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No. 12, Thiruvannamalai Nagar, Saravanampatti - Kalapatti Road, Saravanampatti,  
Coimbatore, Tamil Nadu 641035, India  
Ph: +91 9385339863 | [www.threatenedtaxa.org](http://www.threatenedtaxa.org)  
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Caption: Malabar Slender Loris *Loris lydekkerianus malabaricus* © Dileep Anthikkad.



## A cytomorphological investigation of three species of the genus *Sonchus* L. (Asterales: Asteraceae) from Punjab, India

M.C. Sidhu<sup>1</sup> & Rai Singh<sup>2</sup>

<sup>1,2</sup>Department of Botany, Panjab University, Sector 14, Chandigarh, Punjab 160014, India.

<sup>1</sup>mcsidhu@gmail.com, <sup>2</sup>raibot95@gmail.com (corresponding author)

**Abstract:** Three species of the genus *Sonchus* L. (*Sonchus asper*, *S. oleraceus* and *S. wightianus*) were collected from the Malwa region of Punjab during 2019 to 2020. These species were studied for cytomorphological variations. The species under investigation were identified based on their morphological descriptions. *Sonchus asper* (L.) Hill and *Sonchus wightianus* DC. possess the same number of chromosomes ( $2n=2x=18$ ) whereas *Sonchus oleraceus* (L.) L. is tetraploid with  $2n=4x=32$  chromosomes. Chromosome number of *S. wightianus* ( $2n=2x=18$ ) was worked out for the first time from the state of Punjab. *Sonchus oleraceus* has larger pollens than *S. asper* and *S. wightianus*. This study will be useful for researchers, taxonomists and cytologists for accurate identification of these three species.

**Keywords:** Chromosome number, involucre bract, meiosis, palynology, *Sonchus*, taxonomy.

*Sonchus* L. is a member of the family Asteraceae with 95 species distributed throughout the world including western Morocco, Ethiopia, southern Sudan, South Africa, Canary Island, Europe, Iran, Iraq, Egypt, Afghanistan, and Turkistan (Boulos 1960; Cho et al. 2019). *Sonchus* species are annual to perennial herbs with a milky latex. The stem is clasping, toothed or pinnatifid, segmented leaves; terminal, umbellate, yellow, ligulate-homogamous heads; ovoid, ellipsoid, compressed, ribbed achenes with white hairy pappus which are the important features of the genus *Sonchus* L. (Quireshi

et al. 2002; Rahman et al. 2008). Earlier four species of *Sonchus* (*S. asper* Vill., *S. arvensis* L., *S. oleraceus* L., & *S. maritimus* L.) were reported from British India (Hooker 1882) and undivided Punjab (Bamber 1916). Sharma (1990) enlisted *S. asper*, *S. oleraceus*, and *S. wightianus* from Punjab. Later on, Sidhu (1991) recorded *S. asper*, *S. arvensis*, and *S. oleraceus* from the state of Punjab. *Sonchus asper* and *S. oleraceus* were common in the previous studies whereas *S. wightianus* or *S. arvensis* were frequently misplaced under confusing species.

Morphological parameters have been used for the identifications of plant species for a long time. It is one of the basic, simple and cost effective tools. Morphological features such as leaf shape and color; flower color and type; number, position and nature of androecium and gynoecium; shape and type of fruit and seeds are used for identification of species (Singh & Dey 2005). Chromosome number is also important in the identification of species because species, genera and families have their own unique chromosome numbers in general and basic chromosome number in particular. Variations in chromosome numbers are useful in taxonomic studies (Raven 1975; Jones 1979).

The present study is an attempt to differentiate between previously reported two (*S. arvensis* and

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*S. wightianus*) species (Sharma 1990; Sidhu 1991). Therefore, it is important to look into the detailed morphology of the three species under investigation. Keeping this in view, the present study has been planned to characterize three species of *Sonchus* from the state of Punjab based on morphological and cytological observations.

## MATERIALS AND METHODS

### Collection of study materials

The present study has been undertaken in the Malwa region of the state of Punjab, India. The study material of three species of *Sonchus* was collected during 2019 to 2020. The collected plant specimens were cleaned thoroughly, pressed, and dried at room temperature. After this, the plant specimens were pasted on herbarium sheets. Herbarium specimens were deposited in the Herbarium, Department of Botany, Punjab University Chandigarh (PAN-21994, 21996 and 21997).

### Morphological study

Morphological features of a leaf (arrangement, shape, type, color), stem (glabrous, hairy), flower (colour, type, shape), androecium (number, shape, nature), gynoecium (shape, number, nature) were examined to establish the identity of each of the three *Sonchus* species. The available literature (Hooker 1882; Bamber 1916; Turner et al. 1961; Walter & Kutta 1971; Boulos 1972; Hsieh et al. 1972; Nair 1978; Mejias & Andres 2004; Cho et al. 2019) have been looked into to describe the *Sonchus* species in question. The Herbarium, Department of Botany, Panjab University Chandigarh and online Herbaria have also been consulted for identification.

### Meiotic and pollen study

Meiotic analysis has been carried out in three *Sonchus* species to examine their chromosome numbers. Young flower buds were collected and fixed in the fixative (ethanol 3: glacial acetic acid 1) for 24 hours then shifted to 70% ethanol till further use. Anthers were excised from young flower bud on the glass slide having a drop of acetocarmine and crushed with the help of a glass rod. The material was covered with a micro cover-slip and pressed in two folds of filter paper after gentle heating. Slides were observed under the microscope. Photographs of the pollen mother cells containing countable chromosomes have been taken. For pollen study, mature anthers were taken on the slide and squashed in glycerol acetocarmine (1:1), covered with a cover-slip and observed under the microscope after 24 hours. Uniformly stained pollens (S.P.) were considered

fertile whereas, poorly stained or unstained pollens as sterile. The percentage of pollen fertility was calculated using (Pollen fertility = S.P. / Total Pollens x 100) formula. Pollen size has been measured with the help of camera-lucida technique.

## RESULTS AND DISCUSSION

Three species of the genus *Sonchus*, i.e., *Sonchus asper*, *S. oleraceus*, and *S. wightianus* were collected from the Malwa region of Punjab during 2019 to 2020. All the three species are annual with erect habit. Leaves of *S. oleraceus* are smooth, glabrous, and light green whereas they are dark green in the case of *S. wightianus*. In *S. asper*, leaves are spined and bluish-green. Leaves are elliptic-oblong, half amplexicaul with round auricles in *S. asper* and *S. wightianus* but auricles are spreading in the case of *S. oleraceus* (Image 1,2). Similarly, leaf auricles were found to be round in *S. asper* and pointed to acute in *S. oleraceus* (Barber 1941; Quireshi et al. 2002; Cho et al. 2019). *S. asper* and *S. oleraceus* are very similar to each other in flower colour, i.e., pale yellow to dark yellow whereas the flower colour in *S. wightianus* is orange yellow. Involucral bracts are smooth in *S. oleraceus*, glandular hairy in *S. wightianus* and spiny-



Image 1. Habitat of *Sonchus* L. species (a–c): a—*Sonchus asper* | b—*Sonchus oleraceus* | c—*Sonchus wightianus*. © Rai Singh





Image 2. Morphological details of *Sonchus* species (a–d): a—leaf | b—leaf auricles | c—capitulum | d—involucral bracts. © Rai Singh

hairy in *S. asper* (Image 2). Rahman et al. (2008) also observed glandular and hairy involucral bracts in *S. wightianus* which supports the present study. This feature is important and useful for establishing the identity of *S. wightianus*. Achenes are wrinkled with ribs in *S. asper*, compressed in *S. oleraceus* and finely compressed in *S. wightianus* (Image 3).

#### Identification key (morphology)

- 1 (a) Leaf auricles acute..... *S. oleraceus*
- 1 (b) Leaf auricles round..... 2
- 2 (a) Involucral bracts with glandular hairs .....  
..... *S. wightianus*
- 2 (b) Involucral bracts with spiny hairs ..... *S. asper*

Both *Sonchus asper* and *S. wightianus* are diploid and contain  $2n=2x=18$  chromosomes. Nine bivalents were observed at diakinesis and metaphase-I in *S. asper* and equal segregation of chromosomes (9-9) at anaphase-I in *S. wightianus* (Image 4.a,b,d). Razaq et al. (1994) also

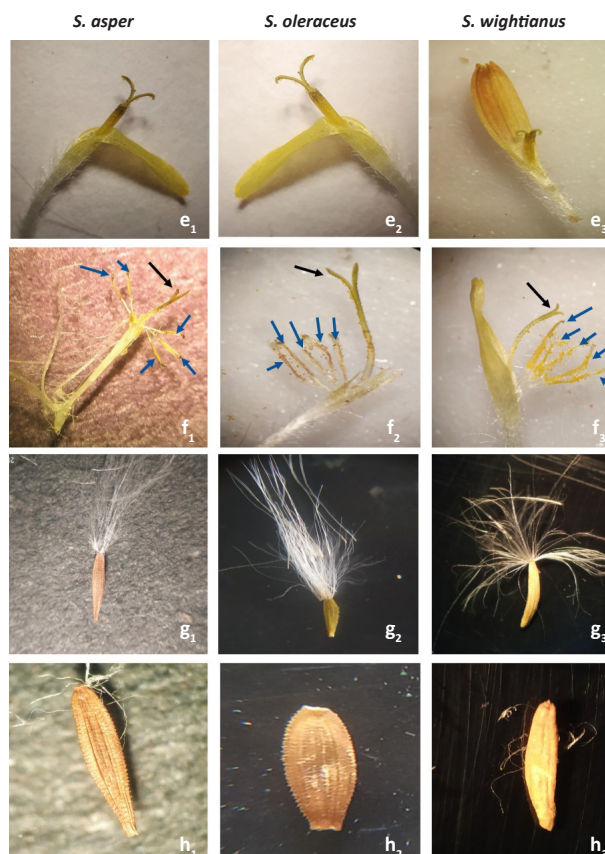


Image 3. Morphological details of *Sonchus* species (e–h): e—flower | f—flower (black arrow showing stigma and blue arrows showing stamens) | g—achene with pappus | h—achene. © Rai Singh

reported chromosome numbers  $2n=18$  in both *Sonchus asper* and *S. wightianus* and  $2n=32$  in *S. oleraceus* from Pakistan.

*Sonchus oleraceus* is a tetraploid and has shown 16 bivalents at diakinesis stage (Image 4c). Present chromosome findings of *S. oleraceus* is in consonance with Ishikawa (1911) who also reported  $2n=4x=32$  chromosome in this species. It has suggested the genetic stability of species even after more than 100 years. But a diploid form of *S. oleraceus* ( $2n=16$ ) and tetraploid ( $2n=32$ ) were previously reported by Marchal (1920) and Cooper & Mahony (1935), respectively. More studies had described *S. asper* as diploid ( $2n=18$ ) and *S. oleraceus* as tetraploid ( $2n=32$ ) (Turner et al. 1961; Walter & Kutta 1971; Boulos 1972; Hsieh et al. 1972; Gupta & Gill 1983; Sidhu et al. 2011; Kaur & Singhal 2015). The variation of chromosome number in *Sonchus* species points towards the incidence of aneuploidy that has happened over time in the genus *Sonchus*.

Pollen size of *S. oleraceus* is  $36.25 \times 32.5 \mu\text{m}$ – $40 \times 33.75 \mu\text{m}$  followed by *S. wightianus* ( $33.75 \times 32.5 \mu\text{m}$ – $36.25 \times 33.75 \mu\text{m}$ ) and *S. asper* ( $31.25 \times 28.75 \mu\text{m}$ – $35$

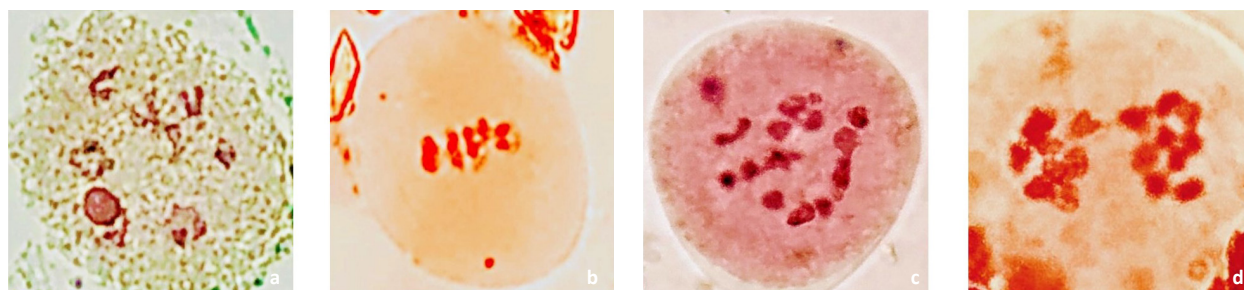


Image 4. Chromosome details of *Sonchus* L. species (a–d): a–b—*S. asper* (n= 9) | c—*S. oleraceus* (n= 16) | d—*S. wightianus* (n= 9). © Rai Singh

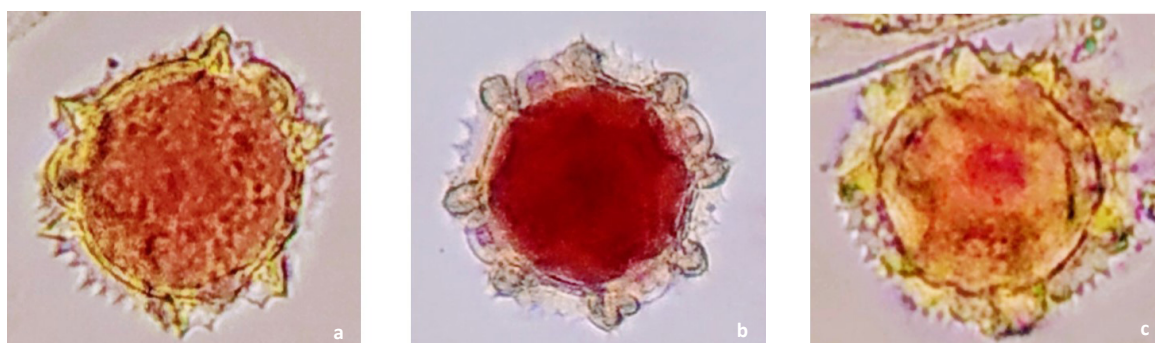


Image 5. Pollen grains of three *Sonchus* L. species (a–c): a—*S. asper* | b—*S. oleraceus* | c—*S. wightianus*. © Rai Singh

x 32.5  $\mu$ m) (Image 5 a–c). Pollen size of *S. asper* and *S. wightianus* is almost similar which may be due to the same number of chromosomes ( $2n=2x=18$ ). Pollens of *S. oleraceus* are larger than the other two species which may be because of its tetraploid ( $2n=4x=32$ ) nature. Pollen fertility was maximum in *S. oleraceus* (94.33%), followed by *S. wightianus* (92.13%) and *S. asper* (88.88%). High pollen fertility in *S. oleraceus* suggested that it is an allotetraploid. These observations are in consonance with Poole (1932) who found that amphidiploids possess a greater degree of pollen fertility.

Earlier three species of *Sonchus* such *S. asper*, *S. oleraceus*, & *S. wightianus* (Sharma 1990) and *S. asper*, *S. oleraceus*, & *S. arvensis* (Sidhu 1991) were documented from the state of Punjab, India. But according to available literature (Shumovich & Montgomery 1955; Mamgain 1998) *S. arvensis* grows exclusively in Europe and is likely confused with *S. wightianus* in India. In literature, from the state of Punjab third species of *Sonchus* was considered as *S. arvensis* but it is actually a *S. wightianus*.

Cytological details of *Sonchus* species are also incomplete from the state of Punjab, India. Previously, Gupta & Gill (1983) had worked out chromosome numbers of three *Sonchus* species (*S. asper* (L.) Hill, *S. brachyotus* DC and *S. oleraceus* L.) from the state of Punjab. However, they have not worked out the

chromosome of *S. wightianus*. Consequently, information about the chromosome number of *S. wightianus* is not known. Therefore, the present study has been carried out for cytomorphological characterization of *Sonchus* species from the state of Punjab India. The findings of the present study will be useful for researchers, cytologists, and taxonomists for correct identification of *Sonchus* species based on morphological, cytological, and palynological details.

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ravi@threatenedtaxa.org

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