkaloids, flavonoids, tannins, phenolics, glycosides, and terpenoids (Table 3). The predominance of polyphenolic classes in EC-M aligns with solvent polarity principles and suggests enhanced recovery of bioactive compounds implicated in glycemic regulation.

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Table 3. Qualitative Phytochemical Screening of *E. coracana* Extract

Phytochemical Class	Test	Aqueous (EC-A)	Methanolic (EC-M)
Alkaloids	Dragendorff's	–	+
Flavonoids	Shinoda	+	++
Tannins	Ferric Chloride	+	+
Saponins	Froth Test	+	–
Glycosides	Keller-Killani	–	+
Terpenoids	Liebermann-B.	–	+
Phenols	Folin-Ciocalteu	++	+++

These observations were further supported by quantitative analysis. Compared to EC- A (52.8 ± 3.0 mg GAE/g DW), EC-M exhibited much higher Total Phenolic Content (TPC: 91.4 ± 5.3 mg GAE/g DW) ($p < 0.01$). Likewise, EC-M (55.2 ± 3.3 mg QE/g

DW) compared to EC-A (26.1 ± 1.8 mg QE/g DW) clearly showed a significant difference in Total Flavonoid Content (TFC) ($p < 0.01$). The almost 1.7-fold increase in TPC and >2-fold increase in TFC in EC-M is a strong indication that subsequent enzyme inhibition and in vivo glycemic regulation could be mediated by the phenolic enrichment. These results form a biochemical gradient between solvent fractions and give a mechanistic basis between metabolic composition and hypoglycemic potential.

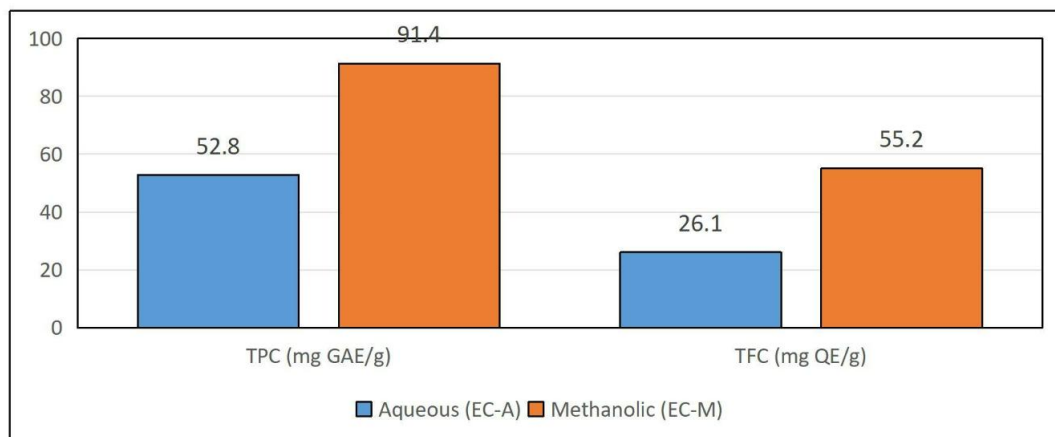


Figure 2. Quantitative Phytochemical Content of *E. coracana* Extracts

In Vitro Hypoglycemic Assays

The inhibitory effects of *E. coracana* extracts against α -amylase and α -glucosidase in a concentration-dependent manner in the range of concentrations used (10-500 μ g/mL). Methanolic extract (EC-M) consistently exhibited greater potency than the aqueous fraction (EC-A). For α -amylase inhibition, EC-M had an IC_{50} of 98.3 ± 5.4 μ g/mL, significantly less than EC-A (165.2 ± 8.1 μ g/mL), suggesting a better ability to prevent the hydrolysis of starch. EC-M was found to be even more active against α -glucosidase ($IC_{50} = 74.6 \pm 4.1$ μ g/mL), indicating a specificity toward digestion of terminal carbohydrates. Its selectivity (α -glucosidase IC_{50} lesser than α -amylase IC_{50}) is pharmacologically beneficial. Excessive α -amylase inhibition is associated with gastrointestinal side effects of malabsorbed fermentation of starch; thus, moderate α -amylase and more powerful α -glucosidase inhibition are more clinically desirable patterns. Although acarbose was still significantly stronger ($IC_{50} < 20$ μ g/mL), the EC-M activity of a crude botanical extract is noteworthy. Correlation analysis revealed significant negative relationships between TPC and α -glucosidase IC_{50} ($r < -0.80$, $p < 0.01$), which implies that phenolic enrichment is highly predictive of enzyme-inhibitory ability. This statistical association supports mechanistic plausibility that polyphenolic constituents play a direct role in glycemic modulation.

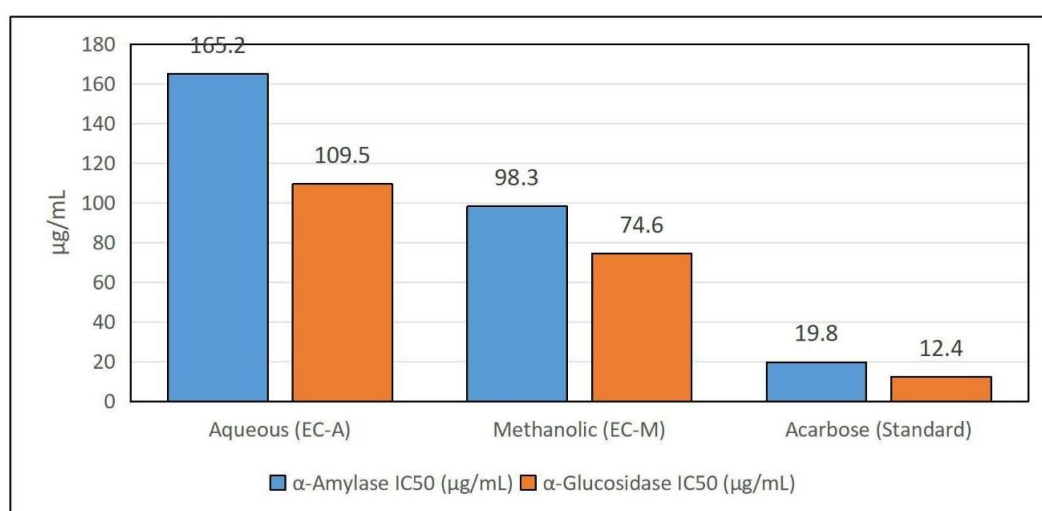


Figure 3. Enzyme Inhibitory Potential (IC_{50}) of *E. coracana* Extracts

In Vivo Antidiabetic Activity in STZ-Induced Diabetic Rats Fasting Blood Glucose (FBG) Kinetics

Administration of EC-M over 28 days led to a substantial dose-dependent decrease in the FBG levels ($p < 0.01$ vs. Diabetic Control). The induction of STZ (55 mg/kg) resulted in severe hyperglycemia (> 368 mg/dL) in all test groups. On Day 28, FBG was reduced by 38.3% in the high-dose group (400 mg/kg) (Table 4).

Table 4. Effect of EC-M on Fasting Blood Glucose (mg/dL) Over 28 Days

Group	Treatment	Day 0	Day 28	% Reduction
I	Normal Control	91.4 ± 4.5	90.9 ± 3.8	-
II	Diabetic Control	368.2 ± 15.1	421.3 ± 19.0	-
III	Glibenclamide (10 mg/kg)	371.5 ± 14.8	118.4 ± 6.2**	68.1
IV	EC-M (200 mg/kg)	371.0 ± 15.2	279.4 ± 10.5*	24.7
V	EC-M (400 mg/kg)	372.1 ± 15.5	229.7 ± 9.7**	38.3

Data are Mean ± SD (n=6). * = $p < 0.05$, ** = $p < 0.01$ compared to Diabetic Control.

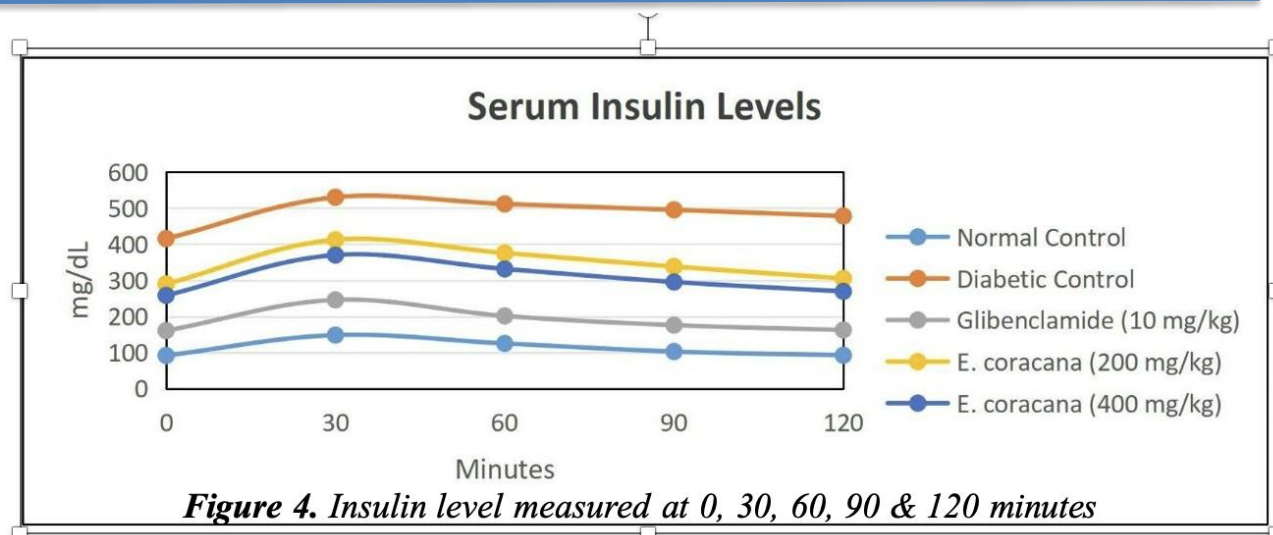
Oral Glucose Tolerance Test (OGTT) and Serum Insulin

The OGTT conducted on Day 21 showed that EC-M increased glucose clearance significantly (Table 5). At 30 minutes, the Diabetic Control reported a blood glucose spike of 530.2 ± 21.0 mg/dL, which was reduced to 370.1 ± 14.0 mg/dL in the 400mg/kg group. Day 28 serum insulin revealed that there were moderate restorative effects of the β -cells. STZ led to a sharp decrease in insulin (5.1 μ U/mL), whereas EC-M treatment positively altered the levels significantly (Table 5) in a dose- dependent manner.

Table 5. OGTT Clearance (120 min) and Serum Insulin Levels (Day 28)

Group	Treatment	OGTT 120 min (mg/dL)		Insulin (μ U/mL)
		0 min	120 min	Day 28
I	Normal Control	91.7 ± 4.4	92.4 ± 4.1	14.8 ± 1.1
II	Diabetic Control	415.6 ± 17.9	478.1 ± 18.8	5.1 ± 0.6
III	Glibenclamide (10 mg/kg)	160.7 ± 8.1**	162.5 ± 7.7**	11.2 ± 0.9**
IV	EC-M (200 mg/kg)	290.0 ± 11.0**	305.1 ± 12.0**	6.2 ± 0.6*
V	EC-M (400 mg/kg)	258.4 ± 10.8**	269.3 ± 10.9**	7.3 ± 0.8*

The OGTT conducted on Day 21 showed that EC-M increased glucose clearance significantly (Table 5). At 30 minutes, the Diabetic Control reported a blood glucose spike of 530.2 ± 21.0 mg/dL, which was reduced to 370.1 ± 14.0 mg/dL in the 400mg/kg group. Day 28 serum insulin revealed that there were moderate restorative effects of the β -cells. STZ led to a sharp decrease in insulin (5.1 μ U/mL), whereas EC-M treatment positively altered the levels significantly (Table 5) in a dose- dependent manner.



Molecular Authentication and Phylogenetic Validation of *Eleusine coracana* Genomic DNA sample of voucher accession SGU-ET-011 (RAGI sample) produced a high-quality template to amplify the locus of chloroplast *rbcL*. The amplification of PCR resulted in a single and discrete band of the correct size, demonstrating specific amplification and no secondary products. The bidirectional sequencing used in Sanger sequencing produced high-quality consensus sequence of 979 base pairs (bp) with trimming of low-quality terminal bases.



Figure 5a) Distribution of the top 100 BLAST Hits for the *rbcL* barcode sequence of *E. coracana*; b) Nucleotide sequence

Table 6. Summary of DNA Barcoding Metrics for *Eleusine coracana*

Sample ID	Assigned Species (BLAST Match)	Barcode Marker	Consensus Sequence Length (bp)	Internal Sequence ID
RAGI	<i>Eleusine coracana</i>	<u>rbcL</u>	979	0725_1014_006_PCR_3_RAGI_RBCL1272R (Consensus) 1

BLASTn Analysis of the consensus sequence using the NCBI nucleotide database showed a 99.89% identity of the nucleotide with the reference sequence of *Eleusine coracana* (GenBank accession: MF114353.1), and a 100 percent query coverage. The observed divergence of sequence was associated with a difference of about one nucleotide, over the 979 bp, alignment which is far below accepted limits of species- level identification in plant DNA barcoding. There were no other species that showed similar identity and coverage values and hence ruled out misidentification or contamination.

Table 7. Definitive BLAST Homology Search Results for *rbcL* Sequences

Sample ID	Top Matched Species (GenBank)	Accession Number	Max Score (bits)	Query Coverage (%)	Percent Identity (%)	E-Value	Identification Status
RAGI	<i>Eleusine coracana</i>	MF114353.1	1650	100%	99.89 %	0	Confirmed

Taxonomic placement was further confirmed by phylogenetic reconstruction by the Neighbor-Joining technique using the Maximum Composite Likelihood (MCL) substitution model. The tree created placed SGU-ET-011 in a deep-rooted clade of *Eleusine coracana*, having more than 70% bootstrap support. Pairwise genetic analysis of divergence showed that there was insignificant divergence (MCL distance < 0.001) between SGU-ET-011 and authenticated *E. coracana* reference sequences. When other methods of inference were used, phylogenetic stability was determined through tree topology.

Table 8. Summary of Sequence Divergence and Phylogenetic Support for Study Samples

Sample ID	Top GenBank Match	Alignment Length (bp)	Estimated Pairwise Genetic Distance (MCL)	Taxonomic Reliability
RAGI	<i>Eleusine coracana</i> (MF114353.1)	862	Near 0.001	High

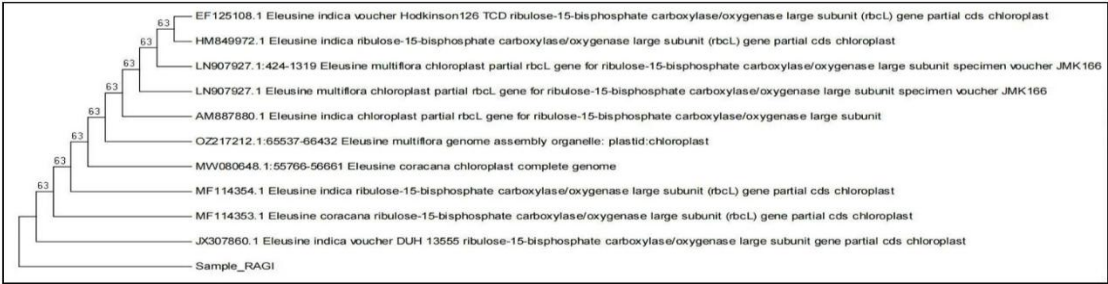


Figure 6. Evolutionary relationships of *Eleusine coracana*, inferred using the Neighbor-Joining method. The phylogenetic tree was constructed based on *rbcL* sequences using the Maximum Composite Likelihood model and supported by 500 bootstrap replicates. The topology confirms the species-level clustering of *Eleusine coracana*

The strong nucleotide homology (99.89%), full coverage of the query, low genetic divergence, and strong bootstrap presence confidently prove the identity of the species of SGU-ET-011 as *Eleusine coracana*. These results exclude the chances of contamination or taxonomic obscurity and achieve a confirmed genetic basis of further phytochemical and pharmacological research provided in this paper.

Discussion

The current study provides a strong ethnobotanical support of *Eleusine coracana* as a dietary supplement in the management of diabetes among tribal South Gujarat groups. The extremely large Informant Consensus Factor (ICF = 0.9287) of diabetes is a sign of homogeneity in species choice among five tribal groups (Gamit, Vasava, Dhodia, Warli, and Kukna) to an extraordinary degree. This agreement is higher than the findings reported in other ethnobotanical studies in other parts of India, where metabolic disorders with a median ICF of 0.75 to 0.85 are reported (Nazar et al., 2024). The high level of consensus indicates that the use of *E. coracana* as a glycemic control is empirically supported traditional knowledge that has been refined over the generations. This is further supported by the Fidelity Level (FL = 87.2%), which implies that citations were linked to hypoglycemic applications specifically and not generalized uses. The Relative Frequency of Citation (RFC = 0.567) indicates a moderate-high cultural significance, which is equal to the range found in traditionally tested antidiabetic plants in the same areas. The population structure exposes a serious issue: 82.5% of knowledge owners

were 46 years or older, and 76.7% said they had over 20 years of experience. This implies that ethnomedicinal knowledge is centralized in old practitioners, which is worrying regarding intergenerational transfer. The same trends are being reported in India and Africa, where modernization is threatening the traditional therapeutic systems based on food (Tadesse et al., 2024).

Sequential extraction demonstrated that variations were largely solvent-dependent and that the variations were of important significance to therapeutic standardization. The elevated yield of aqueous extract (14.1% vs. 8.9% in methanol) indicates the presence of large amounts of water-soluble macronutrients. Nevertheless, the methanolic extract had better secondary metabolite content, as indicated by significantly high Total Phenolic Content (TPC: 91.4 ± 5.3 mg GAE/g DW) and Total Flavonoid Content (TFC: 55.2 ± 3.3 mg QE/g DW). These TPC values are comparable to the higher spectrum of finger millet varieties across the world (45-110 mg GAE/g) according to genotype, growing conditions, and processing methods (Mitharwal et al., 2021). The 1.7fold rise in the TPC and >2-fold rise in

TFC in methanolic versus aqueous fractions highlights the significance of the extraction methodology in order to maximize the bioactive compounds in the extract (Abioye et al., 2022). This has a direct implication on standardized development of nutraceutical formulations.

The findings have great implications on tribal nutrition and management of diabetes in South Gujarat. The tribal population has a diabetes rate of 3.7% to 10.2% (VM et al., 2025), and the metabolic syndrome prevalence rate among tribal adolescents in Gujarat is 3.8%, which suggests the problem of nutrition transition is on the verge of emerging (Shrivastava et al., 2023).

Nevertheless, the considerations of implementation are necessary. The method of processing also has a strong influence on nutritional and therapeutic properties; minimally processed millets were 30% more effective in reducing the meal GI than milled rice and refined wheat (Anitha et al., 2021). Refined products should be discouraged in favor of traditional preparation methods that maintain seed coat and aleurone layer.

Conclusions

The study presents detailed evidence to convince that the traditional use of *Eleusine coracana* is a safe way to manage diabetes in South Gujarat tribal populations. The combination of ethnobotanical validation (ICF = 0.9287, FL = 87.2%), phytochemical characterization (TPC: 91.4 mg GAE/g, TFC: 55.2 mg QE/g), in vitro enzyme inhibition (α -glucosidase IC_{50} : 74.6 μ g/mL) and in vivo antidiabetic activity (reduced FBG 38.3% with 400 mg/kg) and molecular authentication (99.89% identity) establishes a robust scientific foundation for integrating finger millet into evidence-based nutrition and healthcare strategies. These findings contribute to the growing evidence supporting millets as a functional food to prevent and manage diabetes, particularly relevant in the context of the International Year of Millets (2023) and the global efforts addressing nutrition security and chronic disease. Among the tribal groups in South Gujarat and other such areas, *E. coracana* is a culturally acceptable, economically viable and scientifically proven alternative to treating diabetes sustainably.

Combination of traditional food systems and modern scientific validation is an example of a holistic approach to public health that takes into account indigenous knowledge without violating the standards of modern evidence. To achieve the potential of *Eleusine coracana* in terms of tribal nutrition and diabetes management, clinical translation, long-term safety evaluation, and setting policies are to be considered in the future.

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