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FIELD DISTRIBUTION AND HABITAT OF *SILIQUAMOMUM TONKINENSE*, A PLANT SPECIES WITH EXTREMELY SMALL POPULATIONS

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Abstract

Siliquamomum tonkinense is one of the extremely small populations species in China. To support subsequent artificial propagation and reintroduction research of the species, we conducted a field survey across its southeast Yunnan province distribution to assess the current status and drivers of the decline of remaining populations. Geographical distribution, habitat environment, root soil properties, and population characteristics of *S. tonkinense* were examined. We found that the species was mainly distributed in the forested valleys of Ma Lipo, Maguan County, Wenshan Prefecture, Yunnan Province, and Hekou County, Honghe Prefecture, Yunnan province. Specifically, the distribution was from 103° 33′ 85″ to 104° 78′ 58″ E and 22° 57′ 36″ to 23° 03′ 43″ N, and at elevations 546.81-1003.42m but concentrated between 600.54-892.28m. Climate in the distribution area is warm and humid with an average annual temperature of 16.50-17.6°C. The highest and lowest temperatures were 32.4°C and -4.0° C, respectively. Annual precipitation was 1068.0-1410.5mm, mostly from May to September. Average annual relative humidity was 71-84%. *S. tonkinense* grows under the forest with dense vegetation and rich hydrothermal conditions. Populations outside of reserves is mostly close to roads and villages with high human activity, which presents a threat to those populations. Examined populations were mostly declining with low seed germination percentages and few newly germinated seedlings. Plants were scarce in each population, and the populations lacked any clear capacity for natural recovery. Soil was mainly black humus with a pH of 4.0-5.9. Organic matter, nitrogen nutrition, and rapidly available potassium were rich, but the content of other elements was lower. This research established the current geographical distribution, population status, and main ecological characteristics of *S. tonkinense*, and provided a scientific basis for guiding future protection, breeding efforts, and reintroduction.

Key words: *Siliquamomum tonkinense*; Habitat, Biogeography, Soil physical and chemical properties.

Introduction

Plant species with extremely small populations (PSESP) refers to the distribution of wild plants with a narrow or intermittent distribution, due to their own factors or long-term interference by external factors, showing the population degradation and the number of continuous reductions, the number of populations and individuals are very small, has been below the stable survival limit of the minimum survival population, and at any time on the verge of extinction of wild plants(Sun, 2018; Li *et al*., 2023). Age structures for such species are often shifted towards older, non-reproductive classes, and spatial distributions are typically aggregated (Miao *et al*., 2008). For the lack of individuals at reproductive ages, the opportunities to expand ranges and increase population sizes are limited. Most endangered plants have poor seed quality, low thousand-grain weight, poor resistance to change, and low conversion rates of seeds to seedlings, all of which means for small numbers of 1-year-old seedlings and a lack of subsequent resources in the population (Zhong *et al*., 2011). Because of their reduced populations, PSESP may be acutely impacted by species with which they have negative interactions, such as grazers (Ye *et al*., 2022).

Recent rapid human population growth presents many great challenges for the protection of biodiversity. Overexploitation, habitat loss (Liu *et al*.,2023) invasive species, and climate change have pushed many species of flora and fauna to being threatened or endangered with extinction (Mace *et al*., 2011; Ceballos *et al*., 2015). Species with reduced populations are particularly difficult to recover when their habitat is reduced or fragmented because of the resulting obstacles to natural recovery and dispersal (Ma *et al*., 2012; Newbold *et al*., 2015). Information about current distributions and the environmental drivers of those distributions can provide vital guidance for endangered species protection and recovery efforts (Zhang *et al*., 2003).

Siliquamomum tonkinense is a PSESP in the Family Zingiberaceae. It is a perennial herb with erect stems and tall plants, usually 0.6-2m, and is mainly found in Pingbian, Maguan, Hekou, Menghai and other counties in Yunnan Province, China, under tropical valley rainforest or other forest edges at an altitude of 600-800 m (Flora of China, 1981). Microclimate requirements for *S. tonkinense* include high humidity and low direct sunlight. Due to continued habitat loss of this narrowly distributed species, *S. tonkinense* populations are threatened with extirpation. Small, disparate remaining populations mean opportunities for gene flow are limited and possibilities for recovery are less likely without human intervention (Slatkin, 1987). An endemic species to the Yunnan Province, *S. tonkinense* is listed as a national Class II key protected wild plant (approved by the State Council on August 4,1999) and also in the *International Union for Conservation of Nature Red List of Endangered Species* (IUCN) as a critically endangered species (CR).

We investigate the geographical distribution, climatic requirements, habitat, and population characteristics of *S. tonkinense* using data from existing literature and adding new data from field investigations. Physical and chemical properties of soil occupied by *S. tonkinense* population were tested to better understand geographical distributions and habitat requirements, and to provide a basis for the subsequent research, breeding programs, and the overall recovery of populations.

Material and Methods

We collected data on known distributions of *S. tonkinense* by consulting the *' Flora of China', 'Flora of Yunnan', ' Research and Protection of Wild Plants* with *Extremely Small Populations in Yunnan'*, and the *Digital Herbarium of China Records*. We conducted multiple surveys from 2022 to 2023, during the flowering period and fruiting period of the population. In the process of investigation, we conducted a detailed field survey of S. *tonkinense* in the protected area using the line transect methods, recorded the basic situation of S. *tonkinense* population, collected live plants, soil samples and molecular materials. For detected populations, we collected data on geographical coordinates, altitude, topography, geomorphology, hydrothermal conditions, habitat, disturbance factors, population size, and structure.

Longitude, latitude, and altitude were obtained using a GPS locator. Through the *China Meteorological Data Network* ground climate standard value data set (1985-2020) and local climate data records, we obtained climate data for the sites. We collected soil near the roots of five populations that were accessible and had strong representativeness, and, after air drying, these samples were sent to the *Yunnan Jingheng Agricultural Analysis and Testing Co., Ltd*. for testing.

Results

Geographical distribution: *S. tonkinense* is distributed from 103° 33′ 85″ to 104° 78′ 58″ E and 22° 57′ 36″ to 23° 03′ 43″ N, with latitude and longitude spanning 1° 44′ 73″ and 6′ 07″, respectively, and is located in the subtropical monsoon climate zone north of the Tropic of Cancer. We found 7 populations across Xinjie Town, Yuanyang County, Hani and Yi Autonomous Prefecture, Malipo County, Wenshan Zhuang and Miao Autonomous Prefecture, and Maguan County, Wenshan Zhuang and Miao Autonomous Prefecture, all of Yunnan Province. The highest concentrations were in Malipo and Maguan counties (Fig. 1).

Previous records suggested that *S. tonkinense* grows in clusters the tropical valley rainforest or forest edge at an altitude of 600-800m. Among the seven populations found by our survey, the vertical distribution was 546.81-1003.42m, with the lowest at Maguan Gulinqing Jinchang (546.81m) in Wenshan, Yunnan, the highest at Yuanyang New Street (1003.42m) in Honghe, Yunnan, and the high concentration at 600.54-892.28m.

S. tonkinense is distributed in the subtropical monsoon climate zone. The average annual temperature in their distribution is 16.5-17.6°C, the coldest monthly temperature is 9.7-10.2°C, the mean temperature of the warmest month is 20.6-23°C, the extreme minimum temperature is -4.0°C, and the extreme maximum temperature is 32.40°C (Table 1). These data imply the species is adapted for growth in widely variable temperatures, and that temperature does not have a strong effect on driving its distribution. The annual precipitation experienced by the species is high, ranging from 1068.0-1410.5mm, and the annual relative humidity is 71-84%. Rainfall is concentrated in the summer, May to September, and the surface water under the river valley forest is abundant. Water availability is consistent in the distribution and there is no obvious dry and wet season. *S. tonkinense* grows on thick humus soil on the forest floor where the surface water is sufficient and the soil layer is thick. The distribution is located in mountainous areas where the vegetation growth is high. Sufficient surface water, high humidity, and thick surface soil layers all seem to be important contributors to the growth of *S. tonkinense* (Gao *et al*., 2019).

Topography and habitat characteristics: *S. tonkinense* is mainly distributed in valley areas with sufficient hydrothermal conditions and where rainwater collects. High humidity, moist surface soil, dense vegetation, low exposure to direct sunlight, and thick humus soil are consistent conditions for current populations. Populations in Xia Xingqing, Huashan, and Abang in the lower part of MaliPo slope are all near valley streams where rainwater collects. The topography of Bojia, Jinchang and Mogangqing in Maguan Gulinqing is mainly the low-lying tropical valley with karst substrate, which forms a relatively closed small pattern habitat, and the area has sufficient heat, abundant surface water, thick soil layer under the forest, low human disturbance, and relatively high habitat protection (Fig. 2)

Table 1. Climate variation in distribution of *Siliquamomum tonkinense*.

Distribution	AAT (°C)	ATCM (°C)	ATHM (°C)	ELT (°C)	EHT (°C)	FFP (d)	AP (mm)	ARH (%)	HS (h)
XinJie	16.50	10.2	20.6	-2.5	32.40	364	1410.5	71	1753.2
MaGuan	16.9	9.7	21.7	-4.0	32.3	327	1345.0	80	1804.0
MaLiPo	17.6	10.1	23	1.1	30.0	330	1068.0	84	1517.3

AAT: Average annual temperature; ATCM: Average temperature in the coldest month; ATHM: Average temperature in the hottest month; ELT: Extreme low temperature; EHT: Extreme heat temperature; FFP: Frost-free period; AP: Annual precipitation; ARH: Annual relative humidity; HS: Hours of sunshine

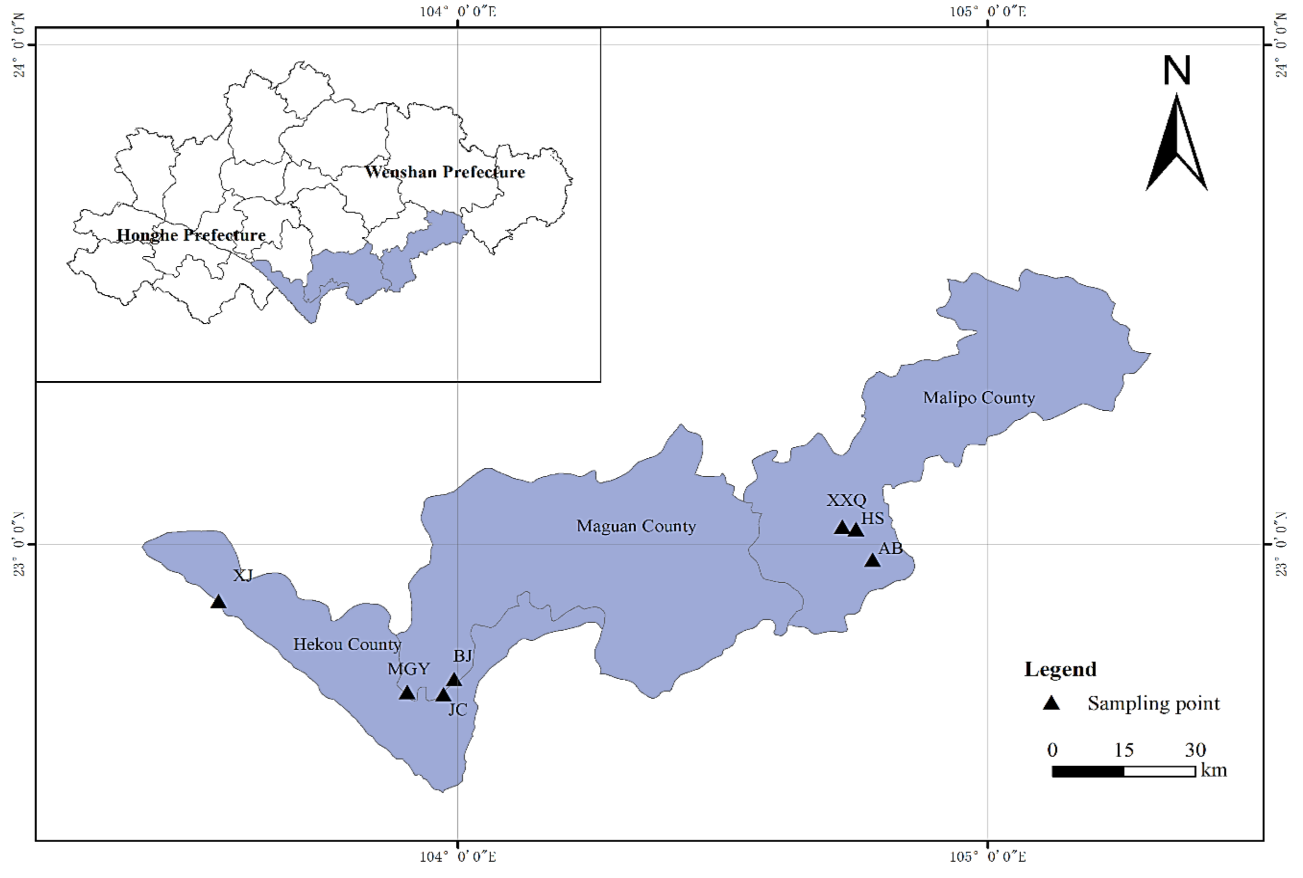

Fig 1. Distribution of *Siliquamomum tonkinense* and sample sites examined.

Fig. 2. Morphological characteristics and habitat of *Siliquamomum tonkinense*. **(a)** Inflorescences of *S. tonkinense* **(b)** *S. tonkinense* buds **(c)** Greenhouse transplants of flowering *S. tonkinense* **(d)** Fruit of the *S.tonkinense* **(e)** and **(f)** Habitat of the *S.tonkinense*.

Table 2. Soil characteristics for *Siliquamomum tonkinense* populations.

Population	pH	OM ($g\cdot kg^{-1}$)	TN ($g\cdot kg^{-1}$)	TPH ($g\cdot kg^{-1}$)	TPO ($g\cdot kg^{-1}$)	HN ($mg\cdot kg^{-1}$)	AP ($mg\cdot kg^{-1}$)	RAP ($mg\cdot kg^{-1}$)	EC ($^{1/2}ca2^{+}$)/kg	EM ($^{1/2}mg^{2+}$)/kg
XinJie	4.8	93.5	4.53	0.92	19.4	595	15.0	216	3.5	1.3
XiaXingQing	4.0	56.5	2.84	0.36	21.8	286	9.4	80	25.2	0.5
A′Bang	4.3	48.8	2.58	0.36	40.1	322	5.8	64	2.3	0.4
BoJia	5.5	39.4	2.42	0.26	40.9	337	2.6	153	6.1	1.5
MoGangQing	5.9	50.9	2.86	0.33	36.1	323	2.1	85	10.1	2.4

OM: Organic matter; TN: Total nitrogen; TPH: Total phosphorus; TPO: Total potassium; HN: Hydrolyzed nitrogen; AP: Available phosphorus; RAP: Readily available potassium; EC: Exchangeable calcium; EM: Exchangeable magnesium

Table 3. Characterization of studied *Siliquamomum tonkinense* populations.

Population	Latitude (N)	Longitude (E)	Elevation (m)	Population characteristics
XinJie	22°95′61″	103°56′99″	1014.63	There were 3 plant groups, Group 1 consisted of 13 plants with an average plant height of 115cm, one fruit and 23.5cm fruit length. Group 2 consisted of 7 plants with an average plant height of 95cm, one fruit and a fruit length of 17.8cm. Group 3 consisted of 10 plants with an average plant height of 87cm and no fruit.
XiaXingQing	23°03′50.43″	104°72′57.43″	884.47	There were 4 plant groups , Group 1 consisted of 7 plants with an average plant height of 58cm, 2 fruiting plants and 13.2cm fruit length. Group 2 consisted of 13 plants with an average height of 92cm and no fruit in inflorescence. Group 3 consisted of 8 plants with an average height of 95cm and flower buds. Group 4 consisted of 11 plants with an average plant height of 92cm.
HuaShan	23°03′02.66″	104°75′20.78″	906.14	There were 9 plants in 1 population, the average plant height was 116cm, the fruit was 1, and the fruit length was 14.2cm.
A’Bang	22°96′93.62″	104°78′26.58″	626.85	There were 15 plants in 1 population with an average plant height of 116cm and no fruit.
BoJia	22°73′03.05″	103°99′29.82″	600.54	The were 2 plant groups, Group 1 consisted of 9 plants with average plant height 153cm, fruitless. Group 2 consisted of 3 plants with an average plant height of 80cm and no fruit.
JinChang	22°69′90.53″	103°97′33.26″	546.81	There were 2 plants groups, Group 1 consisted of 4 plants with an average plant height of 82cm and no fruit. Group 2 consisted of 5 plants with an average plant height of 97cm and no fruit.
MoGangQing	22°70′35.70″	103°90′48.84″	637.74	There were 3 plants, Group 1 consisted of 8 plants with an average plant height of 159cm, and 4 of the results were cracked. Group 2 consisted of 6 plants with an average height of 195cm, and 2 of the results were cracked. Group 1 consisted of 26 plants with an average plant height of 96cm and no fruit.

Physical and chemical properties of soil: The soil of the focal Ginger population is black humus. Acidity was high, with pH values 4.0-5.9 (Table 2). Compared to the nutrient classification standards of the second national soil survey (Soil survey technology in China, 1992), soil organic matter, total nitrogen, and hydrolyzable nitrogen were rich and significantly higher than the first-grade standard. Phosphorus content, on the other hand, low. Except for the population in Xinjie, which was grade 2, the other populations were at levels of grade 4 and 5 or less, and available phosphorus and readily available potassium were also at grade 3 or less. Exchangeable calcium and magnesium content were low, both being below the level 5 standard. This may be related to the habitat being mostly under forest with dense vegetation and good hydrothermal conditions. In addition, the composition of available potassium in different populations was very different. The minimum value was detected in Abang (64 $mg \cdot kg^{-1}$) and the maximum value was in Xinjie (216 $mg \cdot kg^{-1}$), a difference of 3.4X. This may be related to the composition of parent rock and metabolic processes of microorganisms and plants in the soil. In summary, the soil is rich in organic matter, nitrogen nutrition, and available potassium, and shows strong acidity, but the content of other nutrients is low or lacking.

Population characteristics and current status: In this survey, there were 7 *S. tonkinense* populations and 16 plant groups (Table 3), including 10 with less than 10 species, with 10-20 plants, and 1 with more than 20 plants. According to the literature, *S. tonkinense* is a perennial herb with upright stems and relatively tall adult forms, usually 0.6-2m. Among the 7 populations investigated in this survey, the tallest individual was in Mogangqing No.2 at 195cm and the shortest was in Xiaxingqing No.1 at 58cm. Average plant heights of populations ranged from 90-120cm. In this survey, 80% of *S. tonkinense* populations were composed of adult plants. Seedlings were rare, and the age structures suggested the natural regeneration of the populations is unlikely. The seedling stage is one of the most important stages in plant life cycles. The establishment and survival of seedlings are related to the size and persistence of the population, and seedlings are key for not only renewal and continuation of individual species’ populations, but also maintenance of high biodiversity communities (Zhang *et al.*, 2005). Only a small number of new seedlings were found in Xiaxingqing, Bojia and Mogangqing, and no new seedlings were found in other populations. In the population with seedlings, they accounted for less than 30% of the population. We also found that the seed setting rate of *S. tonkinense* was low, and only a small amount of fruit could be observed in a limited portion of the population. We found the flowering period was different from October to June of the next year, and the fruiting period lasted until September of the next year. These findings may explain both the limited number of new seedlings and the degradation of age structures of *S. tonkinense*.

Discussion

According to the '*Yunnan National Key Protected Wild Plants*', *S. tonkinense* populations are rare. In our survey, 7 populations were found in Malipo, Maguan and Honghexinjie in Wenshan, Yunnan, including 3 populations and 6 plant groups in Malipo, and 3 populations and 7 plant groups in Maguan. We assessed the current status of populations and found that, like many rare and endangered plants, the distribution is narrow and remaining populations are few. Providing clear spatial data will be beneficial for formulating conservation strategies and a basis for systematic protection efforts. Information on the population status (Song et al., 2023), location of habitats, soil and climate characteristics, potential distribution, and other factors such as age structure is crucial for establishing a species' conservation priority, habitat requirements, drivers of species distribution, and the development of future conservation measures (Varghese *et al*., 2006). Although we provide information about the current distribution of *S. tonkinense*, we notably only examined current protected areas. Future work using our findings as a baseline should aim to determine the existence of the species outside of these protected areas as additional populations may prove vital in conservation and recovering efforts (Tang *et al*., 2019).

There are two primary categories of factors that drive PSEP to the brink: internal and external. Internal mechanisms include lacking the ability to obtain the nutrients and energy needed for growth and reproduction, such as through the disruption of reproduction (Tang *et al.*, 2011), pollen limitation (Hu *et al*., 2022), low fruit setting rate (Szczecińska *et al*., 2016), low seed germination percentage, high seedling mortality, low genetic diversity, and low adaptability, which leads to an inability to achieve population renewal (Deng *et al*., 2020; Li *et al*., 2014). External factors include historical changes in geology, glacial effects (Wang *et al*., 2023), cattle grazing, human excavation, habitat fragmentation, and habitat degradation (Sun *et al*., 2005). *S. tonkinense* lives in the humus soil under forests in the valleys and river valleys of the subtropical monsoon climate. The hydrothermal conditions are abundant, and the soil has good physical and chemical properties and rich nutrients. Climate and soil conditions are not likely limiting the growth and development of *S. tonkinense*. Due to their location in dense forest and an extended fruiting period, the species is intensely exposed to frugivore activity. *S. tonkinense* also has a low fruit-bearing percentage, low germination rate, few new seedlings, and a single reproduction mode. Grass fruits are frequently planted under the forest by nearby residents for sustenance, and this which destroys *S. tonkinense* habitat. Hence, *S. tonkinense* populations are in decline due to both internal and external factors.

Because their extant distribution in the wild is scarce, *S. tonkinense* populations will require protection measures to avoid extinction. First of all, protecting and restoring existing habitats is inherently necessary for species survival (Benayas *et al*., 2009). Like most species, habitat destruction is a primary cause of the decline of *S. tonkinense* populations, particularly that resulting from cultivation of *Amomum tsaoko*, a species with similar habitat requirements. Secondly, both the in-situ conservation and ex-situ protection of *S. tonkinense* individuals are necessary. Populations surveyed for this study were located near roads and villages with high human activity. While protective measures outside of reserves might be more difficult, implementing such measures inside of reserves can and should done as soon as possible. Seedlings cultured in nurseries should be used to boost wild populations and establish additional populations to ensure long-term viability (Liao *et al*., 2023). The last two protective measures of note would be the establishment of a germplasm resource bank of *S. tonkinense* and increasing public awareness. Germplasm resources are not only useful for effective artificial cultivation of seedlings for reintroduction (Guerrant *et al*., 2007; Goderfriod *et al*., 2011), but they are also important links in research and utilization. For occupants of reserves, education on biodiversity, habitat protection, and the laws and regulations of the state regarding the protection of rare and endangered animals and plants would be helpful. The long-term viability of *S. tonkinense* hinges on comprehensive coordination by researchers, regulatory agencies, and the public, and the success of this requires the rapid formulation of efficient and effective protection measures.

Conclusions

In this study, 7 populations of *S. tonkinense* were found, the species' distribution and resource quantity in Yunnan were examined, and a clear documentation of their population status was established. At the same time, it was found that the tolerance of *S. tonkinense* to temperature was relatively wide, and it was mostly distributed under forests and river valleys with rich humus soil and high hydrothermal conditions. The distribution is narrow, the habitat is fragile, the numbers of and sizes of existing populations are very small, and most are declining. The causes are related to both the species' germination rate and human interference. Our research effectively characterized the geographical distribution, resource status, and main ecological characteristics of *S. tonkinense*, and provided a strong scientific basis for future resource protection, breeding programs, and site selection for restoring populations.

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