ETHNOBIOLOGY AND CONSERVATION

RESEARCH ARTICLE

Ethnobiology and Conservation, 14:34 (14 October 2025)
doi:10.15451/ec2025-10-14-34-1-13
ISSN 2238-4782 ethnobioconservation.com

Rediscovering Crops: DNA species identification and ethnobotanical investigation of chenopods in the eastern Himalayas

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ABSTRACT

One strategy responding to global food security challenges is to enhance the utilization of ancient and under-exploited niche crops. As Andean Quinoa has garnered considerable attention, research on chenopods in Asia has not kept pace. This study launched a field investigation on the cultivation and use of chenopods in the southern foothills of the eastern Himalayas, an area that may have served as a center for chenopod domestication. By employing DNA analyses, we identified the species involved. As highly adaptable crops, Chenopods are typically cultivated using intercropping practices to support sustainable agricultural production in this region. Our findings reveal for the first time that the chenopods cultivated in this region originated from *Chenopodium album*, but over time, due to prolonged human selection and environmental influences, distinct morphological traits and genetic variations have emerged, leading to localized varieties. Based on phylogenetic, morphological and spatial distribution analyses, we propose the existence of multiple independent domestication centers of chenopods in Asia. This study not only sheds light on the origin and domestication of chenopods in Asia but also underscores the strategic potential of their nutritional value and unique cultivation practices in addressing climate change, enhancing global food security, and contributing to the achievement of the Sustainable Development Goals.

Keywords: DNA Sequencing, Niche Crops, Crop Utilization, Cultivation Practices, Food Security.

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SIGNIFICANCE STATEMENT

Chenopods, as resilient and nutrient-dense niche crops, hold a distinctive advantage in agricultural production, particularly in alpine and ecologically fragile environments. Although quinoa has been known as a climate-smart “super crop”, research on chenopods in Asia has not kept pace even though their charred seeds have often been recovered in large quantities from archaeological sites, indicating a long history of utilization. In this study, we conducted a systematic ethnobotanical field investigation in the eastern Himalayas, a proposed potential domestication center, documenting the practices of cultivation, processing, and consumption of chenopod crops in remote mountainous regions. By employing DNA sequencing, we also reveal for the first time that cultivated chenopods in East Asia originate from *Chenopodium album*, marking a distinction from the chenopods cultivated in other regions worldwide, possibly suggesting the existence of independent domestication centers in Asia. The research will not only help to understand the species origin, domestication, and processing history of chenopods in East Asia but, more importantly, it provides actionable insights into resilient and adaptable alternative crops and cultivation practices for future agricultural systems.

INTRODUCTION

The global food system is currently confronted with a multitude of complex challenges. Climate change, regional conflicts, population growth, and economic slowdown are becoming increasingly severe (FAO et al. 2023). Meanwhile, the trend toward monoculture in modern agriculture has led to increased productivity and simplified operations in the short term (Franco et al. 2022), it has also brought about detrimental impacts, including reduced biodiversity of the farmland systems, soil degradation, and environmental pollution (FAO 2023). To effectively tackle these challenges, it is imperative to promote sustainable agricultural practices, develop resilient value chains, and make better use of ancient, under-exploited niche crops (Mabhaudhi et al. 2019; Azam-Ali 2010).

Chenopods, such as *Chenopodium quinoa* and *C. pallidicaule*, are important niche crops. They are gluten-free and nutrient-rich, require minimal maintenance during cultivation, exhibit resistance to a variety of pests and diseases, as well as remarkable resilience to drought and salinity, allowing them to thrive in infertile soils (Murphy et al. 2015). In arid regions, these niche crops provide essential land cover that mitigates soil degradation, and helps to promote biodiversity and sustainable land restoration, thereby enhancing the stability of agro-ecosystems (Repo-Carrasco 2003; Tang et al. 2017). Currently, these crops are leading the way in global grain production trends as cereal alternatives, commonly referred to as pseudocereals (Pirzadah et al. 2020; Bazile et al. 2016).

Quinoa (*C. quinoa*), an ancient crop that is native to the Andes (Martinez et al. 2015; Popenoe et al. 1989), is often described as an “all-purpose grain” because of its critical contributions in ensuring local food security, nutritional health, and poverty alleviation. With its global dissemination and promotion, the Food and Agriculture Organization of the United Nations designated 2013 as the International Year of Quinoa (UN 2011). In addition, *C. berlandieri* ssp. *jonesianum* has been confirmed to be one of the key crops in the prehistoric Eastern Agricultural Complex in the woodlands of Eastern North America, providing an important source of food for prehistoric societies along with other locally domesticated crops such as sunflowers and sumpweed, revealing the existence of a unique and complex indigenous agricultural civilization on the North American continent before corn dominated agriculture. *C. berlandieri* ssp. *nuttalliae* was an important food crop of the Aztec Empire (14th-16th centuries) in Mesoamerica, holding cultural significance in religious practices, including sacrifices (Wilson et al. 1979; Safford 1918), and is nowadays grown as a minor crop on the central highlands of Mexico (Glore 2006). *C. pallidicaule*, known as cañahua, is another highly adaptable plant with a protein content of 13.8%, higher than that of quinoa and wheat. Currently, it is primarily grown on the Altiplano Plateau at elevations above 3,800 m in South America, one of the most challenging agricultural regions in the world. Cañahua has been vital for generations of indigenous peoples, helping to combat malnutrition linked to poverty and ensuring regional food security (Gade 1970).

Nowadays, cultivated chenopods are also grown in Asia. Along the line of the Himalayas, the Hengduan Mountains, and the Southeast Downs, feature unique topographic and climatic conditions, along with a history of both natural and anthropogenic selection. These factors have contributed to the development of crops in this region with strong environmental adaptability, positioning it as a key area for the cultivation of niche crops like chenopods (Partap et al. 1998). However, there is a deficiency in surveys and corresponding collection and conservation of germplasm resources. Cultivated chenopods known in Asia include *C. album* in the southern foothills of the central Himalayas (Partap et al. 1998 1985a 1985b), *C. formosanum* in eastern Asia (Wang 2012; Wang 2018), and *C. giganteum* (Yang 2023), *C. bryoniifolium* and *C. album* (Huang et al., 2022) in central Asia. Current evidence suggests that Asia could be an independent center of origin for chenopod crops, with the possibility of multiple domestication centers throughout the region.

One of the most notable regions for chenopod cultivation is the southeastern Tibetan Plateau, situated in the eastern Himalayas and adjacent to the Hengduan Mountains. (1) The geographical environment closely resembles that of areas where many chenopod crops thrive. This mountainous environment region (IBCAS et al., 1988), consistent with, for example, the Andes Plateau and the woodland of Eastern North America, presents challenging subsistence conditions that may have prompted local ancestors to expand the utilization of some niche crops. (2) Chenopod plants in the region have a long history of use. Prehistoric archaeological sites with systematic archaeological excavations and archaeobotanical work reveal that *Chenopodium* seeds are among the most common plant remains, even outnumbering those of agricultural crop remains. For example, the sites of Bangga (75189 seeds, 3004-2160 BP) (Tang et al. 2021), La phob (8836 seeds, 4825-3897 cal BP) and Klu Lding (37392 seeds, 3840-3240 cal BP) in the Yarlung Zangbo River basin (Wang et al. 2024), the Karuo (555 seeds, 4750-2650 BP) (Song et al. 2021); and the Haimenkou (15911 seeds, 3550-2250 BP) sites in the Lancang River Basin (Min 2013; Xue et al. 2013), etc., therefore, some researchers proposed that chenopods may have been cultivated during prehistorical periods (Gao 2020; Guedes 2018). (3) The use of chenopods has persisted into the modern era in this region. In the 1980s, the Tibet Crop Variety Resources Survey first discovered the cultivation of chenopod crops, primarily in the agroforestry districts of southeastern Tibet, at elevations below 2500 m, including parts of Mêdog Country, where the local people eat their seeds (Wang 1983, 1995), but the specific chenopod species used, as well as how they were processed and consumed, have not been determined.

Given this, we conducted a field survey in the Mêdog region of the eastern Himalayas to explore the traditional cultivation techniques and farming systems for chenopod crops currently in use. Additionally, we collected the germplasm resources and identified the species through DNA analysis. Our research is devoted to providing insights into modern crop cultivation and agricultural production, while also deepening our understanding of the link between chenopod remains excavated from archaeological sites and the livelihoods of ancient peoples, including their role in prehistoric diets and subsistence strategies.

MATERIAL AND METHODS

Ethnobotanical fieldwork

Mêdog County is administratively under the jurisdiction of Nyingchi City, Xizang Autonomous Region,China, located in the lower reaches of the Yarlung Zangbo River geographically, at the eastern end of the Himalayas (Figure 1). The terrain descends from north to south, with elevations ranging from 200 to 7782 m, which demonstrates pronounced altitudinal zonality. This region experiences an average annual temperature of 16~18°C and receives between 2000 and 3000 mm of precipitation each year. The landscape is characterized by a dense network of rivers, extensive marshlands, and striking snow-capped mountains and glaciers.

It is mainly inhabited by the Moinba, Lhoba, Zang, and Han ethnicities in Mêdog. Due to the barrier of the Himalayan Mountains, along with complex geological conditions and specific historical factors, productivity levels have remained low, and practiced slash-and-burn agriculture has been practiced for a long time (Deng 2011). Therefore, more traditional livelihoods have been retained, creating a valuable opportunity for our study.

We carried out a 10-day ethnological field survey in Mêdog in August 2023, covering 8 villages across 5 townships (Figure 1). By the ethnographic fieldwork specifications (Meier zu Verl et al. 2024), we clarified the objectives, defined the geographical scope, reviewed relevant studies, formulated a survey outline, and ensured a shared understanding of key survey segment. During the field survey, we collected 5 seed samples and 21 stem and leaf samples which will be used for laboratory morphological analysis and DNA identification. All biological material collected was deposited in Bioarchaeology Laboratory, Lanzhou University. Additionally, we documented local practices related to the cultivation, processing, and consumption of chenopods.

Next-generation DNA sequencing and data analysis

To obtain detailed species information on cultivated chenopods crops in the southern foothills of the eastern Himalayas and to explore their relationship with the domesticated *C. formosanum* (locals refer to it as djulis) in eastern Asia, cultivated samples MdC1 and MdC2, weed sample MdW1 obtained during the investigation, and TwC1 purchased from Taiwan (Figure 2). The DNA extraction, library preparation and sequencing of four samples were all sent to Annoroad Gene Technology (Beijing) Co., Ltd. All specimens were cleaned according to the experimental require-

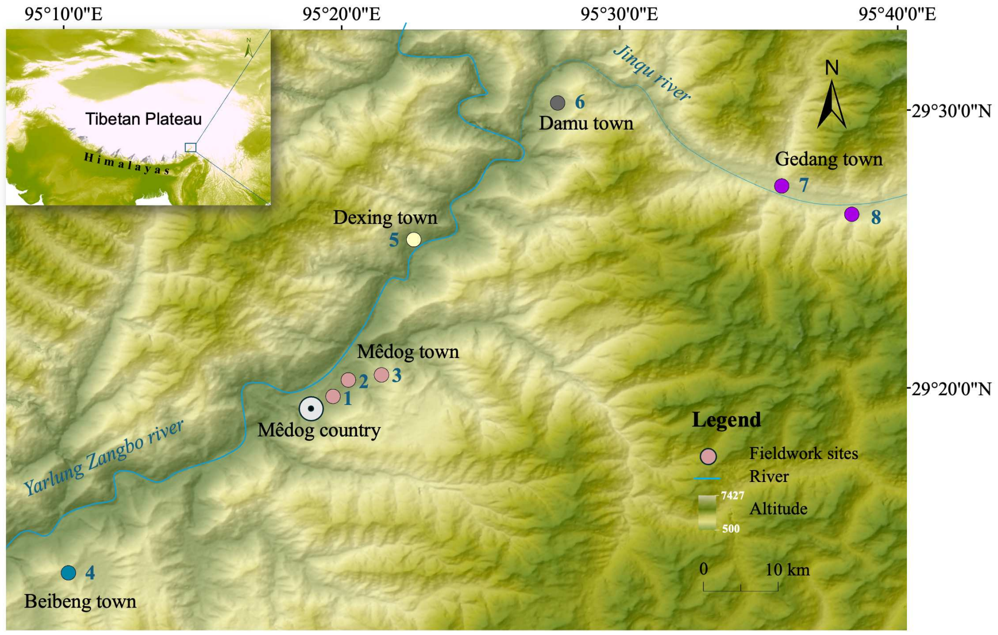

Figure 1. Distribution of survey sites for chenopods in Mêdog County, Nyingchi City, Tibet. Different towns are represented by dots of varying colors. 1, Yadong village, Mêdog town, 1006 m; 2, Mêdog village, Mêdog town, 1112 m; 3, Bari village, Mêdog town, 1646 m; 4, Gelin village, Beibeng town, 1738m; 5, Deguo village, Dexing town, 856 m; 6, Damu village, Damu town, 1575 m; 7, Sangzhenka village, Gedang town, 1978 m; 8, Deji village, Gedang town, 1711 m.

ments before being sent for DNA extraction. They were initially rinsed with ddH_2O to eliminate any surface dust or dirt, and the remaining moisture was blotted using lint-free paper. The tissues were rapidly flash-frozen using liquid nitrogen and transferred into 50 mL centrifuge tubes. To ensure the experiment proceeded smoothly, each sample weighed no less than 0.5 g, with careful attention given throughout the process to prevent cross-contamination between samples.

Genomic DNA was extracted from all four samples using a universal magnetic bead-based kit for plant and animal tissues. Specifically, MdW1 was derived from leaf tissue, whereas MdC1, MdC2, and TwC1 were obtained from seeds. All DNA extraction procedures were performed at Annoroad Gene Technology (Beijing) Co., Ltd. Following DNA extraction, libraries directly sequenced by the whole genome shotgun were prepared according to the TruSeq DNA Sample Preparation Guide (Illumina, 15026486 Rev.C). The constructed libraries were initially quantified using Qubit 2.0 Fluorometer (Thermo Fisher Scientific), followed by fragment size distribution analysis on an Agilent 2100 Bioanalyzer. Libraries passing quality control were then precisely quantified by Q-PCR(Real-time PCR). Paired-end sequencing (2×150 bp) was subsequently performed on an Illumina NovaSeq 6000 platform.

The raw sequencing data were processed using “cutadapt 1.12” (Martin 2011) to remove adapter sequences from each DNA library sequencing file and filter out short fragments with lengths less than 30 bp. Next, the “aln” and “samse” functions in the “BWA 0.7.15” (Burrows-Wheeler Alignment) software (Li et al. 2010) were used to align the fragments to the Maturase K (matK) gene sequence of *C. album* (GenBank: KX676641), discarding sequences with an alignment quality score below 30. SAMtools v1.3.1 software (Li et al. 2009) was used to sort sequences by their 5’ end on the reference genome, remove PCR duplicates with “rmdup”, and merge them into bam files. The final matK sequence of the specimen was exported as a FASTA file using the "-doFasta" function in the angsd-0.916 software (Korneliussen et al. 2014).

To analyze the samples from this study within a broader spatial and temporal framework, we downloaded matK gene sequences from 12 *Chenopodium* species via GenBank to build a reference database (Additional File 1). Using the previously described methods, we identified the species of the four samples in this study. Sequence alignment was performed online through the CIPRES (`http://www.phylo.org`), and phylogenetic trees were constructed using RAxML v 8.0 (Stamatakis 2014) and MEGA11 (Tamura et al. 2021) with 1,000 bootstrap replicates to assess node support. The phylogenetic tree was visualized and output using Figtree v1.4.4 (Rambaut 2018).

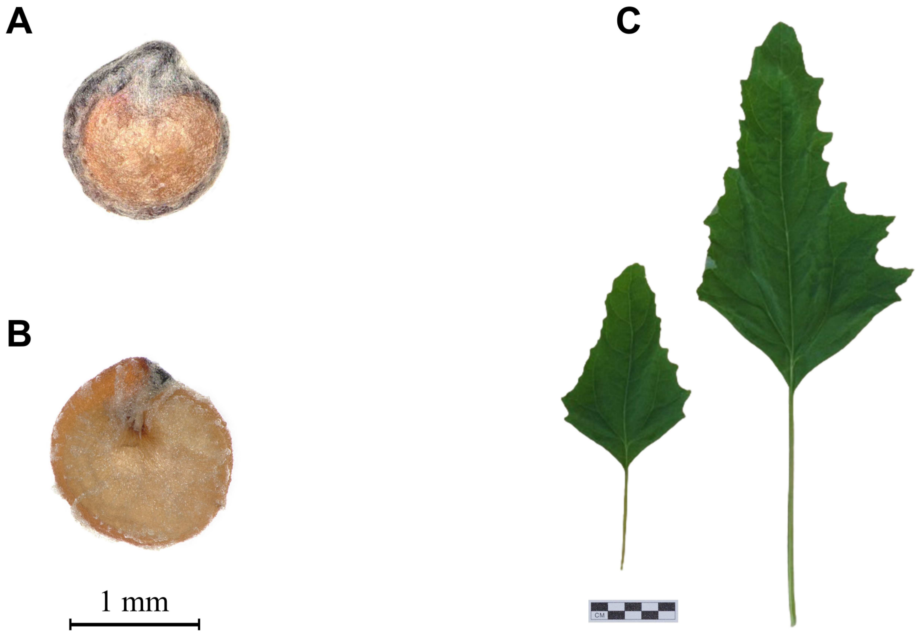

Figure 2. Samples for DNA sequencing. (A-B), seeds and leaves of chenopods crops in the Mêdog County; (C), seeds of *C. formosanum* from Taiwan.

RESULTS AND DISCUSSION

DNA species identification of chenopods in the eastern Himalaya

The results of aligning the raw sequencing data of the four samples to the *C. quinoa* matK gene sequence are as follows (Table 1). As all four samples are modern, the matK sequences depths obtained are relatively high, which show mean coverage higher than 100x.

To reconstruct the phylogenetic relationships and investigate the phylogenetic status of the analyzed samples among the *Chenopodium*, the four newly obtained matK sequences were aligned with 57 *Chenopodium* matK sequences from GenBank, including *C. acuminatum*, *C. album*, *C. hybridum*, *C. iljinii*, *C. rubrum*, *C. serotinum*, *C. strictum*, and *C. urbicum*, with *C. quinoa* as the outgroup to construct a maximum-likelihood tree (Figure 3).

All *Chenopodium* matK sequences were clustered into two groups: *C. quinoa* and other *Chenopodium* species. The matK sequences from three samples obtained from Mêdog County in this study clustered within the branch containing modern *C. album*, indicating that they belong to *C. album*. The TwC1 collected from Taiwan also falls into the same branch, suggesting that djulis is likely a local variety of *C. album*.

Table 1. Information about the DNA analysis results

Experimental code	Number of reads	Coverage (X)	Location	Status
MdC1	1357	164.841	Mêdog	cultivated
MdW1	1252	174.543	Mêdog	weedy
MdC1	754	111.74	Mêdog	cultivated
TwC1	1524	197.731	Taiwan	cultivated

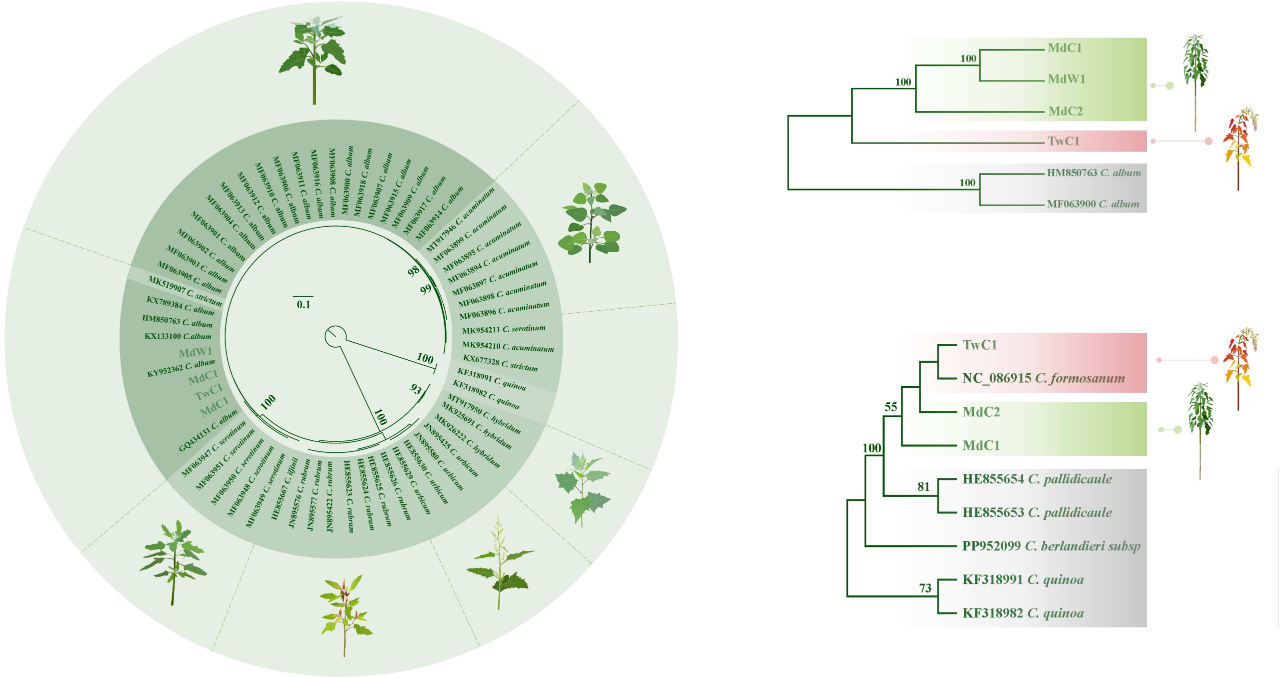

Figure 3. Phylogenetic analysis of *Chenopodium*. (A), Maximum-likelihood phylogenetic tree of the genus *Chenopodium* based on 803 bp of matK gene sequences including four sample sequences obtained in this study; *C. quinoa* was selected as outgroup; (B), Neighbour-joining tree of the six samples; (C), Neighbour-joining tree of cultivated chenopods around the world. Branch labels show bootstrap values derived from 1,000 replications.

Cultivation, consumption and processing of chenopods

Chenopods, locally known as "Xiaermu" by the Moinpa and Lhoba of Mêdog County in southeastern Tibet, are annual crops. Although there is a lack of documented historical evidence regarding their cultivation, oral testimonies from local informants suggest that chenopods have a long-established history as a widely grown crop in the region. Our field investigation across five towns in Mêdog County revealed that chenopods are consistently cultivated in all surveyed villages. While not a staple crop, they play a significant role in local subsistence strategies.

Morphology

Common cultivars of chenopods have tall plants, mostly over 3 m in height, and the stalks are erect and robust, characterized by green or purplish-red stripes. The leaves are broadly lanceolate or rhombic-ovate with an irregular serrated margin, the petiole is nearly two-thirds the length of the leaf (Figure 2B) and the upper leaves often show a purple hue. The flowers have bisexual, paniculate inflorescences, featuring a perianth divided into 5 lobes with longitudinal ridges on the abaxial surface. In contrast, wild *C. album* plants are generally shorter, typically less than 1.5 m tall, and tend to be more branched.

The seed has truncate margin, circular embryo, smooth surface, 1-1.5 mm in diameter. The pericarp is translucent, adhering to the testa surface, which is light yellow and thin (Figure 2A), so that the seeds can be easily crushed. In contrast, wild *C. album* seeds are usually biconvex, with a black seed coat that is thick and hard.

It can be observed that compared to wild *C. album*, the cultivated chenopods in this region have undergone obvious changes in plant height, branching, and seed morphology.

Cultivation practices

Multistorey cropping involves the practice of planting shorter companion crops beneath taller ones in a staggered arrangement to optimize the use of land, light, and heat resources. Chenopods are typically intercropped with corn, chili pepper (*Capsicum annuum*), and taro (*Colocasia esculenta*) in Mêdog. Occasionally, they are mixed with foxtail millet (*Setaria italica*), common millet (*Panicum miliaceum*), finger millet (*Eleusine coracana*), lemon (*Citrus limon*), sesame (*Sesamum indicum*), sweet potato (*Ipomoea batatas*), and scallion (*Allium fistulosum*), etc. (Figure 4A).

Companion cropping is when one crop occupies the main field while another is planted in a boundary about 1 m wide in a fence-like manner. This farming method is also common in Mêdog. For instance, in Deguo Village, Dexing Town, it was found that chenopod was planted around the perimeter of sesame fields (Figure 4B). Similarly, in the Damudo Lhoba Ethnic Town, chenopod was planted in the periphery of dryland rice fields.

Pure cropping means the cultivation of a single crop on a plot of land. This type of cultivation of chenopods is relatively rare, during the survey, it was only observed in a small plot in the Damu Lhoba Ethnic Town (Figure 4C), where the chenopod plants were comparatively shorter.

Sowing, harvesting and consumption

Chenopods are cultivated as an annual crop in both horticultural fields and farmland in Mêdog, with sown in April to May and harvested in October to November. The timing of these activities varies with heat caused by altitude, as the elevation increases, both sowing and harvesting time will be relatively delayed (Additional File 2). It is usually sown by scattering without topsoil or covering with a light layer of soil. Thinning management is performed when the plants reach a height of 15-20 cm. As they grow, the bottom leaves are removed to encourage upward growth and used as livestock feed. Depending on soil fertility and growing conditions, plants reach heights of 1-4 m, with a recorded maximum of 4.3 m during the survey. Growth conditions significantly affect yield; under optimal conditions, a single plant can produce at least 0.5 kg of seeds, while in less favorable environments, 10 or more plants may be required to achieve the same yield.

Historically, chenopods were cultivated by slash-and-burn techniques, and now, practices commonly involve the application of manure to enhance yields. Chenopods grown in horticultural fields typically receive organic manure, whereas those in farmland are often treated with chemical fertilizers or left unfertilized.

In line with most cereal processing methods, the processing of chenopods in the Mêdog region involves several steps, including threshing, winnowing, pounding, etc. Once the crops reach maturity, only the inflorescences are cut off and sun-dried, then placed on a stone slab and threshed by striking with wooden sticks. Following threshing, a round dustpan is used to separate the chaff from the seeds (Figure 5A). For smaller quantities, husking is done by hand-rubbing (Figure 5B-C), whereas larger amounts require cooperation, using wooden mortars and pestles to repeatedly and gently pound the seeds to remove the husks (Figure 5D-E). Following a second round of winnowing, the clean seeds are obtained (Figure 5F) and stored in sealed containers in a cool, dry place.

The primary edible component of chenopods is the seeds, though the young shoots and tender leaves are also consumed as vegetables. The seeds are prepared in several ways: (1) they are often mixed and steamed with staple foods such as rice and corn to add flavor; (2) the seeds are roasted and ground into flour, which is used to make foods like tsampa; (3) they can be used for brewing alcohol.

DISCUSSION

Phylogenetic and morphological reflections of linkages between samples

The results of DNA sequencing and phylogenetic analysis showed that the cultivated sample MdC1 from Deguo village is more closely related to the weedy sample MdW1 collected from Bari village than to another cultivated sample MdC2 from the same location. Besides, the cultivated sample TwC1 (*C. formosanum*) shows a more distant phylogenetic relationship to these 3 samples (Figure 3B), even though all belong to *C. album* group.

In terms of plant morphology, the leaves of MdC1 (Figure 6A) are relatively broad, with irregular serrated margins and a mostly rhombic shape, and the stalk was erect and robust, with no branching (Figure 6B), which is generally consistent with the cultivated chenopods in other areas of Mêdog. However, the leaves of MdC2 (Figure 6C) are smaller, narrow-oval, or lanceolate, with smooth margins, while the stalk was also erect and robust, but frequently branched (Figure 6D). Additionally, MdC1 shows greater morphological similarity to MdW1 regarding its leaf and stalk characteristics, with broader leaves and fewer branches (Figure 6E). The TwC1 (*C. formosanum*), at maturity, features red leaves and stalk, with broad leaves, slender erect stalk, and no branching (Figure 6F).

Based on the integrated results of phylogenetic analysis and morphological observations, the following conclusions can be drawn: (1) MdC1 exhibits closer phylogenetic and morphological affinity to MdW1. With an average height exceeding 3 m, MdW1 is hypothesized to represent a feral form of the cultivated chenopod, having reverted to a wild state following its escape from intensive cultivation and management. (2) MdC1 and MdC2 display significant morphological divergence, despite both being cultivated in Deguo village. We can infer that MdC2 represents a distinct variety, refers to a group of individuals or populations within the same species that have developed stable,

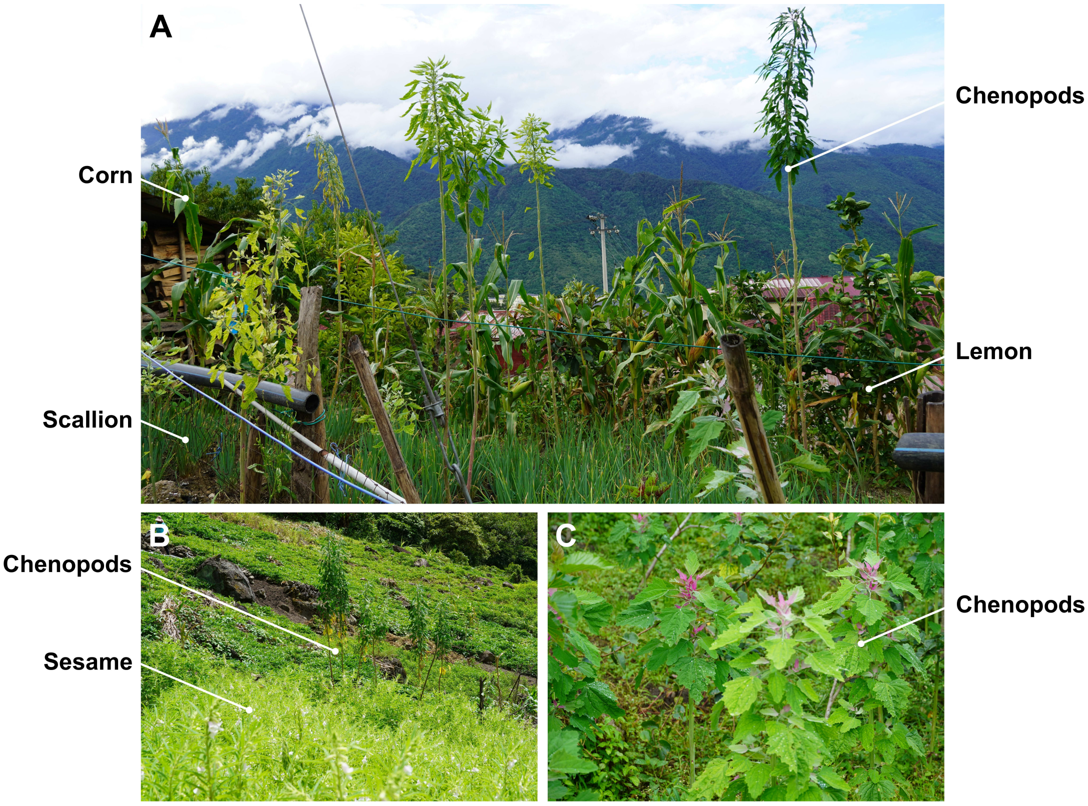

Figure 4. Cultivation patterns of chenopods. (A), multistorey cropping of chenopods and other crops; (b), chenopods are intercropped around the sesame field; (c), chenopods are cultivated purely in the field.

Figure 5. The processing of chenopods crops. (A), using a winnowing dustpan to lift seeds to remove husk; (B-C), shell the grain by hand rubbing and blow off husk; (D-E), using a wooden mortar and dustpan to shell grain and remove husk; (F), pure grain seeds.

heritable morphological differences due to environmental variations, while there is no distinct zonal distribution in geographical terms. (3) TwC1, aside from its color, shares notable morphological similarities with MdC1. Combined with the DNA species identification results, TwC1, domesticated in eastern Asia, may represent a distinct subspecies, which refers to a population within a species that has developed certain mor-

Figure 6. Morphological comparison of 3 different Chenopods crop samples and 1 weedy sample. (A-B), leaves and stem of MdC1; (C-D), leaves and stem of MdC2; (E) plant of MdW1; (F) plant of TwC1.

phological differences after being geographically, ecologically, or seasonally isolated from other populations of the same species.

DNA species identification of chenopods in the eastern Himalayas

Current findings suggest that chenopods cultivated in the Himalayan region and eastern Asia have a common origin, indicating a shared ancestor (with species identification in the southern foot in the central Himalayas primarily relying on morphological analysis), multiple lines of evidence indicate that the domestication process of chenopods in different parts of Asia may have occurred independently. The foothills of the Himalayan and the eastern Asia are geographically distant, appearing as distinct hotspots in terms of their regional distribution. Phylogenetic analyses reveal that TwC1 clusters on a separate branch from those cultivated in Mêdog (Figure 3B). Morphologically, the chenopods cultivated in these two regions also exhibit noticeable differences in plant color and seed morphology. While this implies that significant regional variation may exist in the domestication and cultivation pathways of chenopods between the Himalayas regions and eastern Asia, further research is needed to clarify the relationship between chenopods cultivated in the eastern and central Himalayas, which are geographically closer.

Further comparison of the matK gene sequences of *C. berlanderi* from North America, *C. quinoa*, and *C. pallidicaule* from South America reveals that samples from the Asian region cluster on separate branches (Figure 3C). This indicates that the chenopods cultivated in Asia originated from entirely different species. Cultivated and domesticated chenopods from various regions exhibit unique regional differences in genetic diversity, morphological characteristics, and environmental adaptability. These differences likely reflect the selection pressures they experienced during the domestication process, influenced by differing ecological environments, agricultural practices, and cultural contexts.

In this, we speculate that Asia may represent an important center of origin for chenopod crops, with multiple sites of cultivation and domestication. In these centers, humans have independently selected *C. album* around residential areas to domesticate, adapting it to their specific environment and the requirements of society.

Insights from Chenopods farming practices

Not only in the Mêdog region but also in various ethnographic and modern agricultural studies from other areas, there is a predominant reflection of intercropping practices involving chenopods with staple crops, such as Himachal Pradesh in the southern slopes of the Himalayas (Partap et al. 1985a, 1985b) and Taiwan in East Asia (Fogg 1983).

As auxiliary plants for human consumption, intercropping with staple crops facilitates crop management and optimizes the use of land and labor time (Gao et al. 2019). By cultivating crops with varying heights, nutrient needs, and fertility periods in a layered arrangement, the system maximizes sunlight exposure and improves ventilation and light penetration among field crops (Yuan et al. 2008). Moreover, intercropping boosts the diversity of soil microbial species, upgrades nutrient structures, and regulates the balance of salinity and pH, thereby heightening soil fertility and effectively mitigating pest and disease issues caused by continuous cropping (Bybee-Finley et al. 2018).

By 2050, global food production will need to double, yet the area of arable land will likely remain unchanged, water resources and other inputs are expected to decrease, and climate change will continue to alter the conditions for crop cultivation, bringing new challenges to food production (FAO et al. 2023). Future food production increases may not be concentrated in the core agricultural regions, but rather necessitate the cultivation of resilient crops and the development of suitable agricultural ecosystems in marginal agricultural areas (Partap et al. 1998). Many local varieties are experiencing a rapid decline due to ecological, economic, and other factors (Huang et al. 2022). Conserving the genetic resources of these traditional crops is crucial for preserving cultural diversity and providing an affluent genetic background and biological resources for future agricultural development. With systematic research and effective promotion strategies, domesticated chenopods in Asia are expected to play a greater role in global agricultural production and food security.

CONCLUSION

This study conducted DNA sequencing analysis on chenopods in Mêdog County, located at the eastern end of the Himalayas. The results revealed that it originated from wild *C. album*, the same as the *C. formosanum* in East Asia, as well as those in the central Himalayas. This suggests that they share a common ancestor and were independent evolution and domestication in different regions, providing new evidence for the origin and domestication of chenopod crops in Asia.

After internal ethnographic fieldwork locally, this study gathered insights into the cultivation, sowing, harvesting, and consumption practices of chenopods. It was found that the local cultivation of chenopods employs a mixed intercropping system designed to increase yields and optimize the use of heat and land resources. This not only affords valuable information for understanding, conserving, and utilizing niche crops like chenopods but also offers some thoughts about cultivation strategies to cope with climate change and optimize soil resource use.

Future research should integrate interdisciplinary methods, combine archaeology, anthropology, and agricultural sciences to explore the cultivation and domestication history, cultural significance, and utilization potential of chenopods in Asia, promote its role in global food security and nutritional health, and facilitate its wide application in modern agricultural practices.

ACKNOWLEDGMENTS

We would like to express our gratitude to the Forestry and Grassland Bureau of the Tibet Autonomous Region and S.Tordner of the Cultural Relics Bureau of Bomi County for their support and assistance during the survey. This work was funded by the National Natural Science Foundation of China (42371171), National Natural Science Foundation of China (41971231), Youth Innovation Promotion Association of the Chinese Academy of Sciences (2022068), Open Foundation of MOE Key Laboratory of Western China's Environmental System, Lanzhou University and the Fundamental Research Funds for the Central Universities (lzujbky-2024-jdzx01).

DATA AVAILABILITY

The data used to support the findings of this study are available from the corresponding author upon reasonable request.

CONFLICT OF INTEREST

The authors have no conflicts of interest to declare.

CONTRIBUTION STATEMENT

Conceived of the presented idea: TY, XY, YG (initial of the authors).
Carried out the experiment: TY, QY, YT (initial of the authors).
Carried out the data analysis: TY, LW (initial of the authors).
Wrote the first draft of the manuscript: TY, LW (initial of the authors).
Review and final write of the manuscript: XY, YG (initial of the authors).
Supervision: XY (initial of the authors).

DISCLOSURE OF AI USE

Artificial intelligence tools (ChatGPT by OpenAI) were used solely to improve the clarity and readability of the manuscript. No part of the scientific analysis, data interpretation, or conclusions was generated by AI. The authors take full responsibility for the content of this publication.

REFERENCES

Azam-Ali SN (2010) **Fitting underutilised crops within research-poor environments: Lessons and approaches.** *South African Journal of Plant and Soil* 27(4): 293-298. doi: 10.1080/02571862.2010.10639997.

Bazile D, Jacobsen S-E, Verniau A (2016) **The global expansion of Quinoa: Trends and limits.** *Frontiers in plant science* 7, 622. doi: 10.3389/fpls.2016.00622.

Bybee-Finley KA, Ryan MR (2018) **Advancing Intercropping Research and Practices in Industrialized Agricultural Landscapes.** *Agriculture* 8(6), 80. doi: 10.3390/agriculture8060080

Deng C (2011) **Research on sustainable development strategy of ecological environment in Mêdog County.** MSc dissertation, Northwest A&F University, Yangling, China.

FAO, IFAD, UNICEF, WFP & WHO (2023) **The state of food security and nutrition in the world 2023. Urbanization, agrifood systems transformation and healthy diets across the rural-urban continuum.** Rome, Italy. doi: 10.4060/cc3017en.

FAO (2023) **World Food and Agriculture – Statistical Yearbook 2023.** Rome, Italy. doi: 10.4060/cc8166en.

Fogg WH (1983) **Swidden cultivation of foxtail millet by Tai-wan Aborigines: A cultural analogue of the domestication of Setaria italica in China.** In: Keightley DN (ed) The origins of Chinese civilization. University of California Press, Berkeley, pp. 95-115.

Franco S, Pancino B, Martella A (2022) **Assessing the presence of a monoculture: from definition to quantification.** *Agriculture* 12(19), 1506. doi: 10.3390/agriculture12091506.

Gade DW (1970) **Ethnobotany of cañihua (*Chenopodium pallidicaule*), rustic seed crop of the Altiplano.** *Economic Botany* 24(1): 55-61. doi: 10.1007/BF02860637.

Gao A, Hu X, Jiang S (2019) **Survey of Crop Cultivation Resources in Key Areas of China.** China Agricultural Science and Technology Press, Beijing, China.

Gao Y (2020) **Archaeological Observation on The Utilization of Genus *Chenopodium* Linn during the Pre-Qin period in Southwest China.** *Journal of Tibet University (Social Science Edition)* 35(02): 17-26. doi: 10.16249/j.cnki.1005-5738.2020.02.003.

Glore AG (2006) **Domesticated *Chenopodium* in North America: Comparing the past and the present.** PhD Thesis, Washington University in St. Louis, Saint Louis, USA.

Guedes DJ, Hein A (2018) **Landscapes of Prehistoric Northwestern Sichuan: From Early Agriculture to Pastoralist Lifestyles.** *Journal of Field Archaeology* 43(2): 1-15. doi: 10.1080/00934690.2018.1423830.

Huang X, Dai L (2022) **The Overview of Crop Germplasm Resources in Yunnan.** Science Press, Beijing, China.

Institute of Botany, the Chinese Academy of Sciences (IBCAS), Institute of Changchun Geography, the Chinese Academy of Sciences (ICGCAS) (1988) *Xizang vegetation.* Science Press, Beijing, China.

Korneliussen TS, Albrechtsen A, Nielsen R (2014) **ANGSD: Analysis of Next Generation Sequencing Data.** *BMC Bioinformatics* 15(1), 356. doi: 10.1186/s12859-014-0356-4.

Lane P (2005) **Barbarous tribes and unrewarding gyrations? The changing role of ethnographic imagination in African archaeology.** In: Stahl AB (ed) African Archaeology: A Critical Introduction. Blackwell, Oxford, pp. 24-54.

Li H, Durbin R (2010) **Fast and accurate long-read alignment with Burrows-Wheeler transform.** *Bioinformatics* 26(5): 589-595. doi: 10.1093/bioinformatics/btp698.

Li H, Handsaker B, Wysoker A, Fennell T, Ruan J, Homer N, Marth G, Abecasis G, Durbin R, & 1000 Genome Project Data Processing Subgroup (2009) **The sequence alignment/map format and SAMtools.** *Bioinformatics* 25(16): 2078-2079. doi: 10.1093/bioinformatics/btp352.

Mabhaudhi T, Chimonyo VGP, Hlahla S, Massawe F, Mayes S, Nhamo L, Modi AT (2019) **Prospects of orphan crops in climate change.** *Planta* 250: 695-708. doi: 10.1007/s00425-019-03129-y.

Martin M (2011) **Cutadapt removes adapter sequences from high-throughput sequencing reads.** *EMBnet.journal* 17(1): 10-12. doi: 10.14806/ej.17.1.200.

Martinez EA, Fuentes F, Bazile D (2015) **History of quinoa: Its origin, domestication, diversification and cultivation with particular reference to the Chilean context.** In: Murphy KS, Matanguihan J (eds) Quinoa: Improvement and Sustainable Production. Wiley-Blackwell, Hoboken, pp. 19-24.

Meier zu Verl C, Meyer C (2024) **Ethnomethodological ethnography: Historical, conceptual, and methodological foundations.** *Qualitative Research*

24(1): 11-31. doi: 10.1177/14687941221129798.

Min R (2013) **Comprehensive Study on Jianchuan Haimenmen Site.** *Academy* (15): 6-9. doi: 10.3969/j.issn.1674-4810.2013.15.004.

Murphy K, Matanguihan J (2015) **Quinoa: Improvement and sustainable production.** Wiley-Blackwell, Hoboken, pp. 2-11.

Partap T, Joshi BD, Calwey NW (1998) **Chenopods: *Chenopodium* spp.** International Plant Genetic Resources Institute (IPGRI), Rome, Italy.

Partap T, Kapoor P (1985a) **The Himalayan grain chenopods. II. Comparative morphology.** *Agriculture Ecosystems Environment* 14(3-4): 201-220. doi: 10.1016/0167-8809(85)90036-2.

Partap T, Kapoor P (1985b) **The Himalayan grain chenopods. I. Distribution and ethnobotany.** *Agriculture, Ecosystems & Environment* 14(3-4): 185-199. doi: 10.1016/0167-8809(85)90035-0.

Pirzadah TB, Malik B (2020) *Pseudocereals as super foods of 21st century: Recent technological interventions.* **Journal of Agriculture and Food Research** 2, 100052. doi: 10.1016/j.jafr.2020.100052.

Popenoe H, King S, León J, Sumar KL, Vietmeyer N, Dafforn M (1989) **Lost crops of the Incas.** The National Academy Press, Washington, USA.

Rambaut A (2018) **FigTree v.1.4.4.** [`http://tree.bio.ed.ac.uk/software/figtree`] Accessed 15 February 2025.

Repo-Carrasco R, Espinoza C, Jacobsen SE (2003) **Nutritional value and use of the Andean crops quinoa (*Chenopodium quinoa*) and kañiwa (*Chenopodium pallidicaule*).** *Food reviews international* 19(1-2): 179-189. doi: 10.1081/FRI-120018884.

Safford WE (1918) ***Chenopodium nuttalliae*, a food plant of the Aztecs.** *Journal of the Washington Academy of Sciences* 8(15): 521-527. `https://www.jstor.org/stable/24521339`.

Song J, Gao Y, Tang L, Zhang Z, Tang MH, Xu H, Wangyal T, Yuan H, Li L, Li Y, Wangdue S, Liu X, Lu H (2021) **Farming and multi-resource subsistence in the third and second millennium BC: archaeobotanical evidence from Karuo.** *Archaeological and Anthropological Sciences* 13, 47. doi: 10.1007/s12520-021-01281-9.

Stamatakis A (2014) **RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies.** *Bioinformatics* 30(9): 1312-1313. doi: 10.1093/bioinformatics/btu033.

Tamura K, Stecher G, Kumar S (2021) **MEGA11: Molecular Evolutionary Genetics Analysis Version 11.** *Molecular Biology and Evolution* 38(7): 3022-3027. doi: 10.1093/molbev/msab120.

Tang L, Lu H, Song J, Wangdue S, Chen X, Zhang Z, Liu X, Boivin N, Spengler RB (2021) **The transition to a barley-dominant cultivation system in Tibet: First millennium BC archaeobotanical evidence from Bangga**. *Journal of Anthropological Archaeology* 61, 101242. doi: 10.1016/j.jaa.2020.101242.

Tang Y, Tsao R (2017) **Phytochemicals in quinoa and amaranth grains and their antioxidant, anti-inflammatory, and potential health beneficial effects: a review.** *Molecular nutrition & food research* 61(7), 1600767. doi: 10.1002/mnfr.201600767.

Tibet crop variety investigation group (1987) **Collection on Tibetan Crop Varieties investigation.** China Agricultural Science and Technology Press, Beijing, China.

UN (2011) **Resolution 66/221, International Year of Quinoa.** [`https://digitallibrary.un.org/record/724509?v=pdf`] Accessed 15 February 2025.

Wang B (2012) **Discussion on the cultivation model of *Chenopodium formosanum* in Taiwan Province.** *Agricultural Newsletter of Taitung* 80: 1-4.

Wang T (1983) **Coarse grain crop resources in Shannan and Qamdo regions of Tibet.** *Crop Germplasm Resources* (04): 21-24. doi: 10.19462/j.cnki.1671-895x.1983.04.005.

Wang T (1995) **Exploitation and protection of crop germplasm resources in Tibetan Plateau.** In: Proceedings of the symposium on the Tibetan Plateau and global change. China Meteorological Press, Beijing, pp. 155-160.

Wang Y (2018) **Agricultural transformation from millet to *Chenopodium*: Community resilience of a tea group in post disaster reconstruction.** *Journal of North Minzu University* (3): 136-143. doi: 10.3969/j.issn.1674-6627.2018.03.019.

Wang Y, Yang Q, Yang J, Sharma S, Huang Y, He W, Tsring T, Hu S, Chen S, Sun Q, You T, Gu Z, Tsho Y, Wangdue S, Chen Z, Gao Y, Yang X, Chen F (2024) **The evolution of cropping structure in prehistoric Xizang**. *Science Bulletin* 69(24): 3959-3967. doi: 10.1016/j.scib.2024.06.025.

Wilson HD, Heiser CB (1979) *The origin and evolutionary relationships of 'huauzontle' (**Chenopodium nuttalliae** Safford), domesticated chenopod of Mexico.* **American Journal of Botany** 66(2): 198-206. doi: 10.2307/2442525.

Xue Y, Martello RD, Qin L, Stevens CJ, Min R, Fuller DQ (2022) *Post-Neolithic broadening of agriculture in Yunnan, China: Archaeobotanical evidence from Haimenkou.* **Archaeological Research in Asia** 30, 100364. doi: 10.1016/j.ara.2022.100364.

Yang Z, Xu L, Wen X (2023) **The cultivation and utilization of *Chenopodium giganteum*, a specialty niche crop.** *Xiandai Nongcun Keji* (1): 17-18. doi: 10.3969/j.issn.1674-5329.2023.01.013.

Yuan Y, Liu G, Chen L (2008) **Research progress in crop mixtures.** *Crop Research* 22(S1): 381-385. doi: 10.16848/j.cnki.issn.1001-5280.2008.s1.020.

Received: 15 February 2025
Accepted: 02 October 2025
Published: 14 October 2025

Editor: Guillaume Odonne

Additional Files

Add File 1. Information of Maturase K reference sequences for 68 *Chenopodium*.

Species	GenBank ID	Number
Chenopodium formosanum	NC_086915	1
Chenopodium acuminatum	MT917946, MK954210, MF063899, MF063898 MF063897, MF063896, MF063895, MF063894	8
Chenopodium album	HM850763, GQ434131, KX133100, KX789384 KY952362, MF063900, MF063901, MF063902 MF063903, MF063904, MF063905, MF063906 MF063907, MF063908, MF063909, MF063910 MF063911, MF063912, MF063913, MF063914 MF063915, MF063916, MF063917, MF063918	24
Chenopodium hybridum	MT917950, MK925691, MK926222	3
Chenopodium iljinii	HE855667	1
Chenopodium rubrum	JN895422, JN895576, JN895577, HE855623 HE855624, HE855625, HE855626	7
Chenopodium serotinum	MK954211, MF063951, MF063950, MF063949 MF063948, MF063947	6
Chenopodium strictum	KX677328, MK519907	2
Chenopodium urbicum	JN895580, JN895425, HE855629, HE855630	4
Chenopodium pallidicaule	HE855654, HE855653	2
Chenopodium quinoa	KF318982, KF318991	2
Chenopodium berlandieri ssp *nuttalliae*	PP952099	1

Add File 2. Chenopods crops cultivation in different villages in Mêdog County.

Sites	**Cropping practice**				**Intercropping crops**	**Sowing**	**Harvest**	**Altitude**	**Fertilization**
	Pure	Multistorey	Horticultural	Field					
Yadong		✓	✓	✓	corn, pepper, lemon, cabbage, foxtail millet, common millet, finger Millet, tea tree	March	October	1006 m	manure & inorganic fertilizer
Mêdog		✓	✓		corn, pepper, long beans, lemon	March	October	1112 m	manure
Deguo		✓		✓	corn, sesame, taro, sweet potato, pepper	March	October	856 m	no fertilization
Bari		✓	✓		corn, amaranth, scallion, peppers	April	November	1646 m	unknown
Damu	✓	✓	✓		corn, pepper, dryland rice, scallion, snap bean, long beans	April	November	1575 m	no fertilization
Sangzhenka		✓	✓		beans	April	November	1978 m	unknown
Deji		✓	✓		corn, pepper, taro, peach, sorghum	April	November	1711 m	manure