A karyomorphological study in *Ledebouria crispa*, Asparagaceae

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**ABSTRACT:** A karyomorphological study was made in *Ledebouria crispa* S.Venter (Asparagaceae). The chromosomes at resting stage were classified as the diffuse type, while those at mitotic prophase were classified as the continuous type. The chromosome number was 2n = 36 reported here for the first time. The 36 chromosome sets at mitotic metaphase showed gradual decrease in size from the longest to the shortest chromosomes and they showed symmetric karyotype due to arm ratio with 34m+2sm. Thus, the 18 pairs of chromosomes classified were comprised from two similar chromosomes and thus, it was concluded as a diploid.

**KEYWORDS:** Asparagaceae, Chromosomes, Karyomorphological study, *Ledebouria crispa*

*Ledebouria* Roth is a genus distributed in India, Madagascar, and sub-Saharan Africa, mainly southern Africa (Manning et al. 2002). This genus is concluded in the family of Asparagaceae, and the species of the genus show deciduous or weakly evergreen perennials and have bulbs. Fifty-three accepted species were reported as this genus (The Plant List 2013). The chromosome numbers of the 20 species of *Ledebouria* reported up to the present were 2n = 16, 20, 22, 24, 26, 28, 30, 34, 38, 40, 44, 45, 46, 48, 55, 58, 60, 64, 66, 68 and 70 (Raghavan and Venkatasubban 1940a, b; Sheriff and Murthy 1946; Delay 1947; Rao 1954, 1956; De Wet 1957; Giménez-Martin 1959; Fernandez and Neves 1962; Mahalakshma and Sheriff 1970; Sharma 1970; Ratter and Miline 1973; Sen 1973a, b; Gill 1978; Sheriff 1979; Sarkar et al. 1980; Valdes-Bermejo 1980; Sheriff and Rao 1981; Subramanian 1981; Vij et al. 1982; Chakraborty and Sen 1983; Vijayavalli and Mathew 1988, 1990; Dixit et al. 1989; Naik 1989; Chakravarty and Sen 1992; Venter 1993; Stedje 1996; Johnson and Brandham 1997; Sheeba and Vijayavalli 1998; Chala 2006; Wetschnig et al. 2007; Haque and Ghosh 2015; Rahangdale and Rahangdale 2016).

*Ledebouria crispa* S. Venter was recorded from Northern Province (the name was changed from Limpopo Province) by Venter and Edwards (1998). This species is highly localized yet, and is probably the one with the most undulate leaves in the genus *Ledebouria* (Hankey 2016). No chromosome data has been documented in *L. crispa*. Thus, the karyomorphological study in *L. crispa* was here made for the first time.

**MATERIALS AND METHODS**
The plants (Fig. 1A) used for the observation were introduced from a nursery located in Osaka Pref., Japan. Somatic chromosomes were observed in growing root tips by the aceto-orcein squash method as in Hamatani et al. (1998). Root tips were harvested and pretreated in 2 mM 8-hydroxyquinoline at 20°C for 2 hrs. before they were fixed in the 3 : 1 ethanol and Acetic acid at ca 10°C for 10 min and stored at below freezing temperature for a few days. Then, they were macerated in the 1:1 mixture of 45% acetic acid and 1 N hydrochloric acid at ca 60°C for 1 min. They were then, stained and squashed in 2% aceto-orcein.

The resting nuclei and somatic prophase chromosomes were classified according to Tanaka (1980) and those on mitotic metaphase chromosomes by centromeric position according to Levan et al. (1964).

**RESULTS**
The results of the observations on chromosome are described in Fig. 1B-E.

The chromosomes in the resting stage showed diffuse type (Fig. 1B). And, the chromosomes in the mitotic prophase showed continuous type (Fig. 1C).

The chromosome number was 2n = 36 (Fig. 1D, E). It was reported for the first time.

On the mitotic metaphase, the 36 chromosomes showed gradual decrease in size from the longest to the shortest (Fig. 1E). And it showed symmetric karyotype due to arm ratio with 34m+2sm. All chromosomes were classified in 18 pairs each were comprised from two similar chromosomes, then it was decided as a diploid.

**DISCUSSION**
On genus *Ledebouria*, chromosome number of 2n = 38 was not reported. On the other hand, Jessop (1970) reported n = 18 or 19 on *L. floribunda*, and Jessop (1972) reported n = 18 on *L. concolor* on the meiosis cells.
On genus *Ledebouria*, detail researches were held on *L. revoluta*. 2n = 20, 22, 26, 30, 38, 40, 44, 45, 46, 58, 60, 64, 68 were reported as the chromosome number of of *L. revoluta* (Raghavan and Venkatasubban 1940a, b; Sheriff and Murthy 1946; Rao 1954; Ratter and Milne 1973; Sen 1973a, b; Sheriff 1979; Sarkar et al. 1980; Valdes-Bermejo 1980; Sheriff and Rao 1981; Subramanian 1981; Chakraborti & Sen 1983; Vijayavalli and Mathew 1988, 1990; Dixit et al., 1989; Nair 1989; Chakravarty and Sen 1992; Stedje 1996; Johnson and Brandham 1997; Sheeba and Vijayavalli 1998; Haque and Ghosh 2015), this species has a large variation of the chromosome number. And n = 10, 11, 17 and 30 were reported on *L. floribunda* (Jessop 1972) which was reported n = 18 or 19 (Jessop 1970), this species also showed a large variation on the chromosome number.

*L. floribunda* and *L. revoluta* are the species which have wide distribution area in genus *Ledebouria*, and they have a habitat near the distribution area of *L. crispa* which is a quite endemic species (Venter 2008). Meanwhile, Venter (2008) suggested that *L. crispa* and *L. undulata* were closely related each other. About the chromosome number of *L. undulata*, Giménez-Martin (1959) reported as 2n = 30. I wish to have chances to research the karyotypes of *L. floribunda*, *L. revoluta*, *L. undulata*, and so on, and to compare with it of *L. crispa*, in the future.

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