

Phenotypic Variations in *Camptotheca Decaisne*

Shiyu Li*

National Center for Pharmaceutical Crops, Arthur Temple College of Forestry and Agriculture, Stephen F. Austin State University, Nacogdoches, TX 75962, USA

Abstract: Variations in shape, size, color, and texture of fruits, shape and size of cotyledons, shape, venation pattern, surface and margin characters of mature leaves of 40 populations (either living plants or herbarium specimens) representing all known taxa of *Camptotheca* have been analyzed within and between populations. A majority of the observed phenotypic variations for most morphometric measures was among populations. This is probably due largely to selfing and related matings common to both natural and cultivated populations, resulting in the current population structure that now displays a highly fragmented distribution pattern. Trees of different generations from the same seed source but growing in different locations demonstrated minimal variations in both means and frequency distributions of leaf vein number, fruit length, fruit colors, and cotyledon vein number. The trees of different taxa even growing in the same location have significant difference in main characters particularly fruit color and texture. The key diagnostic characters for the identification of taxa of *Camptotheca* are leaf shape, venation type, vein number, stoma size and frequency, outer stomatal rim, subsidiary cell number, and gland size; cotyledon shape, venation type, and vein number; and the fruit surface texture, disc, length, and color.

Keywords: *Camptotheca*, cotyledon shape, fruit color, fruit length, fruit surface texture, gland length, leaf surface, leaf vein number, leaf venation type, phenotypic variations,

INTRODUCTION

Evolutionary ecologists have stressed the importance of understanding the expression and the components of phenotypic variation among and within populations [1-6] and have proposed that a hierarchical perspective may provide insights into the evolutionary history of a species [3]. Phenotypic variations of the *Camptotheca* or any species of the genus have never been analyzed. The primary objectives of this study are: (1) to investigate the pattern of phenotypic variation within and among populations; (2) to determine whether these traits examined are taxon specific and can be used as diagnostic key characters in the genus; and (3) to clarify taxonomic and systematic relationships.

MATERIALS AND METHODS

Plant Materials

General information on the plant materials collected from 1992 to 1995 for this study is presented in (Table 1). The letters A, B, L, and Y followed by numbers designate populations. The letter A refers to the plant materials of *C. acuminata* var. *acuminata* collected directly from China: A1-9 represents the dominant seed sources in current plantations in southern China. A10-16 refers to herbarium specimens. The letter B refers to the materials of *C. acuminata* var. *acuminata* collected from the United States: B1-6 represents mature trees in the country (in 1995). B3a and B3b were collected from the same tree San Antonio, Texas in 1993 and

1995, respectively, and B6a and B6b were collected from the same tree in Huntington, California in 1992 and 1995, respectively. Three of these samples represent first generation (B2) and the third generation (B3 and B4) from the same ancestor see Fig. (1). The letter L refers to populations of *C. lowreyana*: L1, L4, L5, and L6 are herbarium specimens, while L2 and L3 were collected from China. The letter Y refers to population of *C. yunnanensis*: Y1 was collected in China, and Y2 is the herbarium specimen. In addition, the only sample of *C. acuminata* var. *tenuifolia* is the type specimen. The data on *C. acuminata* var. *rotundifolia* are drawn from the original description.

Climatic data including annual precipitation and mean January temperatures were determined for each collection site from the local records (Table 2).

Fruit and Cotyledon Shapes and Sizes

All fruit from herbarium specimens were measured. For our collections, fruits from six individuals were measured, germinated, and grown in the same environments for fruit and seedling phenotypic analyses. For morphometric analysis, length and maximum width of 64 randomly selected mature dry fruit from six trees were measured. For seedling analysis, sample size was dependent on the seedlings available. Cotyledon venation type, lateral vein number, blade length, blade width at widest point, and widths at 1/5 distance from the apex and the base were measured at the age of three months.

Fruit Color and Texture

Color is usually diagnostic in plant taxonomy. Careful color evaluation is desirable for botanical descriptions. A

*Address correspondence to this author at the National Center for Pharmaceutical Crops, Arthur Temple College of Forestry and Agriculture, Stephen F. Austin State University, Nacogdoches, TX 75962, USA; Tel: 936-468-2071; Fax: 936-468-7058; E-mail: lis@sfasu.edu

Table 1. Description of the populations of *Camptotheca* surveyed (* Tree age = parent tree age when seeds were collected; ** Seedling age = age in 1997; ---- no data available/unapplicable; A = collections of *C. acuminata* from China; B = collections of *C. acuminata* from the United States; L = *C. lowreyana*; Y = *C. yunnanensis*; A1-9, B1-6, L1-3, and Y1 = living collections; rest = herbarium specimens).

Seed Source	Origin	Mature Tree Population Size	Seed Collection Year	Tree Age * (year)	Germinated Seedling Number	Seedling Age ** (year)
<i>C. acuminata</i> var. <i>acuminata</i>						
A1a. Dakengshan, Huaiji, Guangdong, China	Cultivated	24	1994	18	63	3
A1b. Dakengshan, Huaiji, Guangdong, China	Cultivated (same with A1a)	24	1996	20	33	1
A2. Gushui, Guangning, Guangdong, China	Cultivated (same with A1)	18	1994	18	154	3
A3. Pengzhou, Sichuan, China	Cultivated	c. 300	1994	>10	0	----
A4. Jintang, Sichuan, China	Cultivated	c. 500	1994	>10	22	3
A5. China (F.W. Schumacher Co, MA)	Cultivated	----	1994	>10	0	----
A6. Nanjing, Jiangsu, China	Cultivated	40	1994	25	850	3
A7. China (Lawyer Nursey, MT)	Cultivated	----	1994	>10	220	3
A8. Xuancheng, Anhui	Cultivated	----	1995	>10	350	3
A9. Zhejiang, China (Lawyer Nursey, MT)	Cultivated	----	1991	>10	26	6
A10. Emeishan, Sichuan, China (W. C. Cheng 2231)	----	----	1930	----	----	----
A11. Nanjing, Jiangsu, China (K. S. Chow 80271)	Cultivated	----	1980	----	----	----
A12. Hubei, China (A. Henry 7606)	----	----	1885-88	----	----	----
A13. Guangzhou, Guangdong, China (H. G. Yip 431)	Cultivated	----	1981	----	----	----
A14. Yungshien, Guangxi, China (Steward & Cheo 1180)	----	----	1933	----	----	----
A15. Guilin, Guangxi, China (Wan & Chow 79088)	Cultivated	----	1979	----	----	----
A16. Lushan, Jiangxi, China (David 866, isotype)	----	----	1868	----	----	----
A17. Botanical Garden, Guangzhou, China	Cultivated	3	1996	18	0	1
A18. Botanical Garden, Xian, Shaanxi, China	Cultivated	2	1996	8	24	1
A19. Changan, Shaanxi, China	Cultivated	20	1996	25	52	1
A20. Yangling, Shaanxi, China	Cultivated	5	1996	25	65	1
A21. China (unknown)	Cultivated	----	1996	----	----	----
<i>C. acuminata</i> var. <i>acuminata</i>						
B1. Chico, CA, USA (east tree)	Cultivated (from Jiangsu, China)	1	1992	58	----	----
B2. Chico, CA, USA (west tree)	Cultivated (same with B1)	1	1992	58	----	----
B3a. San Antonio, TX, USA	Cultivated (parent tree from B2)	1	1993	12	20	4
B3b. San Antonio, TX, USA	The same tree with B3a	1	1995	14	125	2
B4. Kingwood, TX, USA	Cultivated (same with B3)	1	1994	12	20	3
B5. Summerville, SC, USA	Cultivated (from Honolulu)	1	1994	24	50	3
B6a. Huntington, San Marino, CA, USA	Cultivated (local source)	1	1992	29	----	----
B6b. Huntington, San Marino, CA, USA	The same tree with B6a	1	1995	32	134	2
B7a. SFA Arboretum, Nacogdoches, TX, USA	Cultivated	1	1996	5	c. 600	1
B7b. SFA Arboretum, Nacogdoches, TX, USA	Cultivated (same tree with B7a)	1	1997	6	----	----
B8. SFA Exper. Forest, Nacogdoches, TX, USA	Cultivated	1	1996	5	0	----
<i>C. acuminata</i> var. <i>rotundifolia</i>	Natural (?)	----	----	----	----	----
<i>C. acuminata</i> var. <i>tenuifolia</i>	Natural	1	1996	----	12	2
<i>C. lowreyana</i>	Natural	----	1930	>10	----	----
L1. Lianxian, Guangdong, China (Gao 50863, type)	Natural	21	1994	>50	22	3
L2. Shiyang, Huaiji, Guangdong, China	Natural	28	1994	>100	35	3
L3. Dakengshan, Huaiji, Guangdong, China	Natural	----	1933	----	----	----
L4. Tongzhong, Huaiji, Guangdong, China (Tsang 23130)	Natural	----	1928	----	----	----
L5. Lechang, Guangdong, China (Jiang 1429)	Natural	----	1936	----	----	----
L6. Yaoshan, Guangdong, China (Wang 40005)	Natural	c. 10	1996	>80	54	1
<i>C. yunnanensis</i>						
Y1. Mengyang, Xishuangbanna, Yunnan, China	Cultivated	c. 250	1994	46	20	3
Y2. Simao, Yunnan, China (Henry 13433)	Cultivated	>100	1996	>30	11	1
Y3. Yangbi, Yunnan, China						
Y4. Kunming, Yunnan, China						

general description of color is inadequate today. To avoid human visual errors, remote sensing technology was applied in the fruit color analyses. Thirty dry fruit from each of 17 populations of *Camptotheca* were selected for chroma and surface texture analysis using remote sensing technology. The procedure involves the following steps: (1) optical digitization using a Sony DXC 327A video camera; (2) recording the digitized image using a "frame grabber" video board (video 7 band) installed in an HP 9000 Model 720 workstation; (3) image processing using ERDAS Imagine 8.1 (ERDAS Inc., 1990) software normalized for contrast and

brightness by setting the Histogram Contrast Adjustment to a linear curve indexed from 0 to 255 for all three channels (red, green, and blue) Fig. (1); (4) definition of the AOI (area of interest) of each fruit manually; (5) adding AOIs to each seed signature and then merging signatures through a classification protocol; and (6) displaying statistics (means and standard deviations) for all three (red, green, and blue) color channels for all seed signatures. Diagnostic differences in seed chroma were recorded indicating that multi-spectral analysis may be a productive area for further research in classifying visual indicators in plant taxonomy.

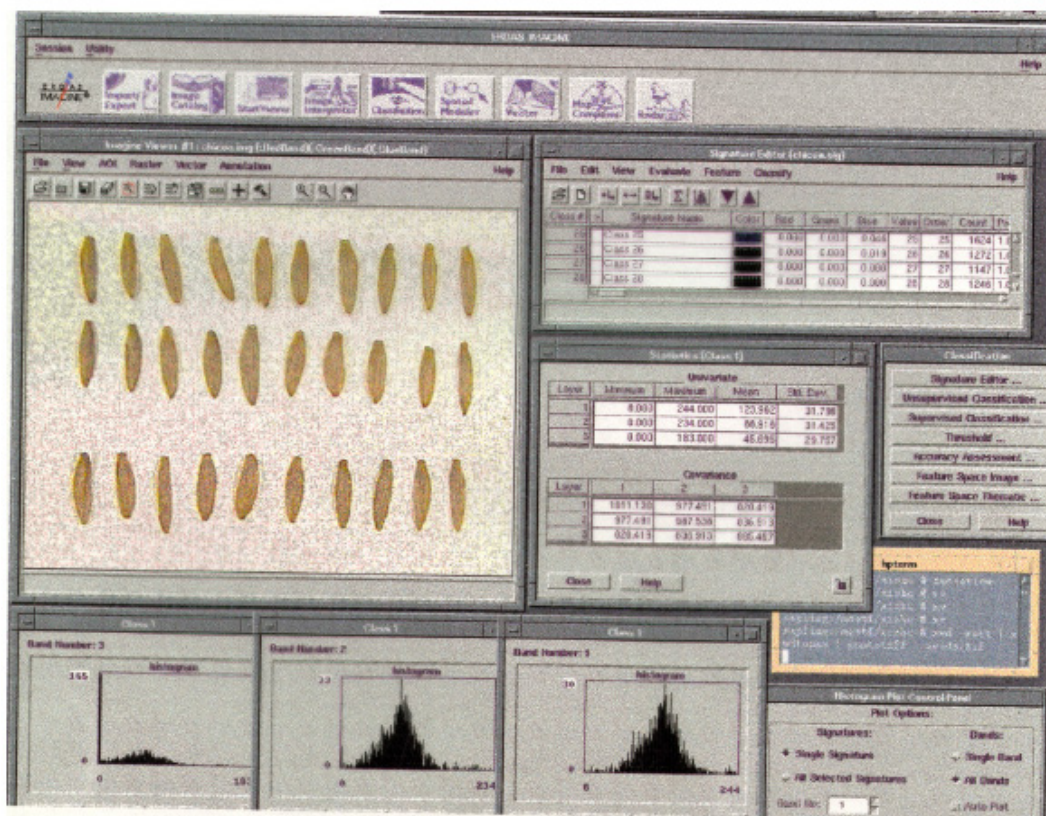


Fig. (1). Analysis of fruit color and size of *Camptotheca* using ERDAS Imagine 8.1 (ERDAS Inc., 1990).

Leaf Shapes, Venation Patterns, and Margin Characters

Each sample (64 mature leaves when possible) was selected from three trees for each of 17 populations/specimens of *C. acuminata* var. *acuminata*, six populations of *C. lowreyana*, and two of *C. yunnanensis*. Leaf shape, vein number, blade length, blade width at widest point, and blade widths at 1/5 the distance from the apex and from the base were recorded for each leaf. Leaf margin characters include three categories: serrate, slightly serrate, and entire was measured for all samples. Both parent trees and seedlings were measured when possible.

Statistical Analysis

Phenotypic variations of populations were compared using ANOVA and Tukey's Studentized Range (HSD) Test for each of the quantitative measures. Means, standard deviations, and variation ranges of all characters were calculated for all samples. Statistical significance levels employed are 0.05, 0.01, and 0.001. Frequency distributions of all characters in each sample are also presented because means-based methods tend to obscure overlap. Pearson correlation coefficients were calculated for major measures. Statistical analyses employed SAS version 6.03 (SAS Institute 1990) at SFASU.

RESULTS

Fruits (Seeds)

All observed samples of *C. acuminata* (var. *acuminata*, var. *rotundifolia*, and var. *tenuifolia*) have a rugose surface

and thick disc when dry. Fruit of *C. lowreyana* and *C. yunnanensis* have a smooth surface and thin disc relative to *C. acuminata* (Table 3). *C. lowreyana*, with a mean fruit length of 29.02 mm, similar to *C. acuminata* var. *tenuifolia* (mean length: 29.80 mm) has larger fruit than either *C. acuminata* var. *acuminata* (22.54 mm) (Fig. 2) or *C. yunnanensis* (22.96 mm). A frequency distribution of fruit length shows the same relationships Fig. (3). The ratio of fruit length by width also displayed significant differences among species: *C. lowreyana*, *C. acuminata* var. *acuminata*, and *C. yunnanensis* have mean ratios of 5.08, 4.18, and 3.61, respectively. In fact, ratio differences are largely contributed by the length differences among species.

A quantitative description of fruit color provides an important and objective measure of phenotypic variation and is diagnostic in taxon identification of *Camptotheca*. *C. yunnanensis* displays significant differences in red, green, and blue chroma from *C. acuminata* and *C. lowreyana* both in terms of means and distribution frequencies for the number of fruit measured (Table 4), Fig. (4).

Cotyledons

Venation type is constant within species, but different between *C. lowreyana* (pinninerved) and *C. acuminata* var. *acuminata* or *C. yunnanensis* (pinnipalmate). Vein number of the cotyledon is also an important character in distinguishing *C. lowreyana* (mean vein number: 6.91) from both *C. acuminata* var. *acuminata* (3.48) and *C. yunnanensis* (3.21) (Table 5); Fig. (5). Vein number is also independent of cotyledon size, the latter often varying with environment.

Table 2. Site descriptions of the populations of *Camptotheca* surveyed (---- no data available).

Population/Location	Latitude	Longitude	Altitude (m)	Lowest Temperature (°C)	Mean January Temperature (°C)	Annual Precipitation (mm)
<i>C. acuminata</i> var. <i>acuminata</i>						
A1a, b. Dakengshan, Huaiji, Guangdong, China	23°50' N	112°20' E	1270	- 5	7	2000
A2. Gushui, Guangning, Guangdong, China	23°45' N	112°25' E	850	- 3	7	2000
A3. Pengzhou, Sichuan, China	31°05' N	103°11' E	950	0	6	1300
A4. Jintang, Sichuan, China	30°49' N	104°22' E	900	- 3	6	1200
A5. China	----	----	----	----	----	----
A6 & A11. Nanjing, Jiangsu, China	32°04' N	118°47' E	75	- 9	2	1100
A7. China (unknown)	----	----	----	----	----	----
A8. Xuancheng, Anhui, China	31°00' N	118°45' E	200	- 7	3	1100
A9. Zhejiang, China	----	----	----	----	----	----
A10. Emeishan, Sichuan, China	----	----	----	----	----	----
A12. Hubei, China	----	----	----	----	----	----
A13. Guangzhou, Guangdong, China	23°08' N	113°20' E	20	3	14	1700
A14. Yunghsien, Guangxi, China	----	----	----	----	----	----
A15. Guilin, Guangxi, China	25°21' N	110°11' E	220	0	9	1950
A16. Lushan, Jiangxi, China	----	----	----	----	----	----
A17. Botanical Garden, Guangzhou, China	34°01' N	108°58' E	620	-11		
A18. Botanical Garden, Xian, Shaanxi, China			600	-18		
A19. Changan, Shaanxi, China						
A20. Yangling, Shaanxi, China						
A21. China (unknown)						
<i>C. acuminata</i> var. <i>acuminata</i>						
B1. Chico, CA, USA (east tree)	39°42' N	121°47' W	70	2	7	660
B2. Chico, CA, USA (west tree)	39°42' N	121°47' W	70	2	7	660
B3a, b. San Antonio, TX, USA	29°25' N	98°30' W	200	4	10	750
B4. Kingwood, TX, USA	29°55' N	94°50' W	20	- 1	12	1100
B5. Summerville, SC, USA	33°02' N	80°12' W	25	- 6	10	1200
B6a, b. Huntington, San Marino, CA, USA	34°07' N	118°06' W	200	5	13	550
B7a, b. SFA Arboretum, Nacogdoches, TX, USA	31°38' N	94°40' W	100	- 12	9	1200
B8. SFA Exper. Forest, Nacogdoches, TX, USA	?	?	----	----	----	----
<i>C. acuminata</i> var. <i>rotundifolia</i>	24°30' N	112°25' E	----	-5	6	2000
<i>C. acuminata</i> var. <i>tenuifolia</i>	24°47' N	112°28' E	750	- 7	5	1500
<i>C. lowreyana</i>	24°28' N	112°30' E	700	- 5	6	2000
L1. Lianxian, Guangdong, China	23°50' N	112°20' E	1270	- 5	7	2000
L2. Shiyong, Huaiji, Guangdong, China	24°05' N	112°25' E	1000	-5	6	2000
L3. Dakengshan, Huaiji, Guangdong, China	25°12' N	113°21' E	----	-7	5	2000
L4. Tongzhong, Huaiji, Guangdong, China	24°30' N	113°08' E	----	-7	5	2000
L5. Lechang, Guangdong, China	22°10' N	100°51' E	724	0	10	2000
L6. Yaoshan, Guangdong, China	23°00' N	101°00' E	2000	0	8	2000
<i>C. yunnanensis</i>	25°45' N	100°00' E	1500	-5	12	1200
Y1. Mengyang, Xishuangbanna, Yunnan, China						
Y2. Simao, Yunnan, China						
Y3. Yangbi, Yunnan, China						
Y4. Kunming, Yunnan, China						

The Pearson correlation coefficient of vein number with cotyledon length, width (W1 and W2) and ratio (R1 and R2) are 0.20, 0.07, -0.05, 0.13, and 0.10, respectively. Cotyledon sizes, especially length, displays a great variation within the species, although it is usually smaller in *C. yunnanensis* than in the other two species (Fig. 5). However, cotyledon shape (R2 = W1/W2) is an ideal character for distinguishing *C. yunnanensis* (linear, mean R2 = 1.40) from *C. acuminata* var. *acuminata* (lanceolate, mean R2 = 2.08) and *C. lowreyana* (lanceolate, mean R2 = 1.98) (see Fig. 5).

Leaves

Leaf shape (R1, R2, R3) and vein number are relatively consistent within populations and even within species relative to leaf size (length and maximum width). Thus, the leaf is a key character in distinguishing species (Table 6). *C. lowreyana* typically has cordate leaves with a mean of 16 veins on each side, *C. acuminata* usually has oval leaves with fewer than 15 veins on each side, and *C. yunnanensis* has elliptic leaves with 14 veins Fig. (6). *C. lowreyana* has a

Table 3. Means and standard deviations of fruit surface, disc, and size in *Camptotheca* (Means with the same letter are not significantly different at the 0.001 level. ---- No data available).

Population	Sample Size	Surface	Disc	Length (mm)	Width (mm)	Ratio (L/W)
<i>C. acuminata</i>						
<i>var. acuminata</i>	1636	Rugose	Thick	22.54 ± 2.87	5.64 ± 0.83	4.18 ± 0.60
A1A	64	Rugose	Thick	23.23 ± 2.10	5.75 ± 0.59	4.09 ± 0.58
A1B	64	Rugose	Thick	27.30 ± 2.74	6.46 ± 0.67	4.26 ± 0.62
A2	64	Rugose	Thick	18.48 ± 1.74	4.41 ± 0.47	4.23 ± 0.54
A3	64	Rugose	Thick	18.48 ± 2.78	5.88 ± 0.73	3.19 ± 0.62
A4	64	Rugose	Thick	23.59 ± 3.14	5.70 ± 0.74	4.19 ± 0.71
A5	64	Rugose	Thick	22.67 ± 1.70	5.94 ± 0.94	3.91 ± 0.65
A6	64	Rugose	Thick	23.19 ± 2.56	5.77 ± 0.61	4.06 ± 0.60
A7	64	Rugose	Thick	24.53 ± 2.25	5.12 ± 0.64	4.86 ± 0.71
A8	64	Rugose	Thick	23.98 ± 2.03	5.47 ± 0.61	4.44 ± 0.63
A9	----	----	----	----	----	----
A10	29	Rugose	Thick	25.24 ± 2.43	5.88 ± 0.68	4.35 ± 0.72
A11	32	Rugose	Thick	22.55 ± 2.68	6.36 ± 0.80	3.57 ± 0.44
A12	46	Rugose	Thick	23.74 ± 1.58	5.68 ± 0.72	4.25 ± 0.66
A13	19	Rugose	Thick	20.58 ± 1.92	4.34 ± 0.73	4.84 ± 0.84
A14	64	Rugose	Thick	19.70 ± 2.23	5.05 ± 0.72	3.96 ± 0.61
A15	64	Rugose	Thick	22.79 ± 1.34	4.95 ± 0.61	4.68 ± 0.61
A16	5	Rugose	Thick	20.60 ± 3.63	4.10 ± 0.22	5.01 ± 0.73
A17	----	Rugose	Thick	----	----	----
A18	64	Rugose	Thick	24.09 ± 2.17	5.97 ± 0.62	4.06 ± 0.42
A19	64	Rugose	Thick	25.80 ± 2.39	6.34 ± 0.60	4.11 ± 0.62
A20	64	Rugose	Thick	23.95 ± 2.16	6.02 ± 0.72	4.03 ± 0.59
A21	64	Rugose	Thick	22.59 ± 2.14	5.46 ± 0.54	4.17 ± 0.55
B1	64	Rugose	Thick	21.03 ± 1.89	4.81 ± 0.52	4.41 ± 0.55
B2	64	Rugose	Thick	23.02 ± 2.00	5.88 ± 0.83	3.99 ± 0.63
B3a	64	Rugose	Thick	23.67 ± 2.10	6.27 ± 0.79	3.82 ± 0.49
B3b	64	Rugose	Thick	23.77 ± 1.99	6.17 ± 0.72	3.89 ± 0.47
B4	64	Rugose	Thick	23.45 ± 2.63	5.62 ± 0.53	4.20 ± 0.57
B5	64	Rugose	Thick	21.59 ± 2.24	6.05 ± 0.60	3.60 ± 0.52
B6a	64	Rugose	Thick	20.17 ± 1.17	5.43 ± 0.33	3.73 ± 0.31
B6b	64	Rugose	Thick	20.16 ± 1.50	5.41 ± 0.48	3.75 ± 0.42
B7A	64	Rugose	Thick	24.05 ± 1.58	6.35 ± 0.48	3.80 ± 0.31
B7B	64	Rugose	Thick	24.05 ± 2.02	6.13 ± 0.44	3.94 ± 0.41
B8	32	Rugose	Thick	19.13 ± 1.24	5.11 ± 0.44	3.76 ± 0.33
<i>C. acuminata</i>						
<i>var. rotundifolia</i>	----	----	----	----	----	----
<i>C. acuminata</i>	10	Rugulose	Thin	29.80 ± 2.74	5.65 ± 0.41	5.31 ± 0.75
<i>var. tenuifolia</i>	193	Smooth	Thin	29.02 ± 3.11	5.77 ± 0.70	5.08 ± 0.61
<i>C. lowreyana</i>	22	Smooth	Thin	33.82 ± 3.63	6.25 ± 0.84	5.47 ± 0.59
L1	64	Smooth	Thin	29.06 ± 2.73	5.62 ± 0.60	5.18 ± 0.34
L2	64	Smooth	Thin	29.00 ± 1.90	5.45 ± 0.43	5.34 ± 0.39
L3	22	Smooth	Thin	25.32 ± 2.15	6.32 ± 0.45	4.02 ± 0.42
L4	11	Smooth	Thin	26.14 ± 2.47	6.64 ± 0.81	3.96 ± 0.36
L5	32	Smooth	Thin	26.69 ± 1.26	6.06 ± 0.69	4.46 ± 0.61
L6	215	Smooth	Thin	22.92 ± 2.76	6.41 ± 0.78	3.61 ± 0.51
<i>C. yunnanensis</i>	64	Smooth	Thin	24.24 ± 2.02	6.26 ± 0.70	3.91 ± 0.44
Y1	23	Smooth	Thin	21.89 ± 3.97	7.35 ± 0.44	2.98 ± 0.53
Y2	64	Smooth	Thin	20.63 ± 2.03	5.93 ± 0.61	3.50 ± 0.40
Y3	64	Smooth	Thin	24.27 ± 1.68	6.73 ± 0.67	3.64 ± 0.44
Y4						



Fig. (2). Variation in mature fruits of *Camptotheca acuminata* cultivated in Nacogdoches, Texas, USA.

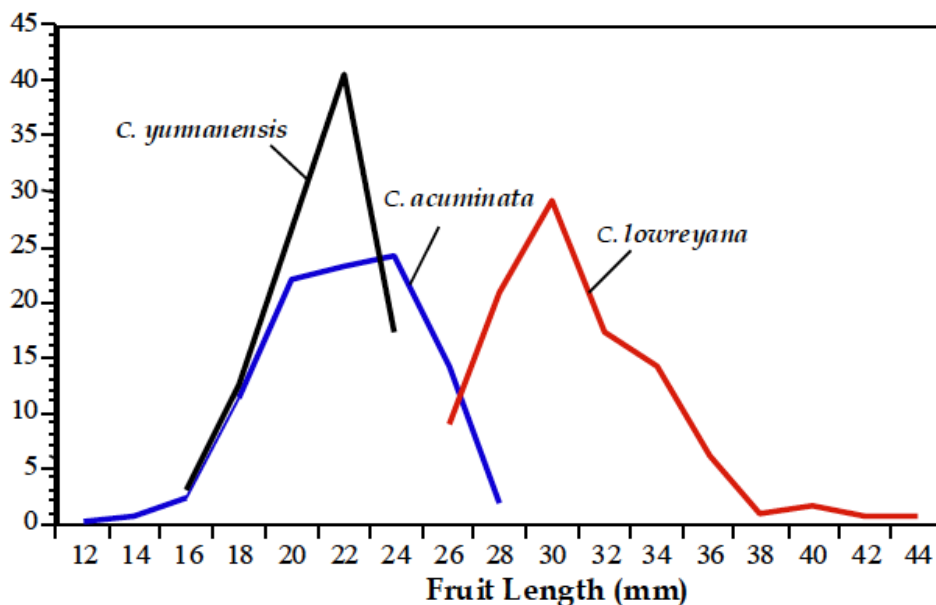


Fig. (3). Frequency distributions of fruit length of *C. acuminata* (n = 1283), *C. lowreyana* (n = 193), and *C. yunnanensis* (n = 87).

wider leaf blade base than apex ($R^2 = 1.55$) with the widest point near the blade base ($R_3 = 3.37$), while both *C. acuminata* and *C. yunnanensis* have balanced leaves ($R^2 = 1.03$ and 1.18 , respectively) with the widest point in the center of the blade or slightly below center ($R_3 = 2.04$ and 2.17 , respectively). However, leaf size, including length and maximum width, is variable with environment and is not a reliable character on which to distinguish the species. Leaf vein number in this study is not strongly related to leaf size; the Pearson correlation coefficient is only 0.54 between vein number and blade length and 0.66 between vein number and blade width.

Leaf margin is not a diagnostic character in distinguishing the taxa of *Camptotheca* because all three species tend to have serrate leaves in the juvenile phase of development (one

or two years old), and gradually become entire after two years (Table 7). This phase variation in leaf margin is not an environmental plasticity due to environmental stimuli; in fact, it is developmental plasticity or fixed phenotypic variation as Bradshaw (1965) noted [7].

DISCUSSION

Character Variations and Key Characters

Evolutionary ecologists have stressed the importance of understanding the expression and the components of phenotypic variation among and within populations [1-6] and have proposed that a hierarchical perspective may provide insights into the evolutionary history of a species [3].

Table 4. Means and standard deviations of fruit color in *Camptotheca* (Means with the same letter are not significantly different at the 0.001 level; ---- No data available; general description: R-red, B-brown, and G-gray).

Population	Sample Size	R.H.S. Color	Red Color (1 - 255)	Green Color (1 - 255)	Blue color (1 - 255)
<i>C. acuminata</i>					
var. <i>acuminata</i>	450	165A-C	139.03 ± 23.28	102.26 ± 16.88	60.27 ± 8.25
A1A	30	165B	107.97 ± 9.82 a	84.10 ± 8.18 a	53.28 ± 6.02 a
A1B	----	165B	----	----	----
A2	30	165B	105.42 ± 8.80 a	81.74 ± 7.28 b	51.38 ± 5.94 b
A3	30	165B	146.27 ± 10.10 e	103.28 ± 7.96 c	57.89 ± 5.04 c
A4	30	165B	118.39 ± 11.48 g	88.61 ± 6.18 d	55.55 ± 4.85 d
A5	30	165B	150.86 ± 11.79 de	110.21 ± 8.91 e	68.01 ± 6.55 e
A6	30	165B	156.04 ± 13.27 cd	113.94 ± 9.78 f	64.13 ± 7.10 f
A7	30	165A	115.81 ± 9.12 g	89.01 ± 6.92 g	60.67 ± 6.59 g
A8	30	165A-B	117.84 ± 9.10 g	86.83 ± 6.03 h	58.67 ± 7.02 h
A9	----	----	----	----	----
A10	----	165B	----	----	----
A11	----	165C	----	----	----
A12	----	165C	----	----	----
A13	----	165B	----	----	----
A14	----	165C	----	----	----
A15	----	165B	----	----	----
A16	----	165B	----	----	----
A17	----	165B	----	----	----
A18	----	165B	----	----	----
A19	----	165B	----	----	----
A20	----	165B	----	----	----
A21	----	165B	----	----	----
B1	30	165B	149.20 ± 10.12 e	104.40 ± 6.45 i	61.52 ± 7.04 i
B2	30	165B	156.85 ± 10.91 c	108.57 ± 6.78 j	66.36 ± 7.11 j
B3a	30	165B	163.34 ± 10.84 b	119.74 ± 7.12 k	68.69 ± 7.98 k
B3b	30	165B	165.34 ± 8.32 b	135.18 ± 6.94 l	61.76 ± 6.17 l
B4	30	165B	165.48 ± 10.71 b	119.31 ± 8.79 m	59.56 ± 7.57 m
B5	30	165B	133.92 ± 15.01 f	97.79 ± 9.44 n	62.82 ± 7.94 n
B6a	----	----	----	----	----
B6b	30	165B	132.79 ± 8.33 f	91.15 ± 6.23 o	54.02 ± 6.11 o
B7	----	165A-B	----	----	----
B8	----	165A-B	----	----	----
<i>C. acuminata</i>	----	----	----	----	----
var. <i>rotundifolia</i>	----	R-B	----	----	----
<i>C. acuminata</i>	30	164B-C	137.15 ± 11.14	112.33 ± 8.78	77.82 ± 7.94
var. <i>tenuifolia</i>	----	164B	----	----	----
<i>C. lowreyana</i>	----	164B-C	----	----	----
L1	30	164B-C	137.15 ± 11.14 f	112.33 ± 8.78 p	77.82 ± 7.94 p
L2	----	164B-C	----	----	----
L3	----	164B-C	----	----	----
L4	----	164B-C	----	----	----
L5	30	164B-C	179.61 ± 12.25 h	143.93 ± 10.12 q	96.07 ± 8.49 q
L6	----	164B	----	----	----
<i>C. yunnanensis</i>	----	164B-C	----	----	----
Y1	----	164B	----	----	----
Y2	----	----	----	----	----
Y3	----	----	----	----	----
Y4	----	----	----	----	----

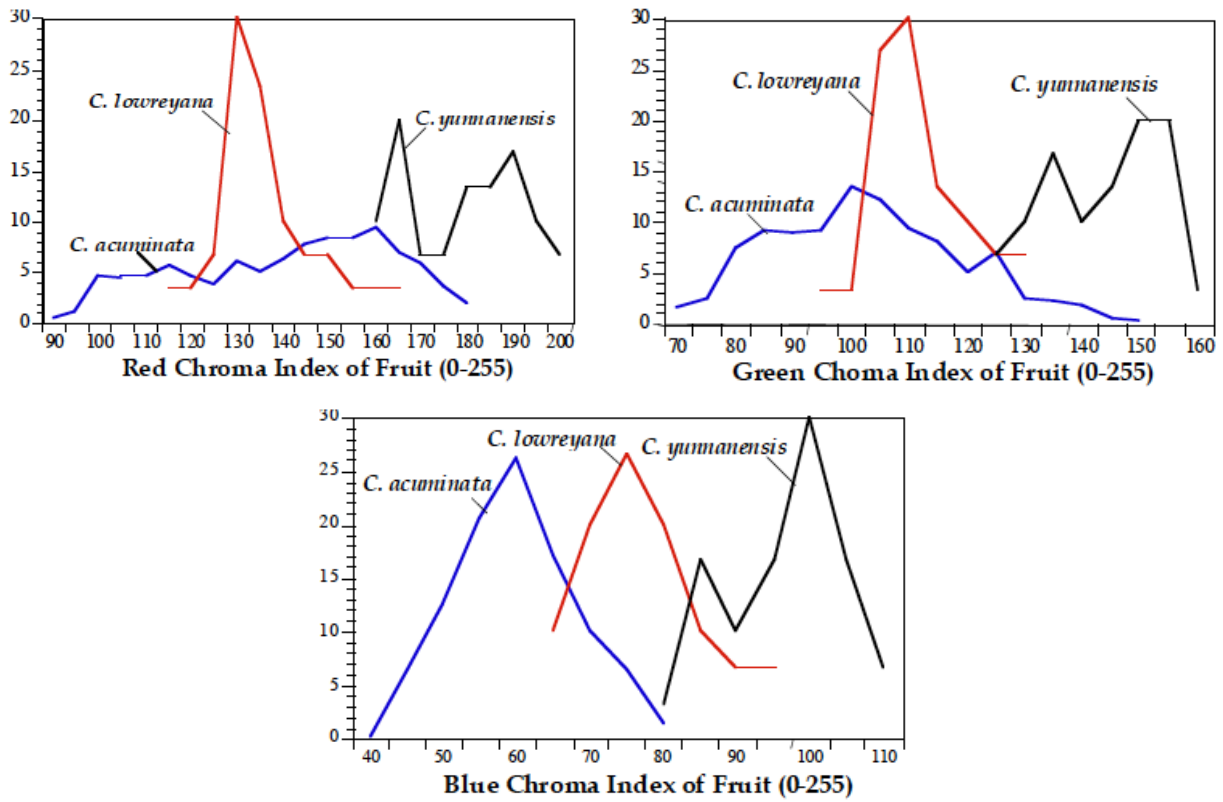


Fig (4). Frequency (% , vertical axis) distributions of fruit color of *C. acuminata* (n = 450), *C. lowreyana* (n = 30), and *C. yunnanensis* (n = 30).

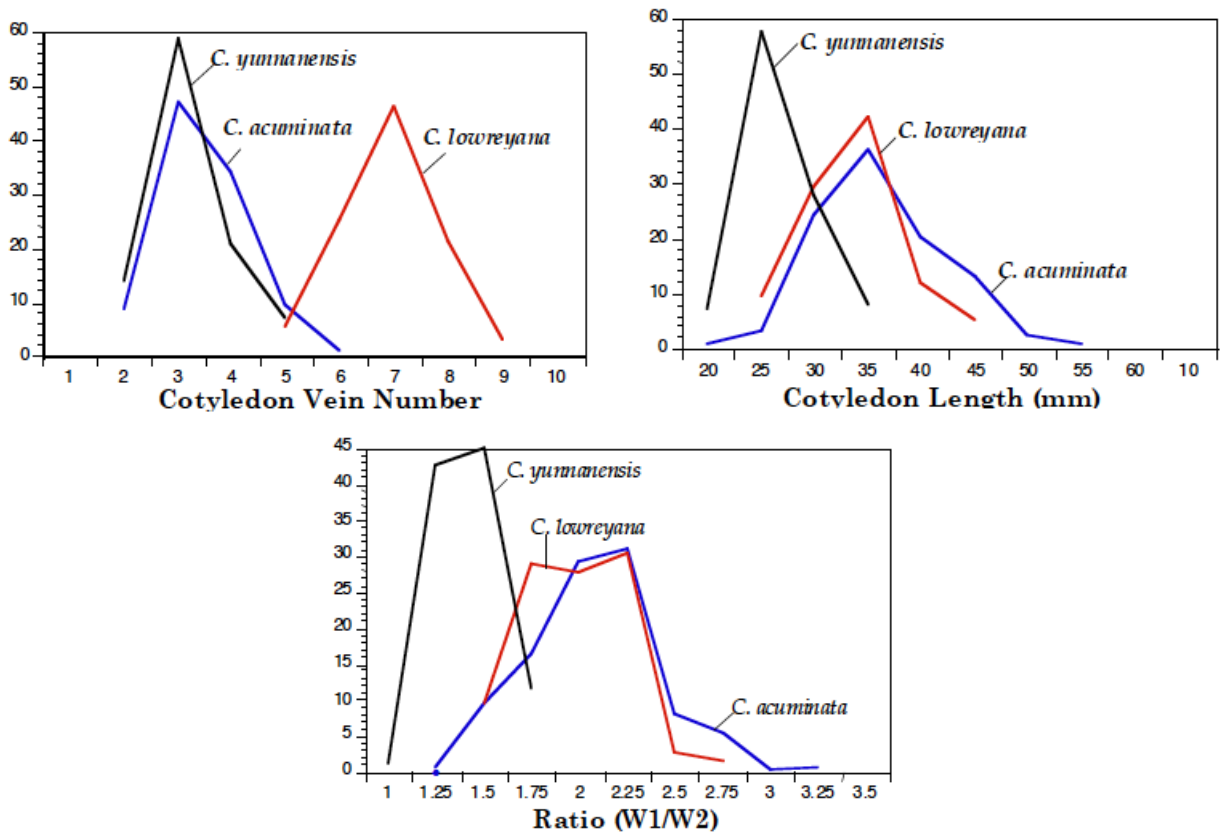


Fig (5). Frequency distributions of cotyledon vein number, length, and size ratio (W1/W2) of *C. acuminata* (n = 583), *C. lowreyana* (n = 76), and *C. yunnanensis* (n = 87).

Table 6. Means and standard deviations of leaf characters in *Camptotheca* (Means with the same letter are not significantly different at the 0.001 level. * C—cordate, E—elliptic, NE—narrowly elliptic, Ol—oval, R—round, Ot—ovate; **according to picture/publication. *** data from three parent trees. ---- no data available. W1 - blade width at 1/5 distance from the base; W2 - blade width at 1/5 distance from the apex; Lm—distance of the widest point from the base along the main vein).

Population	Leaves Surveyed	Typical Shape*	Vein Number (each side)	L (blade length) (cm)	W (blade widest point) (cm)	R1 (L/W)	R2 (W1/W2)	R3 (L/Lm)
<i>C. acuminata</i> var. <i>acuminata</i>	986	Ol	13.88 ± 2.02	15.66 ± 3.11	8.32 ± 1.51	1.89 ± 0.19	1.03 ± 0.23	2.04 ± 0.20
A1a	64	Ol	16.44 ± 1.22	18.58 ± 1.50	9.33 ± 0.82	2.00 ± 0.14	0.98 ± 0.12	2.11 ± 0.21
A1b	64	Ol	13.55 ± 1.48	14.20 ± 2.90	7.32 ± 1.43	1.95 ± 0.17	0.85 ± 0.13	1.95 ± 0.16
A2	64	Ol	14.48 ± 1.32	17.64 ± 1.53	9.22 ± 0.81	1.92 ± 0.16	1.00 ± 0.11	1.99 ± 0.19
A3	----	Ol**	----	----	----	----	----	----
A4	64	Ol	14.34 ± 2.12	16.42 ± 3.62	8.30 ± 1.54	1.97 ± 0.18	0.93 ± 0.12	2.09 ± 0.19
A5	----	Ol**	----	----	----	----	----	----
A6	64	Ol	15.95 ± 1.36	19.37 ± 2.66	9.48 ± 1.64	2.06 ± 0.13	1.12 ± 0.15	2.02 ± 0.15
A7	64	Ol	13.06 ± 1.49	14.16 ± 2.39	7.58 ± 1.15	1.87 ± 0.20	1.02 ± 0.16	1.83 ± 0.23
A8	64	Ol	15.03 ± 1.47	17.37 ± 1.83	9.68 ± 0.89	1.80 ± 0.14	0.99 ± 0.11	2.02 ± 0.11
A9	64	Ol	12.23 ± 1.42	12.06 ± 1.29	6.62 ± 0.87	1.84 ± 0.20	1.54 ± 0.20	2.31 ± 0.17
A10	2	Ol	11.50 ± 0.71	17.00 ± 2.12	8.50 ± 1.41	2.01 ± 0.08	1.76 ± 0.54	2.53 ± 0.08
A11	3	Ol	15.00 ± 2.00	17.50 ± 1.50	9.27 ± 2.19	1.94 ± 0.35	2.01 ± 0.37	2.51 ± 0.16
A12	3	Ol	15.00 ± 0.00	14.67 ± 1.76	6.83 ± 0.29	2.14 ± 0.19	1.75 ± 0.23	2.32 ± 0.07
A13	2	Ol	14.50 ± 2.12	12.25 ± 1.06	6.00 ± 0.71	2.07 ± 0.42	1.27 ± 0.22	1.97 ± 0.28
A14	4	Ol	13.25 ± 1.71	16.13 ± 1.60	7.63 ± 1.11	2.13 ± 0.22	1.77 ± 0.49	2.30 ± 0.22
A15	9	Ol	15.33 ± 1.41	15.61 ± 1.47	7.22 ± 0.57	2.16 ± 0.13	1.63 ± 0.38	2.32 ± 0.16
A16	3	Ol	13.00 ± 1.00	11.50 ± 1.00	5.93 ± 0.60	1.94 ± 0.04	1.60 ± 0.20	2.31 ± 0.21
A17	----	----	----	----	----	----	----	----
A18	64	Ol	12.70 ± 1.16	12.85 ± 2.32	7.73 ± 1.44	1.67 ± 0.11	0.98 ± 0.14	2.04 ± 0.12
A19	64	Ol	12.25 ± 1.79	14.67 ± 2.94	8.08 ± 1.55	1.83 ± 0.21	1.02 ± 0.12	2.04 ± 0.18
A20	64	Ol	12.06 ± 1.48	14.56 ± 2.52	8.74 ± 1.30	1.66 ± 1.29	0.90 ± 0.07	2.16 ± 0.14
A21	----	----	----	----	----	----	----	----
B1	----	Ol**	----	----	----	----	----	----
B2	----	Ol**	----	----	----	----	----	----
B3a	----	Ol	----	----	----	----	----	----
B3b	64	----	14.20 ± 1.18	16.11 ± 0.87	8.32 ± 0.49	1.94 ± 0.09	0.93 ± 0.08	2.02 ± 0.11
B4	----	Ol**	----	----	----	----	----	----
B5	64	Ol	15.69 ± 1.41	18.50 ± 1.75	9.62 ± 1.02	1.93 ± 0.14	0.98 ± 0.11	1.93 ± 0.14
B6a	----	Ol	----	----	----	----	----	----
B6b	64	----	13.88 ± 1.11	15.52 ± 0.93	8.27 ± 0.54	1.88 ± 0.09	0.93 ± 0.12	1.98 ± 0.11
B7a	64	----	11.95 ± 1.69	12.26 ± 2.02	6.70 ± 1.25	1.85 ± 0.19	0.94 ± 0.10	1.93 ± 0.16
B7b	----	R**	----	----	----	----	----	----
B8	----	Ol	----	----	----	----	----	----
<i>C. acuminata</i> var. <i>rotundifolia</i>	----	C/Ot	----	----	----	----	----	----
<i>C. acuminata</i> var. <i>tenuifolia</i>	5	C	10.20 ± 1.10	8.30 ± 1.48	4.70 ± 0.76	1.76 ± 0.12	1.38 ± 0.64	2.01 ± 0.22
<i>C. lowreyana</i>	174	C/Ot	15.96 ± 1.99	15.45 ± 3.09	8.29 ± 1.52	1.87 ± 0.21	1.55 ± 0.35	3.37 ± 0.63
L1	1	C/Ot	16.00	17.00	10.00	1.70	1.70	2.83
L2' ***	20	C/Ot	16.70 ± 1.34	17.85 ± 2.32	10.23 ± 1.24	1.75 ± 0.14	2.22 ± 0.36	3.93 ± 0.82
L2	83	Ot	15.13 ± 1.47	13.12 ± 2.11	7.50 ± 1.38	1.76 ± 0.11	1.42 ± 0.20	3.45 ± 0.57
L3	64	Ot	17.16 ± 1.86	17.84 ± 1.85	8.77 ± 1.01	2.04 ± 0.19	1.46 ± 0.23	3.08 ± 0.46
L4	3	----	13.33 ± 0.58	15.33 ± 1.26	7.83 ± 0.76	1.97 ± 0.23	1.90 ± 0.16	3.19 ± 0.44
L5	3	E	11.00 ± 0.00	12.33 ± 1.15	7.00 ± 0.00	1.76 ± 0.16	2.06 ± 0.24	4.04 ± 1.03
L6	----	E	----	----	----	----	----	----
<i>C. yunnanensis</i>	245	E	14.25 ± 1.57	16.17 ± 2.81	7.28 ± 1.68	2.29 ± 0.44	1.18 ± 0.20	2.17 ± 0.25
Y1	64	E	14.73 ± 1.31	15.64 ± 2.52	8.20 ± 1.34	1.91 ± 0.13	1.30 ± 0.18	2.14 ± 0.15
Y2	6	E	11.83 ± 1.47	14.83 ± 2.01	9.08 ± 1.24	1.64 ± 0.15	1.72 ± 0.30	2.35 ± 0.25
Y3	111		13.73 ± 1.48	16.71 ± 2.54	7.28 ± 1.44	2.33 ± 0.32	1.10 ± 0.13	2.23 ± 0.23
Y4	64		14.91 ± 1.44	15.96 ± 3.41	6.17 ± 1.72	2.66 ± 0.47	1.14 ± 0.20	2.10 ± 0.32

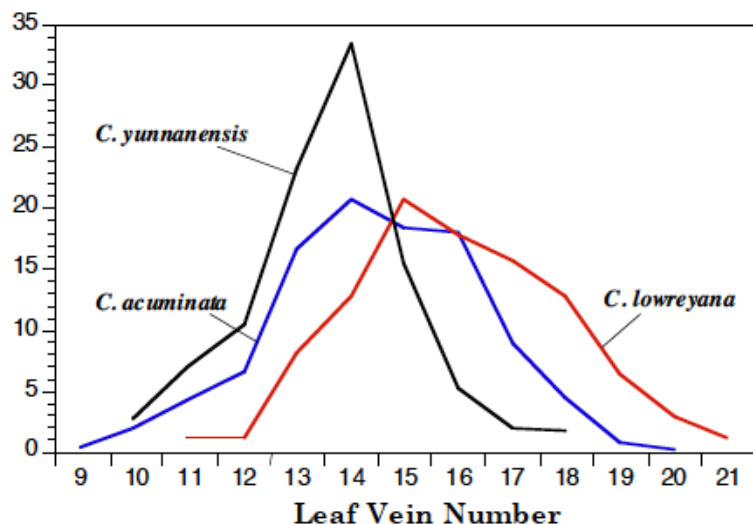


Fig (6). Frequency distributions of leaf vein number of *C. acuminata* (n = 986), *C. lowreyana* (n = 174), and *C. yunnanensis* (n = 245).

Seed size is a critical feature in the life history of a plant [8]. The size of a seed provides a measure of the size of the nutrient reserve provided for the embryo by the parent. Thus, seed size affects not only the dispersal, but also the establishment of seedlings. Seed size within a species is generally regarded as relatively constant [9, 10]. However, considerable variation in seed size is still widely observed among and within populations, within single plants, and even among years in the same plant in many species [2, 11].

The fruit of *Camptotheca* is flattened and samara-like, containing only a single seed. Thus, the fruit of *Camptotheca* is treated the same as any other seed (or fruit). The fruit surface texture and disc are consistent within the populations and species studied.

The cotyledon is a conservative organ in plant life history and is important and often overlooked in plant taxonomy and phylogeny [12, 13]. Cotyledon shape and venation pattern is usually relatively stable within populations and within species and can provide important diagnostic characters for distinguishing the species in some genera [12, 14].

The leaf is commonly used for plant identification. Seedling leaves are also important to *Camptotheca* because of their high concentration of CPTs [15].

Our phenotypic analysis showed that key diagnostic characters for the identification of taxa within *Camptotheca* include leaf shape, venation type, vein number, stoma size and frequency, outer stomatal rim, subsidiary cell number, and gland size (Table 8); cotyledon shape, venation type, and vein number; and the fruit surface texture, disc, length, and color. Other characters could not be shown to be dependable for diagnostic purposes.

Variation within and Among Populations

Camptotheca is a polygamo-monoecious genus. Neither self-fertilization nor agamospermy were observed in *C. acuminata* var. *acuminata* [16] because the stamens are shed nearly one week before the stigma of the same flower becomes receptive. Therefore, cross-pollination is the major breeding system of the species. Indeed, pollination of *Camptotheca*

is obligately entomophilous, and no fruit is produced in the absence of visiting insects (bees, butterflies, flies, and beetles) [16]. Seed set depends on the activities of pollinating insects. Species cross-pollinated by insects generally have a greater proportion of their variation within rather than among populations [17]. Major phenotypic variation was observed among populations for most measures (Table 9). The current population structure and distribution of existing trees may largely shape this variation pattern. *Camptotheca* has a fragmented geographic distribution and populations are well isolated from each other. Each natural population is very small, from 1-10 mature trees according to our observations. Selfing (crossing between male and female structures of the same plant) or related matings (crossing between closely related individual plants) are greatly increased due to the small sizes of populations.

Relatedness is also common in plantations as the plants frequently show the same seed source and may even be ramets of the same clone. Consequently, the variation in both wild populations and plantation trees now existing in China is narrow. The extent of the variation base among trees cultivated in the United States is very narrow. Most of the mature trees can be traced back to the same provenance, and all are solitary except in Chico, California, where two trees are known to exist. Therefore, selfing is the only breeding system for nearly all individual trees in the United States. Consequently, all existing plants received their limited gene pool from parents displaying obvious heterozygosity loss for generations. If the effective population size of *Camptotheca* is 20 under natural conditions, genetic variation will decrease by 2.5% per generation according to Koenig's model $1/2N$ (1988).

A majority of the observed phenotypic variation for most measures was among populations. In addition, related matings or selfing (the most severe form of related matings) is common in both natural and cultivated populations of *Camptotheca*. Therefore, it is not possible to broaden genetic diversity at the species level using only materials from a single population. To conserve genetic diversity of *Camptotheca*, it is necessary to collect seeds from all populations.

Table 7. Leaf margin characters in *Camptotheca* (* according to the original description. ----- No data available).

Population	Parent Trees (>10 years old)				One-Year-Old Seedlings				Two-Year-Old Seedlings			
	Leaves (trees) Surveyed	Serrate (%)	Slightly Serrate (%)	Entire (%)	Leaves (seedlings) Surveyed	Serrate (%)	Slightly Serrate (%)	Entire (%)	Leaves (seedlings) Surveyed	Serrate (%)	Slightly Serrate (%)	Entire (%)
<i>C. acuminata</i> var. <i>acuminata</i>	1768 (54)	0.1	2.6	97.3	9495 (251)	91.9	7.9	0.2	4800 (120)	46.9	21.9	31.2
A1a	200 (10)	1.5	5.0	93.5	800 (20)	80.5	19.5	0	800 (20)	71.5	22.5	6.0
A1b	200 (10)	1.5	5.0	93.5	64 (5)	95.3	4.7	0	-----	-----	-----	-----
A2	200 (10)	0	5.0	95.0	800 (20)	96.0	4.0	0	800 (20)	57.1	23.0	19.9
A3	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
A4	-----	-----	-----	-----	800 (20)	97.5	2.5	0	800 (20)	56.4	27.3	16.4
A5	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
A6	-----	-----	-----	-----	800 (20)	83.3	13.8	2.9	800 (20)	12.7	14.1	73.2
A7	-----	-----	-----	-----	800 (20)	95.5	4.5	0	800 (20)	17.7	22.2	60.1
A8	-----	-----	-----	-----	800 (20)	87.8	12.2	0	-----	-----	-----	-----
A9	-----	-----	-----	-----	800 (20)	95.7	4.3	0	-----	-----	-----	-----
A10	4 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A11	4 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A12	6 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A13	7 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A14	6 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A15	17 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A16	4 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A17	20 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
A18	40 (1)	0	5.0	95.0	128 (6)	93.8	6.2	0	-----	-----	-----	-----
A19	40 (1)	0	0	100.0	139 (5)	87.8	12.2	0	-----	-----	-----	-----
A20	60 (5)	0	3.3	96.7	229 (7)	91.3	8.7	0	-----	-----	-----	-----
A21	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
B1	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
B2	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
B3a	200 (1)	0	2.0	98.5	-----	-----	-----	-----	-----	-----	-----	-----
B3b	200 (1)	0	2.0	98.5	800 (20)	97.0	3.0	0	-----	-----	-----	-----
B4	200 (1)	0	1.5	98.5	800 (20)	87.0	13.0	0	-----	-----	-----	-----
B5	200 (1)	0	14.0	86.0	800 (20)	86.5	13.5	0	800 (20)	66.2	22.1	11.7
B6a	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
B6b	-----	-----	-----	-----	800 (20)	97.9	2.1	0	-----	-----	-----	-----
B7a	50 (1)	0	4.0	96.0	135 (8)	97.8	2.2	0	-----	-----	-----	-----
B7b	50 (1)	0	2.0	98.0	-----	-----	-----	-----	-----	-----	-----	-----
B8	60 (1)	0	5.0	95.0	-----	-----	-----	-----	-----	-----	-----	-----
<i>C. acuminata</i> var. <i>rotundifolia</i> *	7 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
<i>C. acuminata</i> var. <i>tenuifolia</i>	6 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
<i>C. lowreyana</i>	423 (17)	0	1.5	98.5	1600 (40)	16.1	81.9	2.0	1600 (40)	27.8	22.7	49.5
L1	3 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
L2	200 (3)	0	5.0	95.0	800 (20)	9.0	89.9	1.1	800 (20)	25.6	27.8	46.7
L3	200 (10)	0	3.8	96.2	800 (20)	26.2	70.9	2.9	800 (20)	30.0	17.5	52.5
L4	8 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
L5	8 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
L6	4 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
<i>C. yunnanensis</i>	308 (22)	0	2.1	97.9	1419 (34)	70.7	27.6	1.7	800 (20)	28.3	62.1	65.5
Y1	50 (1)	0	0	100.0	380 (7)	89.0	6.3	4.7	-----	-----	-----	-----
Y2	8 (1)	0	0	100.0	-----	-----	-----	-----	-----	-----	-----	-----
Y3	200 (10)	0	2.6	97.4	800 (20)	32.3	67.7	0	800 (20)	28.3	62.1	65.5
Y4	50 (10)	0	1.5	98.5	239 (7)	90.8	8.8	0.4	-----	-----	-----	-----

Table 8. Means and standard deviations of leaf stoma features in *Camptotheca* (Means with the same letter are not significantly different at the 0.001 level. * 120 cells were measured for each population).

Population	Cell Number	Stomatal Density (no./mm ²)	Stoma Length (L) (μm)	Stoma Width (W) (μm)	R (L/W)	Subsidiary Cell Number *	Gland Length (μm)
<i>C. acuminata</i> var. <i>acuminata</i>	600	180.39 ± 35.62	22.85 ± 2.25	15.30 ± 1.91	1.51 ± 0.22	4.86 ± 0.80	34.87 ± 3.92
A1	60	167.99 ± 31.12	24.08 ± 2.18	14.75 ± 1.35	1.64 ± 0.13	4.70 ± 0.69	37.33 ± 2.98
A2	60	125.68 ± 22.64	23.33 ± 2.70	12.50 ± 2.45	1.90 ± 0.31	4.58 ± 0.67	37.75 ± 4.10
A3	----	----	----	----	----	----	----
A4	60	187.20 ± 23.95	20.64 ± 1.50	15.42 ± 1.27	1.34 ± 0.11	4.88 ± 0.84	34.04 ± 4.29
A5	----	----	----	----	----	----	----
A6	60	173.05 ± 29.08	24.18 ± 2.18	15.05 ± 1.65	1.62 ± 0.14	4.96 ± 0.80	38.83 ± 3.43
A7	60	148.38 ± 23.42	24.23 ± 1.68	15.75 ± 1.38	1.55 ± 0.12	4.86 ± 0.81	33.18 ± 2.63
A8	60	196.85 ± 23.18	22.38 ± 1.78	15.56 ± 1.48	1.44 ± 0.12	4.83 ± 0.79	33.45 ± 3.02
A9	60	208.70 ± 21.23	21.73 ± 1.49	15.69 ± 1.41	1.39 ± 0.09	4.95 ± 0.67	35.14 ± 4.71
A10	----	----	----	----	----	----	----
A11	----	----	----	----	----	----	----
A12	----	----	----	----	----	----	----
A13	----	----	----	----	----	----	----
A14	----	----	----	----	----	----	----
A15	----	----	----	----	----	----	----
A16	----	----	----	----	----	----	----
B1	----	----	----	----	----	----	----
B2	----	----	----	----	----	----	----
B3a	----	----	----	----	----	----	----
B3b	60	207.60 ± 28.73	22.44 ± 1.53	16.50 ± 1.21	1.36 ± 0.10	4.88 ± 0.83	34.56 ± 3.30
B4	----	----	----	----	----	----	----
B5	60	189.30 ± 19.36	23.25 ± 2.28	15.17 ± 1.65	1.54 ± 0.12	5.02 ± 0.89	36.42 ± 3.64
B6a	----	----	----	----	----	----	----
B6b	60	199.15 ± 24.79	22.29 ± 2.03	16.60 ± 1.70	1.35 ± 0.10	4.92 ± 0.88	32.10 ± 2.72
<i>C. acuminata</i> var. <i>rotundifolia</i>	----	----	----	----	----	----	----
<i>C. acuminata</i> var. <i>tenuifolia</i>	----	----	----	----	----	----	----
<i>C. lowreyana</i>	120	263.20 ± 60.47	29.33 ± 3.51	20.08 ± 2.11	1.46 ± 0.13	5.23 ± 0.93	44.49 ± 4.51
L1	----	----	----	----	----	----	----
L2	60	297.09 ± 61.59	27.55 ± 2.80	19.63 ± 2.45	1.41 ± 0.11	5.27 ± 0.91	38.61 ± 4.78
L3'	60	229.31 ± 34.15	31.13 ± 3.22	20.53 ± 1.63	1.52 ± 0.13	5.20 ± 0.97	47.98 ± 3.70
L4	----	----	----	----	----	----	----
L5	----	----	----	----	----	----	----
L6	----	----	----	----	----	----	----
<i>C. yunnanensis</i>	60	221.22 ± 46.85	31.05 ± 3.60	20.23 ± 3.48	1.56 ± 0.23	5.83 ± 1.12	32.20 ± 2.70
Y1	60	221.22 ± 46.85	31.05 ± 3.60	20.23 ± 3.48	1.56 ± 0.23	5.83 ± 1.12	35.20 ± 2.70
L7	----	----	----	----	----	----	28.93 ± 2.57

Variations Among Generations

Reproduction from seed is a sexual process that results in genetically variable offspring (Raulston and Tripp 1994). Any population of seedlings demonstrates an amazing array of variability. However, some populations exhibit more phenotypic variation than others do.

Six samples of *C. acuminata* var. *acuminata* in the United States represent four selfing generations from the same provenance (first generation: B1, second: B3, third: B4 and B11, and fourth: B5 and B10). B6 and B7 are two selfing generations from the tree in Honolulu, Hawaii. Each of these eight samples and B1 share the same parental base from the Guangxi or Sichuan provenance (Steward No. 75).

Under different environmental conditions (see Table 1), the composite samples from these trees are close in pedigree and are more similar to each other than other samples in both

means and frequency distributions of leaf vein number, fruit length, fruit colors, and cotyledon vein number (see Tables 3-6). For example, B2, B3, and B4 are similar to each other, and relatively similar to B1 and B6, but less similar to others (e.g., A2 and A6) in fruit length (23.03-23.67 vs. 21.03 of B1 and 20.17 of B6 vs. 23.19 of A6 and 18.48 of A2 and A3), red chroma (156.85-165.48 vs. 149.20 of B1 and 133.92 of B6 vs. 115.81 of A6 and 105.42 of A2) and green chroma (108.57-119.74 vs. 104.40 of B1 and 97.79 of B6 vs. 89.01 of A6 and 81.74 of A2). These data indicate that fruit length and chroma is controlled genetically from generation to generation and is only marginally affected, if at all, by local physical environment.

A major factor contributing to variation in fruit length and chroma is the efficiency of pollination. The reason that no significant variation in fruit length and chroma was

Table 9. Tukey's Studentized Range (HSD) pairwise absolute mean differences for each measure between taxa (W1-blade width of cotyledon at 5 mm from the base; W2-blade width of cotyledon at 5 mm from the apex. Level of significance: ** at $P < 0.01$; *** at $P < 0.001$; ns: not significant at $P < 0.05$. ---- No data available. The difference between means listed in the table is measured as column – row).

Taxon	Measure	<i>C. acuminata</i> var. <i>acuminata</i>	<i>C. lowreyana</i>
<i>C. lowreyana</i>	Leaf vein number	3.51***	
	Leaf blade length (LL)	1.99***	
	Leaf blade width (LW)	1.25***	
	Leaf ratio (LW/LL)	0.01***	
	Fruit length (FL)	8.23***	
	Fruit width (FW)	0.14***	
	Fruit ratio (FW/FL)	0.07***	
	Fruit red color	ns	
	Fruit green color	10.07***	
	Fruit blue color	17.55***	
	Cotyledon vein number	3.43***	
	Cotyledon length (CL)	-1.98***	
	Cotyledon width (W1)	-0.49***	
	Cotyledon width (W2)	ns	
	Cotyledon ratio R1 (CL/W1)	ns	
Cotyledon ratio R2 (W1/W2)	ns		
<i>C. yunnanensis</i>	Leaf vein number	ns	3.43***
	Leaf blade length (LL)	ns	2.17***
	Leaf blade width (LW)	1.98***	3.23***
	Leaf ratio (LW/LL)	0.12***	0.13***
	Fruit length (FL)	1.50***	9.73***
	Fruit width (FW)	0.33***	ns
	Fruit ratio (FW/FL)	0.03***	0.10***
	Fruit red color	40.57***	42.45***
	Fruit green color	41.67***	31.60***
	Fruit blue color	35.80***	18.25***
	Cotyledon vein number	-0.27***	-3.70***
	Cotyledon length (CL)	-9.35***	-7.36***
	Cotyledon width (W1)	-2.58***	-2.09***
	Cotyledon width (W2)	ns	ns
	Cotyledon ratio R1 (W1/CL)	0.43***	0.41***
Cotyledon ratio R2 (W2/W1)	-0.67***	-0.57***	

observed among Chico (B2), San Antonio (B3), and Kingwood (B4) trees is that the pollination pool of *C. acuminata* var. *acuminata* was fixed in these three locations. In both San Antonio and Kingwood, there was only a single mature *C. acuminata* var. *acuminata* tree in 1993 and 1994 when fruits were collected. Chico had only two mature trees during this period. Thus, it is expected that selfing or related matings restricted the genetic variation within those observed plants.

Variation Among Populations in the Same Habitat and Location

Both A1 and L3 samples came from the same habitat and the same general location. The distance between the closest trees within these two populations is about 200 m. L3 had a mean fruit size of 29.00 cm in length, 5.45 mm in width, and 5.34 in ratio, while A1A had a mean fruit size of 23.23 cm in length, 5.75 mm in width, and 4.09 in ratio (see Table 3). They also show significant differences in fruit color and tex-

ture: fruits of L3 have a smooth surface and gray-brown color (chroma indices of 137.15 in red, 112.33 in green, and 77.82 in blue) while those of A1 have a rugose surface and are red-brown in color (107.97 in red, 84.10 in green, and 53.28 in blue) (see Table 5). In addition, samples from each of the two populations differ in cotyledon features, especially venation type and vein number (see Table 5).

Camptotheca acuminata var. *acuminata* is commonly planted in the "four-sides" (waterside, hillside, roadside, and homeside); however, seed sources are widely scattered and dependent on where mature surviving natural trees happen to be found. Provenance tests for optimum ecotype selections have never been done and seed is not commercially available.

It is generally expected that outcrossing plants like *Camptotheca* have small variations among populations and large within populations. However, our study found that a majority of the observed phenotypic variation was among populations for most morphometric characters (Table 10).

Table 10. Sources of morphological variations for the genus *Camptotheca* (W1-blade width of cotyledon at 5 mm from the base; W2-blade width of cotyledon at 5 mm from the apex; VWP-variation within populations; VAP-variation among population; VAS-variation among species).

Character	VWP (% of total)	VAP (% of total)	VAS (% of VAP)
Leaf vein number	5.46	94.54	30.35
Leaf blade length (LL)	31.59	68.41	6.65
Leaf blade width (LW)	17.07	82.93	21.91
Leaf ratio (LW/LL)	20.44	79.56	35.57
Fruit length (FL)	25.91	74.09	73.30
Fruit width (FW)	63.23	36.77	3.31
Fruit ratio (FW/FL)	45.72	54.28	55.31
Fruit red color	23.42	76.58	25.43
Fruit green color	22.14	77.86	51.08
Fruit blue color	28.73	71.27	83.13
Cotyledon vein number	25.32	74.68	99.00
Cotyledon length (CL)	12.93	87.07	27.45
Cotyledon width (W1)	45.80	54.20	97.81
Cotyledon width (W2)	86.73	13.27	90.73
Cotyledon R1 (W1/CL)	57.94	42.06	87.27
Cotyledon R2 (W2/W1)	39.00	61.00	91.37

This is probably due largely to selfing and related matings common to both natural and cultivated populations, resulting in the current population structure that now displays a highly fragmented distribution pattern. This makes it difficult to extend patterns from a single population to the species level in the genus. For genetic resource conservation, it is necessary to collect data from as many populations with as few individuals within the population as possible in order to ensure greater genetic diversity. This is true even if the population consists of only one tree. This means that conservation of all remaining wild trees is imperative. A recommendation to that effect has been submitted to the Chinese government.

As a result of taxonomic treatment based on key characters, the taxon from Yunnan is treated as *C. yunnanensis*. The newly described species, *C. lowreyana*, produces large fruit and high CPT yield. This makes *C. lowreyana* an important candidate for plantation development because drug extract levels normally are greatest in the fruit (Li and Adair 1994). *C. yunnanensis* has a long growing period; this is a characteristic which may make *C. yunnanensis* an important selection for biomass production. The plantation possibilities of both taxa will be explored in future studies.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGEMENTS

Declared none.

PATIENT'S CONSENT

Declared none.

REFERENCES

- [1] Primack, R. B.; Kang, H. Measuring fitness and natural selection in wild plant populations. *Ann. Rev. Ecol. Syst.*, **1989**, *20*, 367-396.
- [2] Wolf, L. L.; Hainsworth, F. R.; Mercier, T.; Benjamin, B. Seed size variation and pollinator uncertainty in *Ipomopsis aggregata* (Polemoniaceae). *J. Ecol.*, **1986**, *74*, 361-371.
- [3] Delesalle, V. A.; Mazer, S. J. The structure of phenotypic variation in gender and floral traits within and among populations of *Spergularia marina*. *Am. J. Bot.*, **1995**, *82*, 798-810.
- [4] Montagnes, R. J. S.; Vitt, D. H. Patterns of phenotypic variation in *Meesia triquetra* (Bryopsida: Meesiaceae) over an Arctic-Boreal gradient. *Syst. Bot.*, **1991**, *16*, 726-735.
- [5] Mitchell-Olds, T. Quantitative genetics of survival and growth in *Impatiens capensis*. *Evolution*, **1986**, *40*, 107-116.
- [6] Venable, D. L. Using intraspecific variation to study the ecological significance and evolution of plant life-histories. In: *Perspectives on Plant Population Ecology*; Dirzo, R.; Sarukhan, J., Eds; Sinauer: Sunderland, Massachusetts, **1981**; pp. 166-187.
- [7] Bradshaw, A. D. Evolutionary significance of phenotypic plasticity in plants. *Adv. Genet.*, **1965**, *13*, 115-155.
- [8] Leshman, M. R.; Westoby, M. Hypotheses on seed size: tests using the semiarid flora of western New South Wales, Australia. *Am. Nat.*, **1994**, *143*, 890-906.

- [9] Stebbins, G. L. Adaptive radiation of reproductive characteristics in angiosperms. II. seeds and seedlings. *Ann. Rev. Ecol. Syst.*, **1971**, *2*, 237-260.
- [10] Harper, J. L. *Population biology of plant*. Academic Press: London, **1994**.
- [11] Sakai, S.; Sakai, A. Flower size-dependent variation in seed size: theory and a test. *Am. Nat.*, **1985**, *145*, 918-934.
- [12] Zhang, R. H.; Liu, H. E.; Wang, Z. T. *Seedling morphology of important trees in China*. Science Press: Beijing, **1993**.
- [13] Huang, P.H. Evolution trends and relationships of some traits from the view of seedling morphology. *J. N.E. For. Univ.*, **1980**, 36-56.
- [14] Li, S. Y.; Ning, Z. Morphology, taxonomy and distribution of the genus *Rhamnus L.* (Rhamnaceae) in the northeastern China. *Bull. Bot. Res.*, **1988**, *8*(2), 71-114.
- [15] Li, S. Y.; Yi, Y. J.; Wang, Y. J.; Zhang, Z. Z.; Beasley, R. S. Camptothecin accumulation and variation in *Camptotheca Decaisne*. *Planta Med.*, **2002**, *68*(11), 1010-1016.
- [16] Chen, L. J.; wang, F. H.; Wu, Y. R. *The pollination biology of Camptotheca acuminata Decne. (Nyssaceae)*. *Cathaya*, **1991**, *3*, 45-52.
- [17] Hamrick, J. L.; Godt, M. J. Allozyme diversity in plant species. In *Plant population genetics, breeding, and genetic resources*, Brown, A. D.; Clegg, M. T.; Kahler, A. L.; Weir, B. S., Eds. Sinauer Sunderland, Massachusetts, **1990**, pp. 43-63.

Received: December 30, 2013

Revised: September 16, 2014

Accepted: September 24, 2014

© Shiyou Li; Licensee *Bentham Open*.

This is an open access article licensed under the terms of the Creative Commons Attribution Non-Commercial License (<http://creativecommons.org/licenses/by-nc/3.0/>) which permits unrestricted, non-commercial use, distribution and reproduction in any medium, provided the work is properly cited.