

Principal component and morphological diversity analysis of Job's-tears (*Coix lacryma-jobi* L.)

Gang Shen^{1,2}, Teerayoot Girdthai^{1*}, Zuo Y. Liu², Yu H. Fu², Qing Y. Meng³, and Fan Z. Liu²

¹Suranaree University of Technology, School of Crop Production Technology, Institute of Agricultural Technology, Nakhon Ratchasima 30000, Thailand. *Corresponding author (teerayoot@sut.ac.th).

²Guizhou Academy of Agricultural Sciences, Institute of Subtropical Crops Research, Guizhou 550006, China.

³Guizhou Normal University, School of Life Sciences, Guizhou 550001, China.

Received: 1 September 2018; Accepted: 22 November 2018; doi:10.4067/S0718-58392019000100131

ABSTRACT

The diversity analysis of Job's-tears (*Coix lacryma-jobi* L.), an ideal healthy food crop, is a prerequisite in breeding programs and germplasm utilization. The aim of this study was to characterize the phenotypic traits of 94 Job's-tears accessions (40 cultivated and 54 wild) collected from different geographic areas in China. Principal component (PC) and genetic diversity analyses were conducted on 12 morphological characters: stem node number, panicle branch number, primer branch nodes, panicles per plant, grain number per plant, plant height, 100-seed weight, total bract surface characteristics, total bract texture, total bract shape, total bract color, and pericarp color. The results showed a high variation among the studied materials. The relationship among traits indicated that some traits could be used for indirect selection to evaluate accessions. Based on PC analysis, the first seven PCs in the experiment can summarize the vast majority of the information about the agronomic traits of the 94 Job's-tears accessions. The accumulative contribution rate accounted for 87.31% of the total variation. Cluster analysis grouped all the accessions into seven clusters, and this revealed that genetic variation was based on variety types, geographic distribution, and morphological characteristics.

Key words: Genetic diversity, Job's-tears, morphological traits, principle component analysis.

INTRODUCTION

Job's-tears (*Coix lacryma-jobi* L.) is an economically important annual crop mainly planted in Asian countries (Diao, 2017). It belongs to the *Coix* genus, *Andropogoneae* tribe within the family *Poaceae*, and is a tall grain-bearing tropical plant (Taylor, 1953; Zhou et al., 2010). Job's-tears is also known as coix seed, tear grass, hato mugi, and adlay. The demand for Job's-tears is rapidly increasing due to its medicinal functions as an ideal healthy food. It has been introduced and grown in almost all the tropical and subtropical areas in the world (Lim, 2012).

Inappropriate cultivation techniques coupled with the long-term use of landrace varieties have caused low seed quality and production. In addition, a few research studies about Job's-tears have been conducted and led to a lack of new varieties with high yield and quality (Huang et al., 2009). Genetic resources are the most important part of a plant breeding program, and diversity analysis is a prerequisite for their more efficient management and utilization. Accurate identification of the genotype is very important during all the plant breeding steps, from initial parent selection to the final utilization of cultivars in production schemes (UPOV, 1991). Moreover, diversity analysis is an essential process to clearly identify the genetic relatedness of the available genetic resources. Modern objectives in plant breeding might be achieved by trait evaluation in genetic resources. Molecular marker methods for genotype description have proved useful, but these methods are expensive and need marker-linked trait information. Morphological characters must be recorded to select parents and their progenies, and they must always be used to describe and classify germplasm. Principle component

and cluster analysis are also useful tools to screen accessions (Karimi et al., 2009). To better conserve and utilize genetic resources, appropriate morphological variables should be carefully chosen (Giraldo et al., 2010). There are few studies that have reported the morphological analysis of Job's-tears accessions. Ten major quantitative traits of 12 Job's-tears germplasms from different regions were analyzed by path and principal component analysis (Li et al., 2010). The main components and cluster analysis of 25 Job's-tears accessions were also conducted in 2013 (Wang et al., 2013). So far, studies on the genetic diversity of Job's-tears have been conducted on accessions mainly originating from the Guangxi, Guizhou, and Yunnan Provinces in southern China (Li et al., 2001; Ma et al., 2010; Wang et al., 2015); however, no systematic study about the morphological diversity of Job's-tears has been reported. Hence, the objectives were to analyze the genetic diversity of 94 Job's-tears accessions from different regions in China and clarify the genetic relationships among the accessions to provide valuable information for the selection of excellent accessions as parents and utilize and conserve the Job's-tears genetic resource.

MATERIALS AND METHODS

Plant material

The 94 Job's-tears (*Coix lacryma-jobi* L.) accessions evaluated in this study are shown in Table 1. Accessions consisted of 40 cultivated and 54 wild accessions that were collected from different geographic regions in seven Chinese provinces (36 Guizhou, 5 Guangxi, 7 Sichuan, 7 Chongqing, 7 Hunan, 19 Yunnan, 4 Hubei, and 9 Qianxinan Institute of Agricultural Sciences accessions). All Job's-tears accessions were collected and multiplied during 2013-2014.

Thirty seeds from each Job's-tears accession were planted in seedling cups. Seedlings with 3-4 leaves were transplanted to the field at the Germplasm Resources Garden of Guizhou Academy of Agricultural Sciences (26°57' N, 106°71' E; 1074.3 m a.s.l.), Guizhou province, China, during April 2015. A completely randomized design (CRD) with three replicates was used. Each replicate consisted of 10 plants with 1 plant per hill. The row and plant spacing were 60 and 40 cm, respectively.

Morphological traits

Twelve different quantitative and qualitative traits were evaluated at harvest time. Five plants (normal growth, uniform performance, disease- and insect pest-free) from each accession were randomly selected for scoring. Seven quantitative characters, including stem node number (SNN), panicle branch number (PB), primer branch nodes (PBN), panicles per plant (PP), and grain number per plant (GNP) were evaluated. Plant height (PH) was measured from ground to tip of the main spike at physiological maturity. The 100-seed weight (SW) was measured with a scale sensitive to 0.1 g. Five qualitative characters, including total bract surface characteristics (TBSC), total bract texture (TBT), total bract shape (TBS), total bract color (TBC), and pericarp color (PC) were also recorded.

Data analysis

For the seven quantitative traits, descriptive statistics were computed for each accession with the Statistical Package for Social Science (SPSS 20.0; IBM, Armonk, New York, USA). The mean, maximum, minimum, standard deviation (SD), and coefficient of variation (CV) were calculated for each of the seven quantitative traits (Table 2). The five qualitative traits were scored according to Li et al. (2015): TBSC: smooth = 1, longitudinal convex stripes = 2; TBT: enamel = 1, crustaceous = 2; TBS: circular = 1, oval = 2, length circle = 3; TBC: white = 1, grayish-white = 2, blue-gray without dark stripes = 3, blue-gray with dark stripes = 4, yellow-white = 5, tawny without dark stripes = 6, tawny with dark stripes = 7, brown = 8, dark brown = 9; and PC: light yellow = 1, yellow = 2, brown = 3. Principal component analysis (PCA) was carried out using the original numerical data of quantitative characters and the assigned qualitative trait data. The Euclidean distance between Job's-tears accessions, based on morphological characteristics, was calculated by the between-group linkage method in SPSS 20.0.

Table 1. The codes and collection location of Job's-tears accessions used in this study.

| Code | Collection location | Type | Code | Collection location | Type |
|------|------------------------|------------|------|----------------------|------------|
| Y1 | Qianjiang Chongqing | Wild | Y48 | Cili Hunan | Wild |
| Y2 | Qianjiang Chongqing | Wild | Y49 | Cili Hunan | Wild |
| Y3 | Qianjiang Chongqing | Wild | Y50 | Cili Hunan | Wild |
| Y4 | Banan Chongqing | Wild | Y51 | Enshi Hubei | Wild |
| Y5 | Nanchuan Chongqing | Wild | Y52 | Shizong Hubei | Cultivated |
| Y6 | Nanchuan Chongqing | Cultivated | Y53 | Xianfeng Hubei | Wild |
| Y7 | Wansheng Chongqing | Wild | Y54 | Xianfeng Hubei | Wild |
| Y8 | Changshun Guizhou | Wild | Y55 | Longling Guangxi | Cultivated |
| Y9 | Changshun Guizhou | Wild | Y56 | Longling Guangxi | Wild |
| Y10 | Changshun Guizhou | Wild | Y57 | Xilin Guangxi | Cultivated |
| Y11 | Zunyi Guizhou | Wild | Y58 | Baise Guangxi | Cultivated |
| Y12 | Huaxi Guizhou | Wild | Y59 | Tianlin Guangxi | Cultivated |
| Y13 | Huaxi Guizhou | Wild | Y60 | Qianxinan Institute1 | Cultivated |
| Y14 | Huaxi Guizhou | Wild | Y61 | Qianxinan Institute2 | Cultivated |
| Y15 | Huaxi Guizhou | Wild | Y62 | Qianxinan Institute3 | Cultivated |
| Y16 | Suiyang Guizhou | Wild | Y63 | Qianxinan Institute4 | Cultivated |
| Y17 | Ziyun Guizhou | Cultivated | Y64 | Qianxinan Institute5 | Cultivated |
| Y18 | Yuping Guizhou | Wild | Y65 | Qianxinan Institute6 | Cultivated |
| Y19 | Meitan Guizhou | Wild | Y66 | Qianxinan Institute7 | Cultivated |
| Y20 | Wangmo Guizhou | Wild | Y67 | Qianxinan Institute8 | Cultivated |
| Y21 | Wangmo Guizhou | Wild | Y68 | Qianxinan Institute9 | Cultivated |
| Y22 | Wangmo Guizhou | Wild | Y69 | Zizhong Sichuan | Wild |
| Y23 | Wangmo Guizhou | Wild | Y70 | Zizhong Sichuan | Wild |
| Y24 | Wangmo Guizhou | Wild | Y71 | Jianyang Sichuan | Wild |
| Y25 | Qianlong Guizhou | Cultivated | Y72 | Jianyang Sichuan | Wild |
| Y26 | Puding Guizhou | Cultivated | Y73 | Jianyang Sichuan | Wild |
| Y27 | Puding Guizhou | Wild | Y74 | Chengdu Sichuan | Wild |
| Y28 | Puding Guizhou | Wild | Y75 | Tianjin Sichuan | Wild |
| Y29 | Sinan Guizhou | Cultivated | Y76 | Maguan Yunnan | Cultivated |
| Y30 | Yinjiang Guizhou | Cultivated | Y77 | Maguan Yunnan | Cultivated |
| Y31 | Yinjiang Guizhou | Wild | Y78 | Qiubei Yunnan | Cultivated |
| Y32 | Wuchuan Guizhou | Wild | Y79 | Xichou Yunnan | Wild |
| Y33 | Wuchuan Guizhou | Wild | Y80 | Luoping Yunnan | Cultivated |
| Y34 | Wuchuan Guizhou | Wild | Y81 | Luoping Yunnan | Cultivated |
| Y35 | Ceheng Guizhou | Wild | Y82 | Luoping Yunnan | Cultivated |
| Y36 | Ceheng Guizhou | Cultivated | Y83 | Luoping Yunnan | Cultivated |
| Y37 | Ceheng Guizhou | Cultivated | Y84 | Quejing Yunnan | Wild |
| Y38 | Xingren Guizhou | Cultivated | Y85 | Wenshan Yunnan | Wild |
| Y39 | Xingren Guizhou | Cultivated | Y86 | Wenshan Yunnan | Cultivated |
| Y40 | Xingrenxiashan Guizhou | Cultivated | Y87 | Shizong Yunnan | Wild |
| Y41 | Xingrenxiashan Guizhou | Cultivated | Y88 | Shizong Yunnan | Cultivated |
| Y42 | Xingyi Guizhou | Cultivated | Y89 | Fuyuang Yunnan | Cultivated |
| Y43 | Guanling Guizhou | Cultivated | Y90 | Fuyuang Yunnan | Cultivated |
| Y44 | Yongshun Hunan | Wild | Y91 | Fuyuang Yunnan | Cultivated |
| Y45 | Sanzhi Hunan | Wild | Y92 | Kunming Yunnan | Wild |
| Y46 | Sanzhi Hunan | Wild | Y93 | Dali Yunnan | Wild |
| Y47 | Sanzhi Hunan | Wild | Y94 | Lijiang Yunnan | Wild |

RESULTS

Quantitative trait analysis

Accessions Y35 (263.4 cm), Y41 (263.3 cm), Y63 (239.4 cm), and Y77 (239.8 cm) had the highest PH values, while Y16 (146.0 cm), Y30 (148.7 cm), and Y84 (59.6 cm) had the lowest PH (Table 2). The highest SNN values were observed in Y3 (13.0), Y23 (12.8), and Y41 (13.4), whereas the lowest SNN values were in Y16 (7.7) and Y39 (7.8). The highest PB was observed in Y25 (26.6), followed by Y36 (16.4) and Y74 (15.7), while the lowest PB was recorded in Y10 (5.0) and Y16 (5.0). The maximum PBN were found in Y35 (8.5), Y63 (8.5), and Y71 (8.7), whereas the minimum value was obtained in Y30 (2.7) and Y52 (1.7). The highest PP values were recorded in Y77 (120.4) and Y88 (117.0), while the lowest PP were in Y7 (19.9) and Y46 (19.9). The maximum GNP was observed in Y92 (316.7), followed by Y68 (302.8), Y3 (279.4), Y40 (273.5), and Y72 (266.9), while the lowest GNP values were in Y44 (51.4) and Y46 (45.0). The SW of all

Table 2. Mean of seven quantitative morphological traits of 94 Job's-tears accessions.

| Accession | PH (cm) | SNN | PB | PBN | PP | GNP | SW (g) | Accession | PH (cm) | SNN | PB | PBN | PP | GNP | SW (g) |
|-----------|---------|------|------|-----|-------|-------|--------|-----------|---------|------|------|-----|-------|-------|--------|
| Y1 | 195.2 | 12.2 | 7.6 | 5.6 | 27.8 | 68.6 | 13.2 | Y48 | 175.4 | 11.2 | 8.0 | 4.2 | 50.6 | 149.5 | 10.2 |
| Y2 | 248.4 | 11.0 | 5.4 | 5.0 | 36.7 | 111.0 | 7.6 | Y49 | 204.8 | 11.4 | 7.7 | 4.0 | 61.0 | 68.0 | 6.8 |
| Y3 | 231.5 | 13.0 | 7.5 | 6.4 | 65.0 | 279.4 | 7.8 | Y50 | 183.6 | 11.8 | 8.6 | 4.0 | 38.0 | 152.6 | 10.3 |
| Y4 | 184.8 | 10.8 | 8.6 | 3.2 | 30.2 | 78.0 | 9.9 | Y51 | 233.1 | 9.4 | 10.1 | 5.3 | 51.8 | 169.7 | 11.4 |
| Y5 | 168.3 | 10.0 | 7.0 | 3.7 | 31.0 | 124.0 | 19.7 | Y52 | 166.0 | 8.3 | 6.7 | 1.7 | 26.3 | 260.4 | 11.5 |
| Y6 | 194.0 | 8.4 | 7.5 | 2.7 | 76.2 | 128.7 | 9.5 | Y53 | 183.4 | 10.4 | 7.4 | 4.0 | 62.3 | 121.5 | 12.3 |
| Y7 | 195.0 | 12.0 | 7.7 | 4.7 | 19.9 | 266.0 | 8.1 | Y54 | 231.7 | 11.7 | 7.0 | 5.0 | 45.9 | 98.0 | 7.9 |
| Y8 | 177.4 | 9.0 | 7.4 | 4.0 | 57.6 | 159.4 | 10.2 | Y55 | 168.3 | 11.0 | 14.4 | 5.6 | 29.0 | 68.4 | 10.2 |
| Y9 | 209.0 | 11.4 | 7.0 | 6.0 | 81.3 | 124.5 | 9.0 | Y56 | 214.2 | 9.4 | 11.9 | 4.8 | 36.7 | 89.8 | 12.5 |
| Y10 | 150.0 | 10.0 | 5.0 | 8.0 | 100.4 | 76.0 | 13.6 | Y57 | 204.9 | 9.1 | 9.9 | 5.8 | 38.1 | 98.2 | 11.8 |
| Y11 | 155.4 | 9.7 | 6.4 | 4.4 | 35.0 | 240.0 | 9.1 | Y58 | 219.1 | 10.0 | 13.8 | 4.7 | 73.1 | 201.8 | 12.5 |
| Y12 | 213.7 | 10.4 | 6.8 | 3.7 | 38.1 | 92.7 | 9.9 | Y59 | 158.0 | 10.2 | 12.8 | 4.9 | 27.9 | 59.2 | 15.7 |
| Y13 | 172.0 | 9.7 | 6.0 | 4.7 | 54.0 | 70.7 | 8.0 | Y60 | 213.3 | 10.6 | 12.0 | 5.8 | 59.2 | 90.6 | 16.1 |
| Y14 | 191.7 | 9.0 | 6.7 | 3.0 | 75.0 | 109.0 | 11.9 | Y61 | 187.7 | 8.3 | 14.1 | 3.1 | 105.1 | 254.3 | 8.1 |
| Y15 | 196.1 | 9.3 | 14.0 | 4.9 | 63.4 | 119.2 | 15.8 | Y62 | 221.1 | 9.4 | 13.6 | 5.8 | 53.9 | 133.9 | 8.3 |
| Y16 | 146.0 | 7.7 | 5.0 | 5.7 | 33.7 | 71.0 | 9.9 | Y63 | 239.4 | 12.6 | 5.5 | 8.5 | 46.3 | 196.0 | 7.4 |
| Y17 | 196.2 | 12.4 | 8.8 | 4.6 | 111.4 | 136.0 | 9.5 | Y64 | 225.6 | 9.9 | 13.6 | 5.1 | 57.4 | 162.4 | 12.6 |
| Y18 | 248.7 | 12.0 | 6.7 | 6.7 | 48.8 | 205.7 | 7.5 | Y65 | 199.1 | 9.1 | 11.8 | 4.6 | 46.3 | 126.1 | 12.0 |
| Y19 | 208.7 | 9.7 | 6.4 | 3.7 | 28.1 | 89.7 | 9.8 | Y66 | 201.6 | 8.4 | 9.2 | 5.2 | 36.2 | 101.7 | 11.6 |
| Y20 | 195.1 | 8.9 | 11.4 | 4.1 | 41.1 | 122.4 | 6.9 | Y67 | 209.9 | 9.0 | 13.9 | 4.3 | 65.6 | 195.4 | 8.1 |
| Y21 | 189.4 | 11.8 | 7.2 | 5.6 | 50.2 | 98.2 | 10.8 | Y68 | 201.9 | 8.8 | 15.2 | 3.8 | 105.8 | 302.8 | 10.3 |
| Y22 | 229.8 | 11.4 | 7.0 | 5.4 | 96.6 | 182.5 | 11.5 | Y69 | 207.7 | 9.0 | 12.8 | 4.6 | 42.2 | 192.3 | 8.5 |
| Y23 | 188.8 | 12.8 | 8.6 | 5.2 | 78.8 | 164.0 | 10.9 | Y70 | 203.4 | 10.2 | 12.2 | 6.7 | 34.2 | 107.6 | 9.8 |
| Y24 | 173.2 | 10.6 | 13.2 | 5.1 | 30.3 | 73.1 | 12.7 | Y71 | 226.4 | 11.2 | 9.3 | 8.7 | 25.6 | 91.0 | 9.2 |
| Y25 | 193.0 | 11.4 | 25.6 | 3.8 | 84.2 | 176.5 | 12.6 | Y72 | 212.1 | 9.6 | 15.7 | 3.2 | 90.0 | 266.9 | 8.5 |
| Y26 | 181.6 | 10.4 | 8.4 | 3.0 | 58.6 | 148.8 | 11.8 | Y73 | 194.8 | 9.1 | 13.6 | 4.6 | 72.1 | 216.1 | 10.0 |
| Y27 | 227.8 | 10.4 | 6.7 | 5.4 | 36.2 | 117.0 | 7.2 | Y74 | 237.4 | 10.8 | 15.7 | 5.3 | 62.3 | 211.9 | 8.7 |
| Y28 | 214.7 | 10.7 | 6.7 | 5.0 | 88.4 | 75.8 | 17.9 | Y75 | 165.0 | 8.3 | 6.4 | 2.7 | 73.1 | 234.0 | 10.9 |
| Y29 | 216.7 | 10.7 | 8.0 | 4.0 | 77.0 | 129.4 | 8.6 | Y76 | 237.4 | 11.4 | 7.4 | 5.0 | 88.0 | 169.2 | 11.5 |
| Y30 | 148.7 | 9.4 | 7.0 | 2.4 | 34.2 | 116.0 | 9.1 | Y77 | 239.8 | 11.2 | 8.4 | 3.8 | 120.4 | 235.8 | 13.4 |
| Y31 | 178.0 | 9.5 | 7.0 | 6.0 | 70.2 | 127.0 | 8.6 | Y78 | 168.2 | 6.9 | 8.4 | 3.4 | 50.2 | 143.4 | 12.8 |
| Y32 | 214.0 | 9.5 | 6.5 | 3.8 | 53.9 | 100.5 | 8.9 | Y79 | 152.5 | 10.0 | 7.5 | 3.5 | 46.0 | 113.5 | 10.2 |
| Y33 | 175.0 | 8.5 | 5.7 | 3.0 | 58.6 | 63.5 | 6.8 | Y80 | 201.6 | 9.7 | 10.9 | 5.9 | 38.3 | 120.8 | 11.5 |
| Y34 | 225.7 | 10.0 | 7.0 | 4.0 | 37.8 | 150.7 | 9.6 | Y81 | 208.9 | 8.6 | 11.4 | 4.6 | 51.9 | 142.0 | 9.0 |
| Y35 | 263.4 | 14.4 | 7.4 | 8.5 | 67.7 | 205.0 | 8.1 | Y82 | 189.4 | 11.8 | 7.2 | 5.6 | 47.8 | 98.2 | 12.5 |
| Y36 | 202.4 | 10.2 | 16.4 | 4.0 | 43.3 | 101.6 | 25.3 | Y83 | 229.0 | 11.4 | 7.2 | 5.2 | 109.0 | 172.5 | 10.6 |
| Y37 | 216.0 | 10.1 | 11.4 | 5.0 | 48.2 | 119.1 | 6.7 | Y84 | 59.6 | 7.4 | 7.7 | 1.8 | 25.6 | 55.5 | 9.1 |
| Y38 | 176.8 | 8.7 | 5.7 | 3.4 | 89.9 | 261.7 | 8.7 | Y85 | 229.8 | 11.4 | 7.0 | 5.4 | 96.6 | 182.5 | 10.4 |
| Y39 | 173.0 | 7.8 | 12.3 | 3.0 | 53.0 | 131.0 | 8.8 | Y86 | 185.7 | 10.7 | 7.0 | 4.4 | 25.6 | 237.0 | 15.2 |
| Y40 | 163.5 | 8.5 | 13.5 | 3.0 | 101.0 | 273.5 | 7.8 | Y87 | 220.4 | 9.6 | 14.4 | 4.3 | 61.6 | 209.6 | 9.5 |
| Y41 | 263.3 | 13.4 | 6.5 | 8.0 | 77.0 | 227.8 | 8.3 | Y88 | 202.8 | 12.6 | 9.2 | 4.4 | 117.0 | 173.0 | 12.5 |
| Y42 | 196.8 | 11.8 | 8.6 | 4.2 | 57.6 | 150.3 | 10.8 | Y89 | 217.4 | 9.7 | 16.7 | 4.8 | 73.7 | 254.2 | 8.2 |
| Y43 | 212.5 | 10.0 | 12.6 | 4.4 | 31.0 | 87.9 | 9.5 | Y90 | 232.9 | 10.1 | 17.1 | 5.0 | 61.4 | 182.2 | 10.1 |
| Y44 | 198.4 | 9.9 | 12.6 | 4.6 | 26.3 | 51.4 | 9.8 | Y91 | 173.0 | 8.2 | 13.5 | 3.0 | 80.2 | 262.0 | 7.9 |
| Y45 | 201.0 | 9.0 | 6.0 | 3.5 | 51.8 | 119.4 | 8.5 | Y92 | 203.4 | 9.7 | 7.0 | 2.0 | 57.4 | 316.7 | 10.1 |
| Y46 | 186.5 | 9.2 | 11.6 | 4.0 | 19.9 | 45.0 | 8.7 | Y93 | 208.7 | 9.8 | 7.0 | 3.0 | 46.6 | 120.0 | 8.4 |
| Y47 | 208.0 | 10.4 | 5.8 | 5.7 | 37.0 | 67.7 | 8.5 | Y94 | 213.5 | 8.8 | 12.1 | 3.6 | 25.6 | 84.1 | 12.6 |

PH: Plant height; SNN: stem node number; PB: panicle branch number; PBN: primer branch nodes; PP: panicles per plant; GNP: grain number per plant; SW: 100-seed weight.

accessions ranged between 6.7 g and 25.3 g. The Y36 accession had the heaviest SW (25.3 g), followed by Y5, Y28, and Y60 with 19.7, 17.9, and 16.1 g, respectively. The lightest SW were observed in Y33 (6.8 g), Y38 (6.7 g), and Y49 (6.8 g).

There were significant genetic variations in all the quantitative traits of cultivated and wild accessions (Table 3). For cultivated species, the PH mean value was 198.6 cm and ranged between 59.6 and 263.4 cm, SD was 32.4 cm, and CV was 16.3%. For the wild species, PH mean value was 200.9 cm and ranged between 148.7 and 263.3 cm, SD was 25.4 cm, and CV was 12.6%. Thus, the variation of cultivated species PH was greater than for wild species. Although the maximum value of cultivated species was similar to that of wild plants, Y35 (263.4 cm) and Y41 (263.3 cm), the minimum was quite different at 56.87 and 150 cm, respectively. Wild species mean variations were higher than for cultivated species in PB, PP, and SW, while SNN and PBN were not significantly different. For GNP, the mean of wild species was higher than for cultivated species at 165.7 and 135.2, respectively.

Table 3. Analysis of seven quantitative morphological traits of 94 Job's-tears accessions.

| Item | Plant height | Stem node number | Panicle branch number | Primer branch nodes | Panicles per plant | Grain number per plant | 100-seed weight |
|-------|--------------|------------------|-----------------------|---------------------|--------------------|------------------------|-----------------|
| | cm | | | | | | g |
| Min | 59.6 | 7.4 | 5.0 | 1.8 | 19.9 | 45.0 | 6.8 |
| | 148.7 | 6.9 | 5.5 | 1.7 | 25.6 | 59.2 | 6.7 |
| Max | 263.4 | 14.4 | 15.7 | 8.7 | 100.4 | 316.7 | 19.7 |
| | 263.3 | 13.4 | 25.6 | 8.5 | 120.4 | 302.8 | 25.3 |
| Mean | 198.6 | 10.3 | 8.5 | 4.7 | 51.4 | 135.2 | 10.1 |
| | 200.9 | 10.0 | 11.1 | 4.5 | 64.4 | 165.7 | 11.0 |
| SD | 32.4 | 1.4 | 2.9 | 1.4 | 21.1 | 65.7 | 2.5 |
| | 25.4 | 1.5 | 4.0 | 1.4 | 27.5 | 63.3 | 3.3 |
| CV, % | 16.3 | 13.2 | 33.7 | 30.3 | 41.0 | 48.7 | 25.2 |
| | 12.6 | 14.9 | 36.4 | 30.1 | 42.6 | 38.2 | 29.9 |

Top value for each item: Cultivated species; bottom value for each item: wild species.

Qualitative trait analysis

Qualitative traits of Job's-tears are shown in Tables 4 and 5. The bract surface characteristic in 80 accessions was smooth (85.1% of the total), and the remaining 14 accessions had longitudinal convex stripes (14.9%). There were 40 accessions with crustaceous bract texture (42.6% of the total), whereas 51 accessions had an enamel texture (57.4%). As for bract shape, five accessions had a circular shape (5.3%), 54 accessions were oval-shaped (57.4%), and 35 accessions length circle shape (37.3%). Regarding TBC, there were 27 white accessions, 6 grayish-white, 3 blue-gray without dark stripes, 3 blue-gray with dark stripes, 1 yellow-white, 4 tawny without dark stripes, 6 tawny with dark stripes, 19 brown, and 25 dark brown that accounted for 28.6%, 6.4%, 3.2%, 3.2%, 1.1%, 4.3%, 6.4%, 20.2%, and 26.6%, respectively. With respect to PC, there were 48 light yellow accessions (51.1%), 12 yellow (12.8%), and 34 brown (36.1%).

Table 4. Qualitative morphological traits of 94 Job's-tears accessions.

| Code | TBSC | TBT | TBS | TBC | PC |
|------|-----------------------------|-------------|---------------|-----------------------------|--------------|
| Y1 | Smooth | Enamel | Oval | Tawny without dark stripes | Light yellow |
| Y2 | Smooth | Enamel | Oval | Brown | Brown |
| Y3 | Smooth | Enamel | Oval | Brown | Brown |
| Y4 | Smooth | Enamel | Oval | Grayish white | Brown |
| Y5 | Smooth | Enamel | Oval | Tawny with dark stripes | Light yellow |
| Y6 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y7 | Smooth | Enamel | Oval | Dark brown | Light yellow |
| Y8 | Smooth | Enamel | Oval | Blue-gray with dark stripes | Light yellow |
| Y9 | Smooth | Enamel | Oval | Brown | Yellow |
| Y10 | Smooth | Enamel | Circular | Yellow white | Brown |
| Y11 | Smooth | Enamel | Oval | Brown | Brown |
| Y12 | Smooth | Enamel | Oval | Tawny with dark stripes | Light yellow |
| Y13 | Smooth | Enamel | Length circle | Tawny with dark stripes | Light yellow |
| Y14 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y15 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y16 | Smooth | Enamel | Oval | Brown | Yellow |
| Y17 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y18 | Smooth | Enamel | Oval | Brown | Yellow |
| Y19 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y20 | Smooth | Enamel | Oval | Blue-gray with dark stripes | Light yellow |
| Y21 | Smooth | Enamel | Oval | Tawny with dark stripes | Light yellow |
| Y22 | Smooth | Enamel | Oval | Grayish-white | Yellow |
| Y23 | Smooth | Enamel | Oval | Grayish-white | Yellow |
| Y24 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y25 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y26 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y27 | Smooth | Enamel | Oval | Brown | Brown |
| Y28 | Smooth | Enamel | Oval | Brown | Yellow |
| Y29 | Longitudinal convex stripes | Crustaceous | Length circle | Brown | Light yellow |
| Y30 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y31 | Smooth | Enamel | Oval | Tawny with dark stripes | Light yellow |
| Y32 | Smooth | Enamel | Oval | Tawny with dark stripes | Light yellow |

Continuation Table 4.

| Code | TBSC | TBT | TBS | TBC | PC |
|------|-----------------------------|-------------|---------------|--------------------------------|--------------|
| Y33 | Smooth | Enamel | Oval | Tawny without dark stripes | Brown |
| Y34 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y35 | Smooth | Enamel | Oval | Grayish-white | Brown |
| Y36 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y37 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y38 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y39 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y40 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y41 | Longitudinal convex stripes | Crustaceous | Length circle | Dark brown | Brown |
| Y42 | Longitudinal convex stripes | Crustaceous | Length circle | White | Brown |
| Y43 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y44 | Smooth | Enamel | Oval | Brown | Yellow |
| Y45 | Smooth | Enamel | Oval | Brown | Yellow |
| Y46 | Smooth | Enamel | Oval | Tawny without dark stripes | Light yellow |
| Y47 | Smooth | Enamel | Oval | Dark brown | Yellow |
| Y48 | Smooth | Enamel | Oval | Tawny without dark stripes | Yellow |
| Y49 | Smooth | Enamel | Oval | Tawny without dark stripes | Light yellow |
| Y50 | Smooth | Enamel | Oval | Brown | Yellow |
| Y51 | Smooth | Enamel | Oval | Brown | Brown |
| Y52 | Longitudinal convex stripes | Crustaceous | Length circle | Blue-gray with dark stripes | Light yellow |
| Y53 | Smooth | Enamel | Oval | Grayish-white | Brown |
| Y54 | Smooth | Enamel | Oval | Dark brown | Light yellow |
| Y55 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y56 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y57 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y58 | Longitudinal convex stripes | Crustaceous | Length circle | Dark brown | Brown |
| Y59 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y60 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y61 | Longitudinal convex stripes | Crustaceous | Length circle | Dark brown | Brown |
| Y62 | Longitudinal convex stripes | Crustaceous | Circular | Blue-gray without dark stripes | Light yellow |
| Y63 | Longitudinal convex stripes | Crustaceous | Length circle | Dark brown | Light yellow |
| Y64 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y65 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y66 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y67 | Longitudinal convex stripes | Crustaceous | Oval | White | Light yellow |
| Y68 | Longitudinal convex stripes | Crustaceous | Oval | White | Light yellow |
| Y69 | Smooth | Enamel | Circular | Dark brown | Brown |
| Y70 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y71 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y72 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y73 | Smooth | Enamel | Oval | Brown | Light yellow |
| Y74 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y75 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y76 | Longitudinal convex stripes | Crustaceous | Circular | Dark brown | Light yellow |
| Y77 | Longitudinal convex stripes | Crustaceous | Length circle | Brown | Brown |
| Y78 | Longitudinal convex stripes | Crustaceous | Length circle | Brown | Brown |
| Y79 | Smooth | Enamel | Oval | Grayish-white | Brown |
| Y80 | Longitudinal convex stripes | Crustaceous | Oval | White | Light yellow |
| Y81 | Longitudinal convex stripes | Crustaceous | Oval | Tawny without dark stripes | Light yellow |
| Y82 | Longitudinal convex stripes | Crustaceous | Length circle | Brown | Brown |
| Y83 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y84 | Smooth | Enamel | Oval | Dark brown | Brown |
| Y85 | Smooth | Enamel | Oval | Brown | Light yellow |
| Y86 | Longitudinal convex stripes | Crustaceous | Length circle | Brown | Light yellow |
| Y87 | Smooth | Enamel | Circular | Dark brown | Light yellow |
| Y88 | Longitudinal convex stripes | Crustaceous | Length circle | Dark brown | Brown |
| Y89 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y90 | Longitudinal convex stripes | Crustaceous | Length circle | White | Yellow |
| Y91 | Longitudinal convex stripes | Crustaceous | Length circle | White | Light yellow |
| Y92 | Smooth | Enamel | Oval | White | Brown |
| Y93 | Smooth | Enamel | Oval | Dark brown | Light yellow |
| Y94 | Smooth | Enamel | Oval | Dark brown | Brown |

TBSC: Total bract surface characteristics; TBT: total bract texture; TBS; total bract shape; TBC: total bract color; FT: fruit type; PC: pericarp color.

Table 5. Conversion value of five qualitative morphological traits of 94 Job's-tears accessions.

| Code | TBSC | TBT | TBS | TBC | PC | Code | TBSC | TBT | TBS | TBC | PC |
|------|------|-----|-----|-----|----|------|------|-----|-----|-----|----|
| Y1 | 1 | 1 | 2 | 6 | 1 | Y48 | 1 | 1 | 2 | 6 | 2 |
| Y2 | 1 | 1 | 2 | 8 | 3 | Y49 | 1 | 1 | 2 | 6 | 1 |
| Y3 | 1 | 1 | 2 | 8 | 3 | Y50 | 1 | 1 | 2 | 8 | 2 |
| Y4 | 1 | 1 | 2 | 2 | 3 | Y51 | 1 | 1 | 2 | 8 | 3 |
| Y5 | 1 | 1 | 2 | 7 | 1 | Y52 | 2 | 2 | 3 | 4 | 1 |
| Y6 | 2 | 2 | 3 | 1 | 1 | Y53 | 1 | 1 | 2 | 2 | 3 |
| Y7 | 1 | 1 | 2 | 9 | 1 | Y54 | 1 | 1 | 2 | 9 | 1 |
| Y8 | 1 | 1 | 2 | 4 | 1 | Y55 | 2 | 2 | 3 | 1 | 1 |
| Y9 | 1 | 1 | 2 | 8 | 2 | Y56 | 1 | 1 | 2 | 9 | 3 |
| Y10 | 1 | 1 | 1 | 5 | 3 | Y57 | 2 | 2 | 3 | 1 | 1 |
| Y11 | 1 | 1 | 2 | 8 | 3 | Y58 | 2 | 2 | 3 | 9 | 3 |
| Y12 | 1 | 1 | 2 | 7 | 1 | Y59 | 2 | 2 | 3 | 1 | 1 |
| Y13 | 1 | 1 | 3 | 7 | 1 | Y60 | 2 | 2 | 3 | 1 | 1 |
| Y14 | 1 | 1 | 2 | 9 | 3 | Y61 | 2 | 2 | 3 | 9 | 3 |
| Y15 | 1 | 1 | 2 | 9 | 3 | Y62 | 2 | 2 | 1 | 3 | 1 |
| Y16 | 1 | 1 | 2 | 8 | 2 | Y63 | 2 | 2 | 3 | 9 | 1 |
| Y17 | 2 | 2 | 3 | 1 | 1 | Y64 | 2 | 2 | 3 | 1 | 1 |
| Y18 | 1 | 1 | 2 | 8 | 2 | Y65 | 2 | 2 | 3 | 1 | 1 |
| Y19 | 1 | 1 | 2 | 9 | 3 | Y66 | 2 | 2 | 3 | 1 | 1 |
| Y20 | 1 | 1 | 2 | 4 | 1 | Y67 | 2 | 2 | 2 | 1 | 1 |
| Y21 | 1 | 1 | 2 | 7 | 1 | Y68 | 2 | 2 | 2 | 1 | 1 |
| Y22 | 1 | 1 | 2 | 2 | 2 | Y69 | 1 | 1 | 1 | 9 | 3 |
| Y23 | 1 | 1 | 2 | 2 | 2 | Y70 | 1 | 1 | 2 | 9 | 3 |
| Y24 | 1 | 1 | 2 | 9 | 3 | Y71 | 1 | 1 | 2 | 9 | 3 |
| Y25 | 2 | 2 | 3 | 1 | 1 | Y72 | 1 | 1 | 2 | 9 | 3 |
| Y26 | 2 | 2 | 3 | 1 | 1 | Y73 | 1 | 1 | 2 | 8 | 1 |
| Y27 | 1 | 1 | 2 | 8 | 3 | Y74 | 1 | 1 | 2 | 9 | 3 |
| Y28 | 1 | 1 | 2 | 8 | 2 | Y75 | 1 | 1 | 2 | 9 | 3 |
| Y29 | 2 | 2 | 3 | 8 | 1 | Y76 | 2 | 2 | 1 | 9 | 1 |
| Y30 | 2 | 2 | 3 | 1 | 1 | Y77 | 2 | 2 | 3 | 8 | 3 |
| Y31 | 1 | 1 | 2 | 7 | 1 | Y78 | 2 | 2 | 3 | 8 | 3 |
| Y32 | 1 | 1 | 2 | 7 | 1 | Y79 | 1 | 1 | 2 | 2 | 3 |
| Y33 | 1 | 1 | 2 | 3 | 3 | Y80 | 2 | 2 | 2 | 1 | 1 |
| Y34 | 1 | 1 | 2 | 9 | 3 | Y81 | 2 | 2 | 2 | 6 | 1 |
| Y35 | 1 | 1 | 2 | 2 | 3 | Y82 | 2 | 2 | 3 | 8 | 3 |
| Y36 | 2 | 2 | 3 | 1 | 1 | Y83 | 2 | 2 | 3 | 1 | 1 |
| Y37 | 2 | 2 | 3 | 1 | 1 | Y84 | 1 | 1 | 2 | 9 | 3 |
| Y38 | 2 | 2 | 3 | 1 | 1 | Y85 | 1 | 1 | 2 | 8 | 1 |
| Y39 | 2 | 2 | 3 | 1 | 1 | Y86 | 2 | 2 | 3 | 8 | 1 |
| Y40 | 2 | 2 | 3 | 1 | 1 | Y87 | 1 | 1 | 1 | 9 | 1 |
| Y41 | 2 | 2 | 3 | 9 | 3 | Y88 | 2 | 2 | 3 | 9 | 3 |
| Y42 | 2 | 2 | 3 | 1 | 3 | Y89 | 2 | 2 | 3 | 1 | 1 |
| Y43 | 2 | 2 | 3 | 1 | 1 | Y90 | 2 | 2 | 3 | 1 | 2 |
| Y44 | 1 | 1 | 2 | 8 | 2 | Y91 | 2 | 2 | 3 | 1 | 1 |
| Y45 | 1 | 1 | 2 | 8 | 2 | Y92 | 1 | 1 | 2 | 1 | 3 |
| Y46 | 1 | 1 | 2 | 3 | 1 | Y93 | 1 | 1 | 2 | 9 | 1 |
| Y47 | 1 | 1 | 2 | 9 | 2 | Y94 | 1 | 1 | 2 | 9 | 3 |

TBSC: Total bract surface characteristics; TBT: total bract texture; TBS: total bract shape; TBC: total bract color; PC: pericarp color.

Correlation coefficient analysis

The Pearson correlation coefficient revealed significant correlations among some variables measured in Job's-tears accessions (Table 6). The PC, TBC, TBS, TBT, and TBSC were positively correlated as well as PH, PBN, and SNN. The PH, PP, and GNP were also positively correlated one with the other. The TBSC and TBT were correlated with PB, PP and GNP. The PB was also positively correlated with TBC and TBS, whereas GNP was correlated with SW.

Principal component analysis

To fully reflect the various factors that played a principal role in the comprehensive indicators, PCA was carried out on seven quantitative traits and five qualitative traits. The Eigenvalues, contribution rate, and accumulative contribution rate were also gathered (Table 7). According to the 85% accumulative contribution rate standard, most agronomic trait

Table 6. Simple correlation matrix for 12 morphological traits of 94 Job's-tears accessions.

| | PH | SNN | PB | PBN | PP | GNP | SW | TBSC | TBT | TBS | TBC |
|------|--------|--------|---------|-------|--------|--------|-------|---------|---------|---------|--------|
| PH | | | | | | | | | | | |
| SNN | 0.51** | | | | | | | | | | |
| PB | 0.08 | -0.17 | | | | | | | | | |
| PBN | 0.49** | 0.57** | -0.08 | | | | | | | | |
| PP | 0.21* | 0.15 | 0.09 | -0.01 | | | | | | | |
| GNP | 0.22* | 0.05 | 0.17 | -0.13 | 0.47** | | | | | | |
| SW | -0.12 | 0.01 | 0.16 | -0.03 | -0.02 | -0.19* | | | | | |
| TBSC | 0.04 | -0.09 | 0.35** | -0.07 | 0.26** | 0.23* | 0.15 | | | | |
| TBT | 0.04 | -0.09 | 0.35** | -0.07 | 0.26** | 0.23* | 0.15 | 1.00** | | | |
| TBS | -0.06 | -0.01 | 0.18* | -0.15 | 0.13 | 0.12 | 0.15 | 0.73** | 0.73** | | |
| TBC | 0.14 | 0.11 | -0.28** | 0.16 | -0.11 | -0.03 | -0.08 | -0.53** | -0.53** | -0.43** | |
| PC | 0.04 | 0.08 | -0.15 | 0.08 | 0.01 | 0.03 | -0.01 | -0.40** | -0.40** | -0.25* | 0.45** |

*, **Significant at $p = 0.05$ and $p = 0.01$, respectively.

PH: Plant height; SNN: stem node number; PB: panicle branch number; PBN: primer branch nodes; PP: panicles per plant; GNP: grain number per plant; SW: 100-seed weight; TBSC: total bract surface characteristics; TBT: total bract texture; TBS: total bract shape; TBC: total bract color; PC: pericarp color.

Table 7. Eigenvalues, proportion of variance, and morphological traits that contributed to first seven principal components (PCs).

| Component | PC1 | PC2 | PC3 | PC4 | PC5 | PC6 | PC7 |
|-------------------------------------|--------|--------|--------|--------|--------|--------|--------|
| Plant height | -0.062 | 0.819 | -0.040 | -0.036 | -0.244 | 0.106 | 0.203 |
| Stem node number | -0.183 | 0.769 | -0.304 | 0.007 | 0.180 | -0.160 | -0.114 |
| Panicle branch number | 0.467 | 0.006 | 0.079 | 0.324 | -0.761 | 0.123 | -0.140 |
| Primer branch nodes | -0.234 | 0.689 | -0.468 | -0.034 | -0.092 | 0.086 | -0.127 |
| Panicles per plant | 0.304 | 0.445 | 0.538 | 0.162 | 0.179 | -0.429 | -0.146 |
| Grain number per plant | 0.275 | 0.371 | 0.738 | 0.008 | -0.027 | -0.016 | 0.172 |
| 100-seed weight | 0.189 | -0.135 | -0.390 | 0.786 | 0.077 | -0.316 | 0.206 |
| Total bract surface characteristics | 0.934 | 0.132 | -0.071 | 0.013 | 0.104 | 0.181 | 0.067 |
| Total bract texture | 0.934 | 0.132 | -0.071 | 0.013 | 0.104 | 0.181 | 0.067 |
| Total bract shape | 0.674 | 0.032 | -0.132 | 0.034 | 0.357 | 0.281 | -0.028 |
| Total bract color | -0.693 | 0.123 | 0.165 | 0.225 | 0.101 | 0.300 | 0.463 |
| Pericarp color | -0.515 | 0.089 | 0.289 | 0.496 | 0.214 | 0.411 | -0.393 |
| Eigenvalues | 3.603 | 2.150 | 1.443 | 1.049 | 0.922 | 0.747 | 0.561 |
| Contribution rate, % | 30.028 | 17.918 | 12.032 | 8.743 | 7.685 | 6.227 | 4.679 |
| Cumulative, % | 30.028 | 47.946 | 59.978 | 68.721 | 76.406 | 82.633 | 87.312 |

information can be summarized in the first seven PCs of the experiment. The accumulative contribution rate accounted for 87.31% of the total variation.

The TBSC, TBT, and TBS were highly loaded in PC1. This indicated that PC1 reflected the main factor of involucre plant parts with an eigenvalue of 3.60, and the contribution rate was 30.03% of the total variation of the studied samples. In PC2, the eigenvalue was 2.15 and the contribution rate was 17.92% of the total morphological variation; among the accessions, these values were mainly explained by PH, SNN, and PBN. This indicated that PC2 was reflected as the main factor of plant type. In PC3, the eigenvalue was 1.44 and the contribution rate was 12.03% of the total variation; GNP and PP was more loaded in PC3. This indicated that PC3 reflected the main factor of yield. In PC4, the eigenvalue was 1.05 and the contribution rate contributed 8.74% of the total morphological variation in these accessions in only SW. The PC5 was 7.69% of the total variation with TBS highly loaded. The PC6 and PC7 accounted for 6.23% and 4.68% and mainly loaded PC and TBC, respectively. In general, for the 12 morphological traits studied, PC1 and PC2 constituted 47.5% of the total morphological variation with most seed-related traits and vegetative traits.

Cluster analysis

According to the cluster analysis results (Figure 1) and the mean value of each group (Table 8), tested Job's-tears accessions were classified in different groups with significant differences in the morphological characteristics. Based on the genetic distance of 10.5, 94 Job's-tears accessions were grouped into seven major clusters.

Figure 1. Cluster map of 94 Job's-tears accessions.

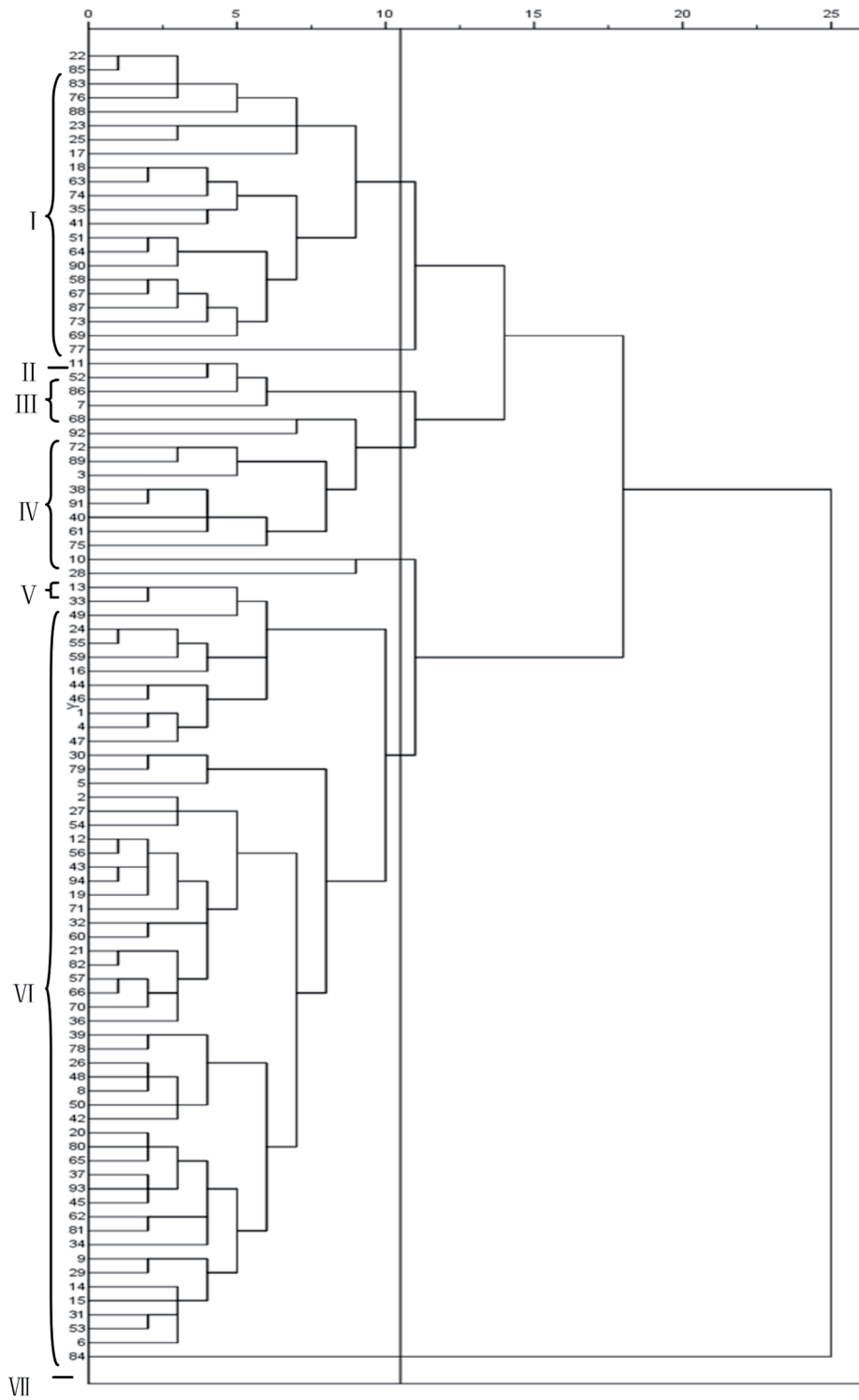


Table 8. Arithmetic mean for traits of each group.

| Group | PH | SNN | PB | PBN | PP | GNP | SW |
|-------|-------|------|------|-----|-------|-------|------|
| I | 223.9 | 11.1 | 11.0 | 5.4 | 74.7 | 187.2 | 10.1 |
| II | 239.8 | 11.2 | 8.4 | 3.8 | 120.4 | 235.8 | 13.4 |
| III | 175.5 | 10.2 | 6.9 | 3.8 | 26.7 | 250.8 | 11.0 |
| IV | 193.2 | 9.3 | 11.5 | 3.5 | 84.1 | 270.5 | 8.8 |
| V | 182.3 | 10.3 | 5.8 | 6.5 | 94.4 | 75.9 | 15.7 |
| VI | 195.6 | 9.9 | 9.1 | 4.5 | 45.5 | 106.3 | 10.6 |
| VII | 59.6 | 7.4 | 7.7 | 1.8 | 25.6 | 55.5 | 9.1 |

PH: Plant height; SNN: stem node number; PB: panicle branch number; PBN: primer branch nodes; PP: panicles per plant; GNP: grain number per plant; SW: 100-seed weight.

Cluster I contained 21 accessions from Guizhou (7), Yunnan (6), Sichuan (3), Qianxinan Institute (3), Hebei (1), and Guangxi (1). The Y22, Y85, Y83, Y76, Y88, Y23, Y25, Y17, Y18, Y63, Y74, Y35, Y41, Y51, Y64, Y90, Y58, Y67, Y87, Y73, and Y69 accessions were classified in this cluster. The main features were PH between 188.8 and 263.4 cm, SNN between 9 and 14.4, PB between 5.5 and 25.5, PBN between 3.8 and 8.5, PP between 42.2 and 117.0, GNP between 136.0 and 227.8, and SW between 7.4 and 12.6 g. This group had relatively high PH, SNN, PB, and PP values (Table 8).

Cluster II contained only Y77. It was a cultivated variety collected in Maguan in Yunnan province, which had the highest PH (239.8 cm), SNN (11.2), and PP (120.4) and higher SW (13.4 g).

Cluster III consisted of 4 accessions from Guizhou (1), Yunnan (1), Sichuan (3), Hebei (1), and Chongqing (1). The Y11, Y52, Y86, and Y17 accessions were classified in this cluster. The main features were PH between 155.4 and 195.0 cm, SNN between 8.3 and 12.0, PB between 6.4 and 7.7, PBN between 1.7 and 4.4, PP between 19.9 and 39.5, GNP between 237.0 and 266.0, and SW between 8.1 and 15.2 g. This group had relatively low PH, PB, PBN, and PP, but it had relatively high GNP and SW.

Cluster IV had 10 accessions from Guizhou (2), Yunnan (3), Sichuan (2), Qianxinan Institute (2), and Chongqing (1). This group had the highest PB (11.5) and GNP (270.5), whereas SW (8.8) was the lowest.

Cluster V only contained Y10 and Y18 collected in two different adjacent counties (Changshun and Puding) in Guizhou province. This group had the highest PBN (6.5). Although this group had lower GNP, with only 78.9, it had the heaviest SW (15.7 g).

Cluster VI had 55 accessions, and this group was collected in Guizhou (24), Yunnan (7), Sichuan (2), Qianxinan Institute (4), Hebei (2), Guangxi (4), Hunan (7), and Chongqing (5). All the characteristics of this group were at a relatively stable intermediate level.

Cluster VII contained only Y84, which was a cultivated variety collected in Quejing in Yunnan province; it had the shortest PH (59.6 cm), the lowest SNN (7.4), PBN (1.8), PP (25.6), and GNP (55.5). In addition, PB (7.7) and SW (9.1) were also relatively lower.

DISCUSSION

Germplasm resource collections are an important step to breed improved crops (Nelson et al., 2011; Andini et al., 2013). Excavating the excellent Job's-tears germplasm was the most important task to improve the species. The results of this study showed that both cultivated and wild accessions have large variations in PB, PBN, PP, and GNP. These accessions have abundant diversity and a great range of optional resources for breeding. The cultivated variety Y36 had 25.3 g SW more than previous findings of 12.5-21.5 g by Wang et al. (2013); it could be used as excellent gene resources to improve varieties with a good combination of agronomic materials such as higher PB and medium PH. The Y92, Y68, Y3, Y40, and Y72 varieties could be used as materials for a high yield variety with more GNP. The cultivated variety Y30 and wild variety Y84, with lower PH, could be used as excellent gene resources for new dwarf varieties. The Y77 accession had higher PH, SNN, PB, GNP, SW, and the highest PP. It was an excellent resource with better comprehensive traits.

Grain yield was closely associated with GNP at harvest (Wang et al., 2013). The analysis of simple correlations among traits revealed that accessions with high PH, SNN, PBN, and PP had high GNP, and positive correlation among accessions suggested that these traits could be used as selection criteria to evaluate accessions. The PP was also positively and

significantly related to GNP, indicating that increasing PP increased GNP. The PP is a good trait for selecting excellent accessions. There were no quantitative traits significantly correlated to SW except GNP, but it was a negative correlation. Generally, when GNP was higher, SW was lighter; therefore, we cannot judge if an accession is good or not only using SW. The qualitative traits had a positive or negative correlation with one another. Some quantitative traits were significantly correlated one another; for example, PB was significantly correlated with TBSC, TBT, TBS, TBC, the PP, and GNP. Positive correlations were recorded for TBSC and TBT. However, the reasons for such a correlation between quantitative traits and qualitative traits are still unclear and need to be studied further.

It is more difficult to analyze the multi-index problems with a number of indicators related one to another. The PCA can simplify multi-index analysis by converting original and more related indices into a new index. Previous studies show that PCA was an effective method for comprehensive crop evaluation (Wang et al., 2013). In the present study, 12 traits of the 94 *Coix* accessions were reduced to seven main components using the PCA method and with the accumulative contribution rate up to 87.31%. The PC1, PC2, and PC3 reflected the effective main factor of involucre, plant type, and yield, respectively. The PC4, PC5, PC6, and PC7 reflected the main factor of SW, TBS, PC, and TBC, respectively. Therefore, 12 trait indices were reduced to seven comprehensive indicators used to represent the original variables, simplify the data, and reveal the relationship between the variables. They can also provide a favorable scientific basis for parent selection in breeding programs (Li et al., 2015). It was also found that PB is one of the most important yield indicators, but it was summarized in PC1 instead of PC2. Therefore, it was necessary to combine the original data and use dialectical analysis methods to remove the apparent phenomenon when we evaluated the quality of germplasm resources by PCA. In addition, breeders must take advantage of the main factors and expand the different traits of breeding materials to accelerate breeding programs for new varieties.

Genetic diversity analysis of germplasms using morphological traits is an initial step for crop improvement (Julia et al., 2016; Peratoner et al., 2016; Loumerem and Alercia, 2016). The variations in morphological traits can be used to classify materials in different groups. The shape, color, and texture of seeds are important to classify *Coix* species (Schaaffhausen, 1952; Rao and Nirmala, 2010). The present study combined quantitative and qualitative traits. The 94 accessions were grouped into seven clusters. Cluster I contained 21 accessions from Guizhou (7), Yunnan (6), Sichuan (3), Qianxinan Institute (3), Hebei (1), and Guangxi (1). The accessions in this group had different plant characteristics that are good for breeding material. Cluster II contained only Y77, which had the highest PH SNN, PP, and SW: this accession can be used as a high-yielding variety and parental material to improve a superior dwarf variety. Cluster III contained 4 accessions, which can be used as excellent resources to promote dwarf, anti-lodging, and high yield varieties. Cluster IV had the highest PB and PP and can be used as excellent parental material to improve crop yield. Cluster V contained only Y10 and Y18 collected from two different adjacent locations, Changshun and Puding in Guizhou province; this indicated that accessions can cluster with geographic locations. Cluster VI was the largest group consisting of 55 accessions. Cluster VII contained only Y84 that had the shortest PH (59.6 cm). It can be used as excellent parent material to cultivate new dwarf and anti-lodging varieties.

Clustering results showed that the accessions collected from different areas could be grouped, such as Y11, Y52, Y86, and Y7 collected from Guizhou, Yunnan, Sichuan, Hebei, and Chongqing, respectively. Some accessions collected from the same area might not be grouped, such as Y35 and Y36 in Cluster I and cluster VI, respectively, which were collected from Ceheng in Guizhou province. This indicated that genetic differences were not influenced by geographic differences, and accessions collected from the same area can be very different as to genetic variation (Li et al., 2010). Genetic differences were based on variety type (wild or cultivated) and qualitative traits were inconsistent with the results of clustering by geographic distribution (Xi et al., 2016). This is because agronomic traits are easily affected by environmental conditions and cultural practices (Bruschi et al., 2003; Wang et al., 2013).

CONCLUSIONS

Morphological diversity analysis in the present study showed high variations among the materials according to ANOVA and simple correlations and multivariate analysis. Significant and positive correlations were found between grain number per plant and among other yield-related attributes. The reasons for such a correlation between quantitative and qualitative traits are still unclear and need further study. However, the information about the correlation of characteristics can be used

for indirect selection and as breeding criteria. The results also indicated that these accessions have the potential to improve yield in the Job's-tears breeding program. Principal component analysis showed that the observed variations were mainly caused by traits such as total bract surface characteristics, total bract texture, total bract shape, plant height, stem node number, and primer branch nodes, and this indicated that their contribution is important to discriminate accessions. Cluster analysis grouped the 94 Job's-tears accessions into seven clusters. This indicated high diversity for most of the traits, and demonstrated that genetic differences cannot only be based on geographic differences, variety type (wild or cultivated), and qualitative traits. Since morphological traits are easily affected by environmental conditions, the genetic relationship cannot only be reflected by the similarity between morphological characteristics.

ACKNOWLEDGEMENTS

The authors would like to thank the Science and Technology Department of Guizhou Province and the Guizhou Academy of Agriculture Sciences for their financial support for the projects of Guizhou talent base construction (Nr QRLF [2016] 22) and Guizhou Academy of Agriculture Sciences special fund (Nr QNKYYZX [2014] 010). The authors would also like to thank the Suranaree University of Technology for their support and technical assistance.

REFERENCES

- Andini, R., Yoshida, S., Yoshida, Y., and Ohsawa, R. 2013. Amaranthus genetic resources in Indonesia: morphological and protein content assessment in comparison with worldwide amaranths. *Genetic Resources and Crop Evolution* 60:2115-2128.
- Bruschi, P., Vendramin, G.G., Bussotti, F., and Grossoni, P. 2003. Morphological and molecular diversity among Italian populations of *Quercus petraea* (Fagaceae) *Annals of Botany* 91:707-716.
- Diao, X. 2017. Production and genetic improvement of minor cereals in China. *Crop Journal* 5:103-114. <https://doi.org/10.1016/j.cj.2016.06.004>.
- Giraldo, E., López-Corrales, M., and Hormaza, J.I. 2010. Selection of the most discriminating morphological qualitative variables for characterization of fig germplasm. *Journal of the American Society for Horticultural Science* 135:240-249.
- Huang, D.W., Kuo, Y.H., Lin, F.Y., and Chiang, W.C. 2009. Effect of adlay (*Coix lachryma-jobi* L. var. *ma-yuen* Stapf) testa and its phenolic components on Cu²⁺-treated low-density lipoprotein (LDL) oxidation and lipopolysaccharide (LPS)-induced inflammation in RAW 264.7 macrophages. *Journal of Agricultural and Food Chemistry* 57:2259-2266.
- Julia, C.C., Waters, D.L.E., Wood, R.H., and Rose, T.J. 2016. Morphological characterisation of Australian ex situ wild rice accessions and potential for identifying novel sources of tolerance to phosphorus deficiency. *Genetic Resources and Crop Evolution* 63:327-337.
- Karimi, H.R., Zamani, Z., Ebadi, A., and Fatahi, M.R. 2009. Morphological diversity of Pistacia species in Iran. *Genetic Resources and Crop Evolution* 56(4):561-571.
- Li, X.H., Huang, Y.Q., Li, J.S., and Corke, H. 2001. Characterization of genetic variation and relationships among *Coix* germplasm accessions using RAPD markers. *Genetic Resources and Crop Evolution* 48:189-194.
- Li, S.Q., Li, X.D., Wang, S.H., and Zhang, Z.L. 2010. Clustering and principal component analysis of introduced black pericarp rice germplasm based on agronomic traits. *Southwest China Journal of Agricultural Sciences* 23:11-15.
- Li, C.H., Wang, Y.Q., Lu, W.J., and Wang, L.H. 2015. The principal component and cluster analysis of agronomic traits of coix germplasm resources in Yunnan. *Journal of Plant Genetic Resources* 16:277-281.
- Lim, T.K. 2012. Edible medicinal and non-medicinal plant. Vol. 1. p. 656-687. Springer, New York, New York, USA.
- Loumerem, M., and Alercia, A. 2016. Descriptors for jute (*Corchorus olitorius* L.) *Genetic Resources and Crop Evolution* 63(7):1103-1111.
- Ma, K.H., Kim, K.H., Dixit, A., Chung, I.M., Gwag, J.G., Kim, T.S., et al. 2010. Assessment of genetic diversity and relationships among *Coix lacryma-jobi* accessions using microsatellite markers. *Biologia Plantarum* 54:272-278.
- Nelson, R.L. 2011. Managing self-pollinated germplasm collections to maximize utilization. *Plant Genetic Resources* 9:123-133.
- Peratoner, G., Seling, S., Klotz, Florian, C., Figl, U., and Schmitt, A.O. 2016. Variation of agronomic and qualitative traits and local adaptation of mountain landraces of winter rye (*Secale cereale* L.) from Val Venosta/Vinschgau (South Tyrol). *Genetic Resources and Crop Evolution* 63:261-273.
- Rao, P.N., and Nirmala, A. 2010. Chromosomal basis of evolution in the genus *Coix* L. (Maydeae): a critical appraisal. *The Nucleus* 53:13-24.
- Schaaffhausen, R.V. 1952. Adlay or Job's tears - A cereal of potentially greater economic importance. *Economic Botany* 6:216-227.

- Taylor, G.D. 1953. Some crop distributions by tribes in upland Southeast Asia. *Southwestern Journal of Anthropology* 9:296-308.
- UPOV. 1991. International convention for the protection of new varieties of plant (UPOV), Geneva, Switzerland. <https://www.upov.int>
- Wang, S., He, J., Nong, M., Zhao, J., and Yang, Z. 2015. Research on SRAP molecular markers in germplasm resources of *Coix lacryma-jobi*. *Chinese Traditional and Herbal Drugs* 46:112-117.
- Wang, S., Zhang, S., He, J., Lu, G., and Yang, Z. 2013. The principal component analysis and cluster analysis of *Coix* resource characteristics. *Journal of Yunnan Agricultural University* 28:157-162.
- Xi, X.J., Zhu, Y.G., Tong, Y.P., Yang, X.L., Tang, L.L., Ma, S.M., et al. 2016. Assessment of the genetic diversity of different Job's tears (*Coix lacryma-jobi* L.) accessions and the active composition and anticancer effect of its seed oil. *PLOS ONE* 11:e0153269. <https://doi.org/10.1371/journal.pone.0153269>.
- Zhou, L.L., Huang, B.B., Meng, X.Z., Wang, G., Wang, F., Xu, Z.K., et al. 2010. The amplification and evolution of orthologous 22-kDa α -prolamin tandemly arrayed genes in *coix*, sorghum and maize genomes. *Plant Molecular Biology* 74:631-643.