

The phylogenetic relationship in the *Lachenalia pusilla* group

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Declaration

“I declare that the dissertation hereby submitted by me for the Magister Scientiae degree at the University of the Free State is my own independent work and has not previously been submitted by me at another university/faculty. I further more cede copy of the dissertation in favour of the University of the Free State”

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List of Abbreviation:

| | |
|-------------------|-----------------------------------------------------|
| % | Percentage |
| °C | Degrees centigrade |
| µl | Microliter |
| 2n | Somatic chromosome number |
| ABI | Applied Biosystems |
| ARC | Agriculture Research Council |
| bp | base pair |
| CO ₂ | Carbon dioxide |
| CTAB | Hexadecyltrimethyl Ammonium Bromide |
| DAPI | 4',6-Diamidino-2-Phenylindole, Dihydrochloride |
| DNA | Deoxyribonucleic Acid |
| dH ₂ O | Distilled water |
| dNTP | Deoxynucleotide triphosphate |
| DMSO | Dimethyl Sulfoxide |
| e.g. | for example |
| EDTA | Ethylene Diaminetetra Acetic Acid |
| Ethanol | Ethyl alcohol |
| Fig. | Figure |
| FISH | Fluorescent <i>In Situ</i> Hybridization |
| G | Gram |
| g. | Gravitational Force |
| GISH | Genomic <i>In Situ</i> Hybridization |
| HCl | Hydrochloric acid |
| i.e. | Id est (that is) |
| INDELS | Insertions/deletions |
| <i>ITS</i> | Internal Transcribed Spacer Region |
| M | Molar |
| MgCl ₂ | Magnesium chloride |
| Min | Minute |
| ml | Milliliter |
| mM | Millimolar |
| N | Gametic chromosome number |
| NaCl | Sodium chloride |
| PCR | Polymerase Chain Reaction |
| <i>psbA-trnH</i> | Intergenic spacer locus |
| <i>rbcl</i> | Ribulose-1, 5-biphosphate carboxylase large subunit |
| RNA | Ribonucleic acid |
| SANBI | South African National Biodiversity Institution |
| subsp. | Subspecies |
| TAE | Tris; Acetic acid; EDTA |
| TE | Tris; EDTA |
| <i>trnF</i> | Transfer RNA gene for Phenylalanine |
| <i>trnL</i> | Transfer RNA gene for Leucine |
| x | Basic chromosome number |

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1. General introduction

Lachenalia Jacq.f.ex Murray is a geophytic genus endemic to the western areas of Southern Africa (Manning *et al.* 2004; Duncan & Edwards 2006).

Common names for *Lachenalia* are Wild Hyacinth, Cape Cowslip (superficial resemblance to genus *Primula*), Leopard Lily (Bryan 1989) (perhaps due to its beautiful black spots and stripes) or in Afrikaans: “viooltjies” or “kalossies” (Crosby 1986). The genus *Lachenalia* is named after Werner de Lachenal (1736–1800), a Swiss professor of botany (Bryan 1989).

The genus consists of about 133 species (Duncan 2012) and comprises approximately 139 taxa (thus subspecies included). More than 80% of the 133 species are found in the Western and Northern Cape (former Cape Province) of South Africa. Duncan (1996, 1999, 2012) recorded that *Lachenalia* species have a geographical distribution from the south-western parts of Namibia, down the western parts of Northern Cape, Western Cape and Eastern Cape Provinces of South Africa; and reaches as far inland as the south-western part of the Free State Province (South Africa). The only limiting factor to the distribution of this genus is its sensitivity to frost (Kapczńska 2009), but even then, some lachenalias survive in extremely cold or high temperatures.

Lachenalia inhabits a very wide variety of habitats, including pure sand on sea level to loam soils at altitudes exceeding 2000 meters or seasonal pools in clay soils (Duncan 2012). The *Lachenalia* specimens can be solitary or found among other vegetation. The flowering

season begins in late March or early April and last until mid-December with the exception of a summer growing species, *L. pearsonii* (Glover) W.F. Barker from Namibia (Duncan 1999).

The centre of origin of *Lachenalia* is the 3319 grid (Worcester) in the Western Cape Province of South Africa with species diversity decreasing towards the margins of its range (Duncan 2005, 2012). Even though *Lachenalia* species are widely distributed, a significant number of species are listed as endangered (10%), vulnerable (17%), considered to be near threatened (2%), critically rare (6%), rare (9%) and declining (2%) (http://www.sanbi.org/index.php?option=com_docman&task=documentdetails&id=43) and the list is increasing dramatically every year (Duncan 2003). In 2012, the number of vulnerable species significantly increased by more than 100%, endangered species showed an increase of 4% and 50% for critically endangered species (Duncan 2012). *Lachenalia moniliformis* and *L. mathewsii* for example, have a restricted distribution contributing to their vulnerability (Duncan 1998). These species, like other endangered species, grows in the area under development for human inhabitants (for example Cape-flats). Critically endangered *L. viridiflora* is threatened by coastal housing development on the Cape west coast (Duncan 2012). Endangered and vulnerable species are threatened by coastal housing development and alien plant infestation respectively (Duncan 2012).

1.1. Medicinal application

Lachenalias are known for their natural biochemical compounds and scents (Duncan 2012). Most of those natural scents are to facilitate and promote pollination and seed dispersal vectors. Even though there's a lot of natural scents and medical importance, phytochemical characterisation of Hyacinthaceae, *Lachenalia* has not been investigated much (Arnold *et al.* 2002). Only *L. flava* (*syn. L. tricolor* Jacq.f.) has been investigated

pharmacologically (Watt & Breyer-Brandwijk 1962). Based on chemotaxonomic trends reported on the subfamilies of the African Hyacinthaceae, *Lachenalia* in the Hyacinthoideae is expected to produce homoisoflavanone (Pohl *et al.* 2000). In South Africa, indigenous bulbous plants, mainly belonging to the Amaryllidaceae and Hyacinthaceae families are normally used (by natives) as disinfectants and anti-inflammatory agents, suggesting some degree of antimicrobial activity (Louw *et al.* 2002). Few authors investigated chemical compositions of *Lachenalia*, chelidonic acid (Ramstad 1953) and flavone sulphates, tricetin, diosmetin and luteolin sulphate (Williams *et al.*, 1976). Recently Langois *et al.* (2005) identified 3-benzylchromone from *L. punctata*.

The other importance of this geophytic genus beside its potential medicinal value is the commercial flora value. *Lachenalia* specimens can be cultivated and, a complete collection of all the species is maintained at South African National Biodiversity Institution (SANBI).

1.2. South African and Global floriculture industry

The global floriculture industry is worth more than US\$33 billion (Boshoff 2010) and is constantly growing. The floriculture export revenue for South Africa amounted to more than R524 million in 2008 (Boshoff 2010). The flower-bulbs market is estimated to be worth more than US\$1 billion (Kamenetsky & Miller 2010). Kleynhans & Spies (2011) described the floriculture industry as a market on the move. It is closely linked to fashion and life style resulting in ever changing demands and requires new floral products. New innovations and adapting to market changes are vital as failure will have catastrophic results for plant growers and breeders. New innovations include, among other, the development of new cultivars and different uses for existing crops (Kleynhans & Spies 2011).

South Africa has an extremely rich plant diversity which consists of approximately 10% of the world's plant species. Five plant species native to South Africa (*Gerbera* L., *Freesia*, *Zantedeschia*, *Gladiolus* and *Ornithogalum*) generated a turn-over revenue of more than €218 million on Dutch auctions in 2009 (Anon 2010 as quoted by Kleynhans & Spies 2011). None of these cultivars have been developed in South Africa (Kleynhans & Spies 2011). The popular crops sold in South Africa are mainly roses ($\pm 30\%$), *Chrysanthemum* (15%), Lilies (10%) and carnations (6%) all of which are mainly produced in greenhouses (Kleynhans & Spies 2011). The rest consists of summer flowers and proteas, which are mainly shade net or field produced (Kleynhans & Spies 2011). Like Dutch auctions, the South African market is dominated by Multiflora auction structures located in Johannesburg and supermarkets (Kleynhans & Spies 2011).

In 1985 The Indigenous Bulb Growers Association of South Africa, marked the genus *Lachenalia* as the second most popular plant in the world following the genus *Gladiolus* (Duncan 1988).

1.3. *Lachenalia* breeding and new cultivars

Lachenalia has been used in a breeding program at the Agricultural Research Council's Vegetable and Ornamental Plant Institute (ARC-VOPI) with the aims to increase commercialized production of *Lachenalia* plants (Du Preez *et al.* 2002) and to develop new cultivars for the international market. Early attempts of commercializing *Lachenalia* cultivars were unsuccessful due to inadequate cultivation procedures (Kleynhans & Hancke 2002), as well as the political isolation of the country until late 1990's (Kapczńska 2009). In 1992 the breeding programme and production of *Lachenalia* increased significantly and in 1998 and 1999 ARC developed a production system to satisfy the commercial growers' requirement

internationally (Kleynhans 2006; Kapczyńska 2009). The intense labour work, technology and breeding management applied by ARC resulted in a revolution of *Lachenalia* hybrids. In 1999 a *Lachenalia* hybrid 'Rupert' was honoured as the best bulbous pot plant (Sochacki 2003). The first inter-species crosses were made in 1968 at ARC-VOPI breeding programme (Kleynhans 2006). Even though this plant has been used in a breeding programme since 1968, it has not really gained much exposure or popularity in the horticultural industry. Bester *et al.* (2009) mentioned obstacles faced by this plant in floriculture industry and the authors further discussed methods that can be used to improve a market for this plant. Hundreds of crossing combinations have been made and more than 25 cultivars have been released (Kleynhans *et al.* 2009b). However, there are external and internal crossing barriers in the genus (Kleynhans 2006) limiting new cultivars development. Kleynhans & Hancke 2002) discussed most of external barrier that exists in *Lachenalia* breeding. Amongst them, the morphological variation in the genus cause a number of isolation barriers (Lubbinge 1980). Variation in the flowering time of species (April to November) makes crossing difficult (Kleynhans & Hancke 2002).

Kleynhans (2006) described a way to overcome external barriers by growing the plants in controlled conditions and by storing of pollen in -4°C or in liquid nitrogen or by storing dry pollen for a 24 months period in a refrigerator. Stored pollen is used to overcome different flowering periods (April to November). Dry pollen stored in a refrigerator retained 80% of its germination ability when stored for up to 24 months (Kleynhans *et al.* 1995). Kleynhans (2006) observed similar results with pollen storage in liquid nitrogen. *Lachenalia reflexa* seed have high initial viability but do not persist in the soil seed bank for more than three years (Kate & Grazyna 2011).

Internal barriers have not been studied in detail (Kleynhans *et al.* 2009b) and in general a study on internal crossing barriers is necessary to assist breeders to understand low cross-ability. Chromosome variation in the genus is linked to external barriers. The study of Kleynhans *et al.* (2012) indicates a correlation between the cross-ability of *Lachenalia* species and their basic chromosome numbers, as well as their phylogenetic relationships.

Incompatibility between species is another isolation barrier among *Lachenalia* accessions (Kleynhans & Hancke 2002). Kleynhans (2002) reported, intra-species crosses between different accessions sometimes overcome accession incompatibility, and recommended combining accessions collected from different areas to improve cross-ability. Incompatibility is associated with flora incompatibility: (1) flower length (and short style) restrict pollen to grow down the style of a longer flower (Lubblinge 1980) which leads to (2) failure of the pollen tube to reach the ovary, (3) abnormal penetration of the pollen tube in ovule, and (4) embryo abortion and non-viable seeds (Kleynhans & Hancke 2002). The *Lachenalia* breeding program is still relatively young as compared to those on other large bulbous crops; therefore it's not surprising that there are still no developed mechanisms to overcome existing crossing barriers.

1.4. Division of genus *Lachenalia*

Lachenalia makes beautiful ornamental plants and is therefore an important genus of the family Hyacinthaceae (Manning *et al.* 2004; Hamatani *et al.* 2007) or *Asparagaceae* Juss, according to the reclassification in 2009 (APG III group 2009).

Lachenalia species are morphologically extremely variable (Duncan 1996, 1998; Duncan & Edwards 2006; Kleynhans 2006) and presents taxonomic difficulties (i.e. due to its high degree of variation, the *Lachenalia* species are easily confused and wrongly identified by

inexperienced identifiers). The number of species in the genus increased as new taxa were added, i.e. the former genus *Polyxena* has been included in the genus *Lachenalia* (Manning *et al.* 2004).

Based on chromosome number and cross-ability, Crosby (1986) divided the genus *Lachenalia* into five provisional groups: (1) *Lachenalia aloides* group, (2) *Lachenalia orchioides* group, (3) *Lachenalia unicolor* group, (4), *Lachenalia unifolia* group and (5) *Lachenalia pusilla* group. *Lachenalia pusilla* group is the smallest of the five groups with only one species, *L. pusilla* Jacq. ($x = 7$). Baker (1897) placed the *Lachenalia pusilla* group into the subgenus *Brachyscypha*. In the Duncan (1988) and Manning *et al.* (2002) classification, *Lachenalia* was divided into five groups and *L. pusilla* species (the only member of *Lachenalia pusilla* group) was placed in group 1 and subgroup 2c.

Recently, Duncan (2012) re-divided subgenus *Lachenalia* into five sections namely, *Lachenalia*, *Urceolatae*, *Oblongae*, *Augustae* and *Latae*. Duncan (2012) grouped *L. pusilla*, the only species belonging to *Lachenalia pusilla* group (Crosby 1986) into subgenus *Lachenalia*, section *Lachenalia* which consists mainly species with $x = 7$ except for only two species *L. unifolia* ($x = 11$) and *L. isopetala* ($x = 10$). Remarkably this dwarf geophyte species (*L. pusilla*) possess homologous morphological characters with the subgenus *Polyxena* species ($x = 12, 13$ and 14) and one subgenus *Lachenalia*, section *Lachenalia* species (*L. barkeriana*, $x = 7$). Additionally, *L. pusilla* and *L. barkeriana* look different from other section *Lachenalia* species, but similar to members of subgenus *Polyxena*.

There is also high morphological resemblance between the sections of subgenus *Lachenalia*. For example, the prostrate leaves of the *Lachenalia pusilla* group are also found in

other sections, *Lachenalia*: *L. trichophylla* ($x = 7$); *Oblongae*: *L. stayneri* ($x = 11$), *L. nardousbergensis* and section *Latae*: *L. nervosa* ($x = 8$).

In this study, *Lachenalia pusilla* group (Crosby 1986) refers to all the species within genus *Lachenalia* with basic chromosome number of $x = 9, 10$ and 13 , plus $x = 7$ and $x = 8$ species that group with these on a phylogenetic tree. These species are believed to have originated from natural hybridization-polyploidization among species with relative basic chromosome numbers.

1.5. Aim

The aim of this study is to determine the phylogenetic relationship within the *Lachenalia pusilla* group ($x = 9, 10, 13$) plus $x = 7$ and 8 species clustering with $x = 9, 10$, and 13 species. To determine whether this group is a hybrid swarm through a combination of cytogenetic (GISH) and molecular systematics (*ITS*, *trnL-F*, *rbcl*, *psbA-trnH*) techniques. Finally, to determine if multiple gene region analysis (*ITS*, *trnL-F*, *rbcl*, *psbA-trnH*) are sufficient for species level phylogeny in the genus.

1.6. Dissertation outline

This dissertation consists of two sections. The first section is a literature review (chapters 1, 2 and 3) and the last section (chapters 4 and 5) is research work. **Chapter 1** gives a general history of the genus *Lachenalia*, its medical application, new cultivar production and *Lachenalia* in the floriculture industry.

Species within the same group (cladogram clade) are closely related. The high number of successful artificial inter-species crosses (ARC *Lachenalia* breeding programme) among the species within the groups support that they are sister species. Therefore, an accurate

Lachenalia classification is a fundamental knowledge for breeders and taxonomists. **Chapter 2** elucidates *Lachenalia*'s classification based on morphological, chromosome number and karyotyping and finally based on molecular systematics.

Phenological, ecology and geographic distribution can provide guidance understanding genus *Lachenalia* phylogenetic relationship and can be used as a guidance to predict the model of species evolution and population establishment. **Chapter 3** briefly gives a general overview of the factors promoting the survival of the genus *Lachenalia*, and recommendation for growing *Lachenalia* species. Phenotypic plasticity, pollination syndrome and polyploidization interactions are beyond the scope of this study, but may have a significant role in *Lachenalia* evolution.

Chapter 4 is a research work chapter. This chapter is designed to map genome origin of taxa with basic chromosome number of $x = 9, 10, 11$ and 13 . The genome of putative parental taxa with $x = 7$ and $x = 8$ is also analysed to determine their genetic constitution. Lastly, briefly troubleshooting suggestion for GISH analysis of the genus *Lachenalia* were given.

Chapter 5 is a research work chapter. Combined molecular analyses of a nucleus-plastid dataset were used to reconstruct the phylogenetic relationship of *Lachenalia* species. To validate the hypothesis that the *Lachenalia pusilla* group is a hybrid swarm, network and dendroscope analyses were incorporate.

The last two chapters **Chapter 6** and **Chapter 7** are general discussion and references respectively.

2. Review of *Lachenalia* classification based on morphological, cytogenetic and molecular data

2.1. Abstract

Recent progress in identification and classification of the genus *Lachenalia* Jacq. f. ex Murray was briefly review in this chapter. Great taxonomic confusion exists in the genus due to high level of diversity of the leaves, inflorescence and flowering time. Polyploidy and aneuploidy, which occur in abundance, may form more morphological variation that may lead to miss identification and classification of the species in this genus. In some cases, the occurrence of B-chromosomes may lead wrong chromosome count that may lead to miss identification. In some cases, accessions with similar ploidy levels differ morphologically in different geographic areas. Additionally, very similar accessions may possess different somatic chromosomes numbers ($2n$) or very different species may have the same somatic chromosome number and similar ploidy level.

Species with basic chromosome numbers of $x = 7$ and 8 are at the bottom of the phylogenetic tree and represent primary chromosomes and other groups scattered around group $x = 7$ and $x = 8$. Interestingly, cytogenetic studies and molecular systematics indicates that there are two separate clades within the $x = 7$ group.

This review emphasises on the classification of the genus *Lachenalia* based on morphological characters, cytogenetical data and molecular data. Secondly, discuss phylogeny relationships unfeasibility within the genus and lastly, discuss the correlation between monophyletic species, basic chromosome number and their cross ability.

Keywords: basic chromosome number, *Lachenalia*, molecular data, morphological data, variation

2.2. Introduction

The dwarf *Lachenalia pusilla* Jacq. geophyte (10- 20 mm) (Nordenstam 1982; Müller-Doblies *et. al.* 1987), is a diploid ($n = 7$). The high morphological variation of *Lachenalia* species might be the by-product of speciation on the species level. For example, *Lachenalia*

hirta ($x = 11$) and *L. unifolia* ($2n = 11$) differ mainly in the presence of hairiness. Van Rooyen *et al.* (2002) suggested that these two species represent two subspecies of the same species. However, morphological dataset placed these species in two distinct groups (Duncan *et al.* 2005). The former *Polyxena* species and some other *Lachenalia* species had been moved within the genus or to the sister genera based on morphological data, and were reclassified or repositioned into a better resolution on phylogenetic tree using molecular data (Pfosser & Speta 1999; Pfosser *et al.* 2003; Spies 2004).

There is a correlation between the cross-ability of *Lachenalia* species and their basic chromosome number (Kleynhans *et al.* 2009b). Similar to Spies (2004), the authors also found a strong correlation between species within a provisional group in their phylogenetic relationship and crossing ability. Thus basic chromosome numbers of *Lachenalia* species can be used to construct their phylogenetic relationship.

In the integrated study presented here, the classification of the genus *Lachenalia* based on (1) morphological data, (2) cytogenetic and karyological data and (3) molecular data was re-evaluate, and where possible suggest few techniques or evolutionary algorithms to improve the phylogenetic modeling of the relationships in *Lachenalia*.

2.3. *Lachenalia* classification

2.3.1. Classification of *Lachenalia* based on morphological data

It is very difficult to study phylogenetic relationships, classification, speciation and other evolutionary biology in *Lachenalia* (Hamatani *et al.* 2008). This unfeasibility of evolutionary determination is due to (but not limited to) high phenotypic variation. There is taxonomic confusion due to high level of morphological variation with overemphasis of minor

morphological difference in *Lachenalia* (Duncan 1992). In contrast, a recent study of Duncan (2012) emphasized that the majority of *Lachenalia* species have clear-cut morphological differences leading to straight forward identification. A broad array of data collection (e.g. morphology, palynology, cytogenetics, anatomy, ecology, biogeography and DNA techniques) may results in an improved classification of *Lachenalia*.

Variation within species occurs in several macro-morphological characters. Plants belonging to the same species display constant features such as bulb shape and size, flower shapes and seed morphologies (Duncan 2005), inflorescences and flora make-up. Resemblance complexes can be differentiated by a combination of morphological features that constitute unique species characters (Duncan 2012) rather than treating each character individual. Species like *L. bifolia*, *L. contaminata*, *L. elegans*, *L. mutabilis*, *L. orchioides*, *L. punctata* and *L. violacea* are highly variable (Duncan 2005), and can be used as a guide for classification based on their morphological characters (i.e. species sharing unique morphological characters similar to those of seven species ('clades') mentioned above can be assumed to be phylogenetically related and falls under the same clade on the cladistics).

Based on morphological data, Baker (1897) classified *Lachenalia* into five subgenera. Crosby (1986) and Manning *et al.* (2002) dividing *Lachenalia* into five groups. Ten subgroups were rearranged based on morphological similarities into five groups by Duncan (1988). However, cladistic analysis based on 73 characters that included flowers, bulbs and seeds indicated that all these morphological data could be inadequate to clarify and justify the species phylogenetic relationships (Duncan 1998; Duncan *et al.* 2005).

Duncan (1988) described a high degree of *Lachenalia* leaf variation based on leaf shape, length, width and leaf number. The leaves vary from short-prostrated and spreading to long-erected and cylindrical shapes (i.e. *Lachenalia contaminata* produces grass-like leaves, whereas *L. latifolia* produces leaves lying horizontally on the ground surface) (Fig. 2.1). Furthermore, leaves can be spotted, banded, smooth or hairy (Kapczńska 2009) (Fig. 2.1). The variation is further extended into inflorescences as they vary from geoflorous raceme to long-pedicelled raceme, or from corymbose recame to widely campanulate perianth shapes (Duncan 1988) (Fig. 2.1). The variation in inflorescences resulted in formation of three main types of inflorescences: the spike, the subspicate inflorescences and the raceme (Duncan 2012) (Fig. 2.1). The other factor which might have promoted the popularity of this genus is the spectrum of different flower colours (Duncan 1988). Flower colours range from yellow, red, purple and green to white with different spot patterns (Fig. 2.1). Furthermore, the production of new cultivars (Fig. 2.1) push the colour range of this genus to levels never imagined. Some species have fragrant flowers, for example *L. convallarioides* (Kapczńska 2009).



Figure 2.1. Examples of the morphological diversity of the leaves and inflorescences/flowers and different somatic chromosome numbers ($2n$) in *Lachenalia*. **A-J**, leaf variation: **A**, long and thick erect leaves, *L. namaquensis*; **B**, prostrated rough leaves, *L. unicolor*; **C**, green grass-like immaculated leaves, *L. zeyheri*; **D**, grass-like with strips from above the ground to the middle *L. unifolia* var. *unifolia*; **E**, solitary edge folded-like leaf with simple trichomes, *L. hirta*; **F**, thin erect, twisted-lanceolate leaves, *L. mediana*; **G**, spotted semi-erect leaves, *L. aloides*; **H**, red

circular cubic postulated leaf, *L. patula*; **I**, suberect spotted leaf, always occur in two, *L. cernua*; **J**, leaves heavily covered with stellate trichomes, *L. trichophylla*. **K-T**, inflorescences and flower variation: **K**, deep-blue spike inflorescence with tubular perianth, *L. viridiflora*; **L**, the geoflorous, subcapitate raceme, *L. violaceae*; **M**, corymbose raceme *L. corymbosa*; **N**, maculated widely campanulate perianth with short-pedicelled raceme, *L. xerophila*; **O**, immaculated white widely campanulate perianth, *L. orthopetala*; **P**, short-pedicelled raceme with urceolate perianth, *L. congesta*; **Q**, unique flower shape of ex genus *Polyxena*; **R**, maroon spike, *L. isopetala*; **S**; long-pedicelled raceme, *L. flava*; **T**, short-pedicelled raceme with postulate urceolate perianth. **U-Y**, Somatic chromosomes number: **U**, $2n = 2x = 20$, *L. undulata*; **V**, $2n = 2x = 28$, *L. cernua*; **W**, $2n = 2x = 16$, *L. capensis*; **X**, $2n = 2x = 18$, *L. mediana*; **Y**, $2n = 2x = 14$, *L. mathewsii*. Photographer: **A, B, D, E-I, L-U**: P. Spies; **C**: R. Kleynhans; **V-Y**: slides prepared by S. Reinecke. Google images: **J**: <https://www.strangewonderfulthings.com>; **K**: <https://www.flickrriver.com>

2.3.2. Classification of *Lachenalia* based on cytogenetics

The other factor resulting into an unconvincing *Lachenalia* phylogenetical relationship is the variation of chromosome numbers. Chromosome numbers of approximately 89 taxa have been determined (Spies *et al.* 2011). The variation in chromosome number is due to different somatic chromosome numbers, which range from $2n = 10$ to $2n = 56$ (Moffett 1936; Sato 1942; Therman 1956; De Wet 1957; Fernandes & Neves 1962; Riley 1962; Mogford 1978; Ornduff & Watters 1978; Nordenstam 1982; Crosby 1986; Hancke & Liebenberg 1990; Johnson & Brandham 1997; Kleynhans 1997; Hamatani *et al.* 1998, 2004, 2009, 2010, 2012; Hancke & Liebenberg 1998; Kleynhans & Spies 1999; Spies *et al.* 2000, 2002; Du Preez *et al.* 2002; Van Rooyen *et al.* 2002).

The taxa with basic chromosome numbers of $x = 7$ and $x = 8$ are more common within the genus, but $x = 5, 6, 9, 10, 11, 12, 13, 14$ and 15 have also been found (Moffett 1936; Ornduff & Watters 1978; Crosby 1986; Johnson & Brandham 1997; Hancke *et al.* 2001; Hamatani *et al.* 2007, 2010, 2012; Spies *et al.* 2008, 2009;). Polyploidy is also frequent in the genus (Johnson & Brandham 1997; Kleynhans & Spies 1999; Spies *et al.* 2000, 2002) and B-chromosomes have been reported in eight species, namely *L. aloides*, *L. anguinea*, *L. bifolia*, *L.*

carnosa, *L. contaminata*, *L. obscura*, *L. reflexa* and *L. splendida* (Crosby 1986; Hancke & Liebenberg 1990; Johnson & Brandham 1997; Kleynhans & Spies 1999; Spies *et al.* 2009, 2011). A chromosome count of $2n = 23$ in an accession of *L. zeyheri* Baker is probably due to the occurrence of a B-chromosome (Hamatani *et al.* 1998). Species with basic chromosome numbers of $x = 7$ have high occurrences of polyploidy (Spies *et al.* 2002).

Chromosome damage occurring during slides preparation might result in chromosome misidentification and B-chromosome identification (Kleynhans *et al.* 2012). And often only one specimen is studied (Spies *et al.* 2011). The small size of the chromosomes within the genus can lead to miss-identification of B chromosomes and unfeasible cytology studies (Hancke & Liebenberg 1990; Spies *et al.* 2000). Therefore, it is important to analyse and determine the chromosome number of several specimen belonging to specific species to have accurate chromosome counts and correctly identify of the presence of B-chromosomes as well as somatic chromosome numbers.

The occurrence of B-chromosomes reported can also lead to taxa and taxonomic confusions based on chromosomal classification. Hancke & Liebenberg (1990) identified and described the properties of B chromosomes in *Lachenalia* for the first time. According to Hancke & Liebenberg (1990) B-chromosomes in *Lachenalia* do not have a specific staining pattern and are similar in size to the smallest chromosome in the normal complement. Due to this behaviour, it is difficult to identify B-chromosomes in the genus *Lachenalia* and results in some erroneous counts reported in literature. Moreover B-chromosomes in *Lachenalia* do not occur in all cells of a specific individual and also not in all accessions of a species (Hancke & Liebenberg 1990).

2.3.2.1. Basic chromosome numbers in the genus

$x = 7$

The basic chromosome number of $x = 7$ is the most common in the genus *Lachenalia*. About 46.5% of *Lachenalia* specimens studied forms an $x = 7$ complex, with 60.8% of those species being diploid (Fig. 2.2) (Johnson & Brandham 1997; Spies *et al.* 2011). Polyploidy in the $x = 7$ complex is easily identified. However, hexaploids or octoploids of $x = 7$ may be confused with uneven polyploids of taxa with $x = 6$ and $x = 8$ (Spies *et al.* 2011), and can also be confused with diploids with $2n = 30$ ($x = 15$) (Johnson & Brandham 1997). These ploidy levels (hexaploids and octoploids) could actually be allotetraploids derived from taxa with $x = 7$ and $x = 8$ following hybridisation and doubling of the chromosome number (Johnson & Brandham 1997). All other varieties of this group other than hexaploids, octoploids and diploids with $2n = 30$ ($x = 15$) are multiples of $x = 7$ making it clear that they must have a basic number of $x = 7$ (Johnson & Brandham 1997; Spies *et al.* 2011). Polyploidy is more frequent in the $x = 7$ group (Fig. 2.2) (Johnson & Brandham 1997; Kleynhans & Spies 1999) and 17.8% of the specimens studied represents polyploids (Spies *et al.* 2011).

Only 3 out of the 273 cytogenetic reports on $x = 7$ taxa represent specimens at uneven ploidy levels, $2n = 3x = 21$ (*L. aloides* and *L. rosea* Andrews) (Crosby 1986) and $2n = 7x = 49$ (*L. bifolia*) (Kleynhans & Spies, 1999). Uneven ploidy levels are extremely rare making up to 1% of the chromosome numbers reported (Spies *et al.* 2011).

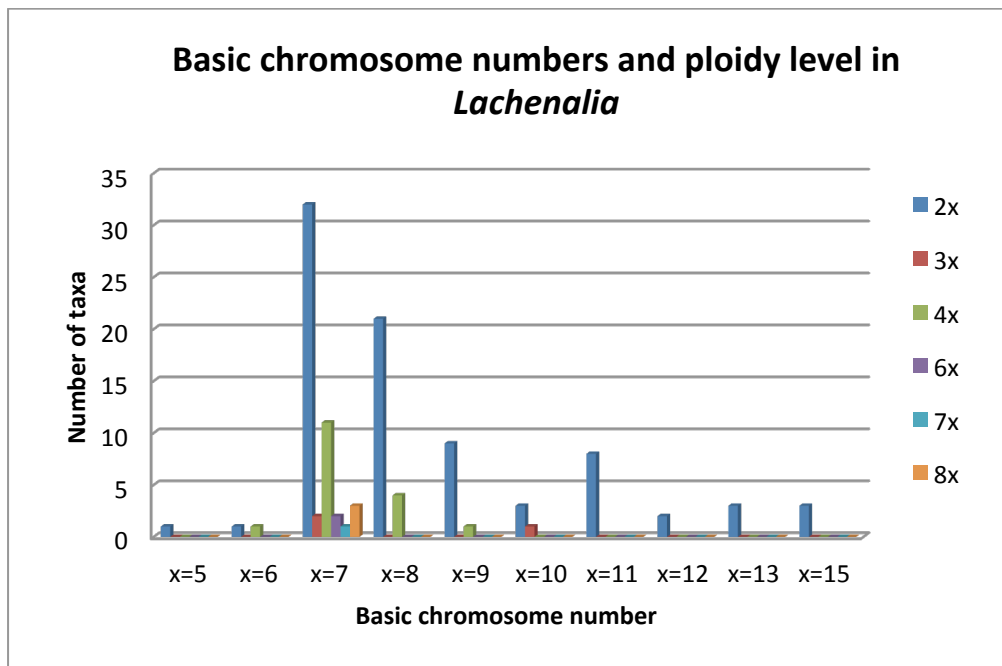


Figure 2.2: Basic chromosome numbers in the genus *Lachenalia* indicating the number of taxa for each basic number and the ploidy levels reported for these basic numbers. Taxa with $2n = 24$ were excluded. (Figure copied from Kleynhans *et al.* 2012 with permission of the authors).

However, the $x = 7$ group has been divided into four taxa due to at least four different karyotypes described in Hamatani *et al.* (2007). Hamatani *et al.* (2007) grouped together *L. longibracteata* E. Phillips, *L. orchioides*, *L. orchioides* subsp. *orchioides*, *L. orchioides* subsp. *glaucina* (Jacq.) W.F. Barker and *L. pusilla* Jacq. as taxa containing chromosomes that gradually decrease in size. *Lachenalia aloides* have two long, four medium and eight short chromosomes. Hamatani *et al.* (2007) further groups *L. algoensis* Schönland, *L. rosea* and *L. viridiflora* in group 2 as they contain karyotypes with six long and eight small chromosomes. Eight short chromosomes were observed in *L. sessiliflora* W. F. Barker and two long and 12 short chromosomes (Hamatani *et al.* 2007). Therefore it seems as if taxa with basic chromosome number $x = 7$ are under chromosomal evolution.

There is a possibility of some $x = 7$ species resulting from hybridization. Hamatani *et al.* (2007) reported *L. longituba* (A.M. van der Merwe) J.C. Manning and Goldblatt with three

long and 25 short chromosomes. Spies *et al.* (2011) concluded that the chromosomes from these are not homologous and this taxon originates through hybridization of taxa with respectively two and four long chromosomes. Spies *et al.* (2011) hypothesised that *L. longituba* originated from an aneuploidy event following hybridization or separate hybridization events involving taxa with different chromosome numbers.

$x = 8$

Basic chromosome number of $x = 8$ is the second most significant group of lachenalias with 19.5% of the taxa having this number (Fig. 2.2) (Spies *et al.* 2011). The $x = 8$ taxa are predominantly diploid with only 9.6% of the taxa at tetraploid level (Du Preez *et al.* 2002; Spies *et al.* 2011). Specimens with $2n = 24$ were excluded from this group (Spies *et al.* 2011). This is because the taxa of $2n = 24$ have only $2n = 24$ with no lower or higher ploidy levels and the probability of so many different taxa being triploid based on $x = 8$ (3.8%) seem improbable (Spies *et al.* 2011). Thus $2n = 24$ taxa is associated with $x = 6$ group rather than a triploid within $x = 8$ taxa. Even though, triploid of some true $x = 8$ species have been recorded in the literature, Spies *et al.* (2011) proposed proper meiotic analyses to provide the answer in this instance.

$x = 9$

This is the third most prominent group in *Lachenalia* with the majority of the specimens being diploids (Spies *et al.* 2011; Kleynhans *et al.* 2012). About 10.7% specimens represent these taxa (Fig. 2.2) (Spies *et al.* 2011). Yet, meiotic studies are required to solve problems regard the possibility of the presences of other basic chromosome numbers such as $x = 6$ or $x = 12$, or is it really a tetraploid (Spies *et al.* 2011). The karyotype of *L. latimerae* W.F.

Barker revealed three long and 15 short chromosomes (Hamatani *et al.* 2007). This indicates again a possible hybrid origin for a non-homologous karyotype (Spies *et al.* 2011).

$x = 11$

This chromosome number exists in 7 members of the genus and only a single tetraploid specimen (Fig. 2.2) was observed with the somatic chromosome number of $2n = 44$. The karyotypes of *L. juncifolia* Baker (Hamatani *et al.* 2007) and *L. zeyheri* (Hamatani *et al.* 2010) showed a gradual decrease in chromosome size. This karyotype has one very small chromosome that may be a B-chromosome thus explaining why somatic chromosome number for *L. zeyheri* is written as $2n = 23$ in Hamatani *et al.* (1998).

Lachenalia latimerae ($2n = 18$), *L. juncifolia* ($2n = 22$), and *L. zeyheri* ($2n = 22$) share common 5S rDNA signals and 18S rDNA near the centromeres, suggesting that these three species are related to each other although they have different chromosome numbers (Hamatani *et al.* 2010). Hamatani *et al.* (2010) observed that *L. latimerae* ($x = 9$), *L. juncifolia* and *L. zeyheri* ($x = 11$) have similar characters in the DAPI and FISH signals than that of *L. pusilla* ($x = 7$) and hypothesized that the speciation of these three species and *L. pusilla* may have occurred from a common ancestor.

Lachenalia hirta (thumb.) Thunb. and *L. unifolia* are closely related (Duncan 1988) based on their morphological similarities. The only predominant feature differing within this two species is that *L. hirta* has hairy leaves whereas *L. unifolia* has smooth leaves (Van Rooyen *et al.* 2002). *Lachenalia unifolia* ($2n = 22$) (Moffet 1936; De Wet 1957; Ornduff & Watters 1978; Crosby 1986; Johnson and Brandham 1997) and *L. hirta* ($2n = 22$) (De Wet 1957; Ornduff & Watters 1978) represents two subspecies of the same species rather than two separate species even though they are highly variable (van Rooyen *et al.* 2002, Duncan 1988).

x = 5, 6, 10, 12, 13 and 14

There are very few species reported with these chromosome numbers (Table 2.1) and further cytogenetic studies especially meiotic studies of these taxa should be done to resolve the actual chromosome numbers controversy. The majority of these species are endangered (SANBI 2009). There are only few species reported in literature with basic chromosome number of $x = 5$, e.g. *L. mutabilis* and *L. marginata* subsp. *neglecta* Schltr. ex G.D. Duncan. *Lachenalia zebrina* W.F. Barker ($2n = 30$) further supports the presence of an $x = 5$ group making *L. zebrina* a hexaploid with a basic number of $x = 5$, rather than a triploid species of $x = 10$. Johnson & Brandham (1997) suggested an alternative origin of $2n = 30$, where a dibasic origin of $x = 7$ crossed with $x = 8$ resulted in a secondary basic chromosome number of 15. Doubling of this chromosome number would results in $2n = 30$ specimens (Spies *et al.* 2011). There are only two species *L. alba* and *L. isopetala* with $2n = 40$. There is no doubt that the $x = 5$ group forms part of a very old polyploidy complex where the diploids are almost extinct (Spies *et al.* 2011). There are no karyotype information available for $x = 5$ taxa.

Table 2.1: List of chromosome numbers and number of diploid versus other ploidy levels reported for *Lachenalia* in literature. (Table copied from Spies *et al.* 2011 with permission of the authors).

| <i>x =</i> | <i>No. of all taxa</i> | <i>No. of diploid taxa</i> | <i>% of diploid taxa</i> | <i>No. of ploidy taxa</i> | <i>% of ploidy taxa</i> |
|--------------|------------------------|----------------------------|--------------------------|---------------------------|-------------------------|
| 5 | 1 | 1 | 0.75 | - | - |
| 6 | 2 | 1 | 0.75 | 1 | 0.75 |
| 7 | 51 | 32 | 24 | 19 | 14.3 |
| 8 | 25 | 21 | 15.8 | 4 | 3 |
| 9 | 10 | 9 | 6.77 | 1 | 0.75 |
| 10 | 4 | 3 | 2.26 | 1 | 0.75 |
| 11 | 8 | 8 | 6 | - | - |
| 12 | 2 | 2 | 1.5 | - | - |
| 13 | 3 | 3 | 2.26 | - | - |
| 15 | 3 | 3 | 2.26 | - | - |
| Total | 133 | 83 | 62.4 | 26 | 19.6 |

According to Spies *et al.* (2011), “ $x = 6$ group is also represented by the same diploid taxon that may form part of an aneuploid series, *L. mutabilis*. Although either aneuploidy or dysploidy may be present, the tetraploid specimens observed may represent a tribasic chromosome number ($x = 7 + 8 + 9$) or a triploid ($2n = 3x = 24$). However, *L. stayneri* W.F. Barker behaves as a diploid based on $x = 12$ or an allotetraploid with $x = 6$ ”. However there is no karyotype information available for this species to support this hypothesis.

There are uncertainties regarding the correct chromosome numbers of *L. mediana* subsp. *mediana* and *L. mediana* subsp. *rogersii* (Bak) W.F. Barker. These species have three different basic chromosome numbers observed for the two varieties, i.e. $x = 7, 9$ and 13 and $x = 13$ for *L. mediana* subsp. *mediana* and *L. mediana* subsp. *rogersii* (Bak) W.F. Barker respectively (Spies *et al.* 2011). The other $x = 13$ species reported are *Lachenalia ensifolia* (Thunb.) J. C. Manning & Goldblatt and *L. paucifolia* (W.F. Barker) J. C. Manning & Goldblatt have three long chromosomes and 23 short chromosomes (Hamatani *et al.* 2007). This is an indication of a possible hybrid origin from *Massonia depressa* Houtt. ($2n = 26$) with a karyotype consisting of four long and 22 short chromosomes (Hamatani *et al.* 2007).

The presence of binding sites for 5S rDNA and 18S rDNA on one chromosome pair, indicate that polyploidy did not contribute to the formation of the higher chromosome numbers observed in *L. paucifolia* ($2n = 26$) and in *L. longituba* ($2n = 28$) (Hamatani *et al.* 2010). However, in *L. vanzyliae* ($2n = 4x = 28$) the polyploids based on $x = 7$ clearly show multiple binding sites with two chromosome pairs providing binding sites for each probe and in *L. orchioides* ($2n = 2x = 28$), two chromosome pairs provide binding sites for each probe plus an additional pair with a binding site for 18S rDNA suggesting a segmental allopolyploid to allopolyploid origin for this species (Hamatani *et al.* 2010).

There is only two species with a basic chromosome number of $x = 14$, *Lachenalia congesta*, karyomorphological analysed for the first time by Hamatani *et al.* (2012), and *L. longituba* reclassified by Manning *et al.* (2004) from *Polyxena longituba* to *Lachenalia*. *Lachenalia longituba* and *L. congesta* both do not have clear DAPI positive bands (Hamatani *et al.* 2011; 2012) compared to other species. In addition to this characteristic *L. longituba* and *L. congesta* have two FISH signals of 5S rDNA sites on distal position of the long arms of the small chromosomes (Hamatani *et al.* 2010). Even though FISH data suggested that *L. congesta* and *L. longituba* had common chromosome feature ($2n = 28$, $x = 14$) and this is unique character among *Lachenalia*, they are separate groups as *ITS* clearly segregate *L. congesta* from *L. longituba* (Hamatani *et al.* 2012).

2.3.3. Classification of *Lachenalia* based on molecular systematic

Molecular data is more reliable in determining phylogenetic relationships than morphological, physiological data (Li 1997) and cytogenetic analysis. Even so, molecular analyses have not yet produced a well-resolved *Lachenalia* evolutionary relationship. Duncan (2012) reported that molecular phylogeny results do not agree with the morphological phylogenetic results.

Spies (2004) determined from a molecular study using the *trnL-F* region that *Lachenalia* species with $x = 7$ and $x = 8$ are very closely related. This was supported by authors such as Hamatani *et al.* (2008) using the *ITS* region. Hamatani *et al.* (2008) found that species with basic chromosome numbers of $x = 7$ and $x = 8$ are genetic homologous and form a distinct clade. However, both Hamatani *et al.* (2008) and Spies (2004) hypothesized that taxa with basic chromosome number of $x = 8$ originates from $x = 7$ or might have evolved from a common ancestor. This is further supported by phylogenetic overlapping of few species such

as *L. sessiliflora* between taxa with basic chromosome number of $x = 7$ or $x = 8$ (Hamatani *et al.* 2008). The other chromosome numbers are not monophyletic (Spies 2004). The $x = 7$ taxa is divided into two distinct clades indicating multiple phylogenetic origin or divergence of $x = 7$ soon after origin, whereas $x = 8$ has a single clade indicating a single point of origin followed by continuous speciation (Spies 2004; Hamatani *et al.* 2008). Most of these findings are supported by fluorescence *In Situ* hybridization studies on *Lachenalia* (Hamatani *et al.* 2007; 2009). Other molecular markers beside *ITS* region such as *atpB* (Pfosser *et al.* 2003), *matK*, *psbA-trnH*, *atpH-I* (Spies *et al.* 2011), *rbcl* (Chapter 5) are also having similar conclusion of *Lachenalia* evolutionary relationships. Multiple gene analysis seem not to improve species phylogenetic resolution significance and there is incongruence within plastid dataset separated, combined and *ITS* dataset (Chapter 5). There basic chromosome number $x = 7$, 8 and 11 and the former *Polyxena* species appear to form monophyletic group. Taxa with other basic chromosome numbers were spread between the $x = 7$ and $x = 8$.

2.4. Conclusion

Diversity of lachenalias with the same basic chromosome numbers (x) but different ploidy levels can be linked to environmental factors and to geographic distribution. Although there is high morphological confusion in the classification of the genus, inflorescences, leaves filament, perianth structures and *etc* variation can be integrated with molecular and cytogenetic phylogenetic observations for much robust phylogeny. Evolutional divergent characters like trichomes and maculation can answer evolutionary models, genetic drift and natural selection concepts.

Molecular systematics of nuclear or chloroplast gene regions may provide a better understanding of the phylogenetic relationships of the species than that of morphological or cytogenetic approaches. However both analyses have not yet produced well resolved phylogenies and may have to be used in conjunction with each other for better phylogeny analysis.

3. Review of factors influencing the survival of the genus *Lachenalia*

3.1. Abstract

The genus *Lachenalia* consists of approximately 133 species. These species vary at phenotypic level (i.e. morphology, size of inflorescences, flower colour, etc.) and cytogenetic composition (dysploidy, aneuploidy, polyploidy, chromosome number, etc.). The diversity of *Lachenalia* flower and floral signals (i.e. flower colour and scent) displayed by members of this genus suggest pollination syndromes such as melittophily and ornithophily. Furthermore, the variation is increased by increased ploidy level and or geographic distribution (i.e. *L. bifolia*). Another survival mechanism of *Lachenalia* is the annual bulb regaining and propagation reproduction acquired by all the members of the genus *Lachenalia*. The ability of this genus to grow in foreign habitats (invasion of *L. reflexa* in Australia as a hazardous weed), feed on low fertile soils and disease tolerance may contribute to its survival. This study reviews most of the factors influencing the performance of this genus and gives a comprehensive summary on growing lachenalias as a garden or a pot plant.

Keywords: bulb, dwarfism character, florescence, Hyacinthaceae, inflorescence, *Lachenalia*, morphology, nutrients, Ornithogalum mosaic virus, pollination, seed, syndrome temperature regime, variation.

3.2. Introduction

South African endemic plants, especially bulbous plants such as *Lachenalia*, are well adaptive and has the potential to become an invasion alien plant (*L. reflexa*). Lachenalias have been cultivated in Britain for more than two centuries (Duncan 2012). In almost all of the northern countries, *Lachenalia* is not hardy enough for outdoor cultivation and must be

kept frost-free to survive (Duncan 2012). Nonetheless, *Lachenalia* seems to possess traits for survival in harsh conditions. The survival of *Lachenalia* species might be, but not limited to factors like: (1) annual bulb character, (2) propagated vegetative by means of adventitious bud formation (Coertze *et al.* 1992), (3) rapid and profuse seedling germination in some species, (4) tolerance to diseases, viruses or pathogens attacking the family *Hyacinthaceae*, and (5) symbiosis with infectious bacteria (Pearson *et al.* 2009).

Some species have unusual dormant characteristics, for instance, *L. lutzeyeri*, *L. montana*, *L. mathewsii* and some other species remain completely dormant for years (Duncan 2012). Floral radiations within the genus promote selective pollinators inducing pollination syndromes (Duncan 2012). Seed production by sexual recombination and vegetative propagation enhance species survival. However, there is strong correlation between *Lachenalia* seed dispersal mechanisms and its restricted geographic distribution.

3.3. Climate and latitude

Lachenalia grows best at cold or moderate temperatures (du Toit 2002), but bulbs grown at low temperatures develop slightly slower than those at moderate and high temperatures (du Toit *et al.* 2002). *Lachenalia* follows a winter growth cycle similar to the Cape floristic region where the group originated (Grace & Van Staden 2003; Duncan 1992). *Lachenalia pearsonii* (Glover) W.F. Barker which inhabits Namibia is an exception and follows a summer rainfall growth cycle (Grace & Van Staden 2003). Another unique feature for this species is its dwarfish character. *Lachenalia pearsonii* might have acquired these characteristics to survive conditions worse than the point of origin (namely hot and dry summer conditions with little rain and cold dry winter).

In spite of the slower development of bulbs subjected to lower temperatures (under commercial cultivation), the quality of these bulbs are actually better than bulbs grown in moderate or high temperatures (du Toit *et al.* 2002). Louw (1991) reported that a drop in storage temperature before planting improves scape elongation and faster growth in cultivar 'Romelia'. Furthermore, bulbs initially exposed to a low temperature have high soluble sugar levels (du Toit *et al.* 2004a). Du Toit *et al.* (2004a) observed that leaves produce copious photosynthates during the growing season. This might be one of the reasons for this genus being well adapted in winter regions, as they utilise stored carbohydrates for growth and development. The starch remains unchanged until the next growing season then it is rapidly burned to support the new plant growth. Thus the bulb serve as a storehouse for the essential growth nutrients and it might be the most fundamental survival trait. Therefore, it can be assumed (based on commercial *Lachenalia* cultivation data) that cold winter temperatures initiate growth and promote best quality inflorescence development (for review, see Louw 1991; du Toit *et al.* 2002; du Toit *et al.* 2004a, 2004b).

The effect of temperature on *Lachenalias'* physiological growth is species specific (genotypic specific). For example, *L. punctata* plants do not have a low temperature requirement for elongation of scape and flowering (Kodaira & Fukai 2005). Further studies are required to find an optimum bulb storage temperature and duration to maximize the number of florets for cultivation purpose.

Under commercial production, it should be noted that failure to flower, low flowering percentage, a lack of uniformity and periodicity of flowering in many geophytes (i.e. *Lachenalia*) depend on many factors, including bulb size and periodicity of inflorescence initiation (Roh 2005). Failure of scape elongation due to death of florets (inflorescence blast)

is caused by unfavourable external factors, such as temperature during bulb production, storage, and forcing (Roh *et al.* 1998) as well as the internal source-sink relationship between inflorescence and other organs (Theron & Jacobs 1996).

3.4. Ecology and adaptative strategies

The distribution area of *Lachenalia* almost covers the western half of South Africa and southern Namibia (Duncan 1988, 1999) (Fig. 3.1). *Lachenalia* occurs in all the nine biomes and in 162 vegetation types of Southern Africa (Mucina & Rutherford 2006). One hundred and twenty-five species are endemic to the nine biomes recorded in South Africa, with some inhabiting more than one biome (Duncan 2012). Van Wyk & Smith (2001) documented the presence of lachenalias in all three primal floristic regions.

Species are morphological highly variable in the winter rainfall zone of the Western Cape, and there is a remarkable decrease in species diversity towards the summer rainfall zone of the Eastern Cape (Duncan 2012). Few species, mostly in the summer rainfall region, have a wider distribution range (Duncan 2012) and can inhabit higher attitudes and feed in different soil types (Duncan 1996, 1999). There are no records of *Lachenalia* species surviving in frost zone areas (Kapczyńska 2009). Nevertheless, Duncan (2012) documented *L. attenuata*, *L. canaliculata*, *L. congesta*, *L. isopetala* and *L. longituba* as cold tolerant species, as they can survive temperatures below -15°C for short periods.

Lachenalia patula (a xerophytic species) has small bulbs and leaves. This species is endemic to the bare Karoo desert in the Knersvlakte. It has evolved “lush” semiterete leaves and small bulbs surrounded by extremely tough cartilaginous outer scales (Duncan 2012) as adaptive features. *Lachenalia namibiensis* from south-western Namibia shares almost xero-



Figure 3.1. Geographical distribution of genus *Lachenalia*. **A**: contains high number of species per grid (≥ 20 species per grid), **B**: contains low number of species per grid (≤ 10 species per grid) (Duncan 2012).

phytic lachenalias (Duncan 2012). All the species in this genus, except *L. pearsonii*, has tough bulbs in order to survive hot and dry summer months when in a dormant state (Duncan 2012). *Lachenalia*s growing in areas with prolonged droughts and erratic rainfalls are adapted to remain dormant for years until favourable conditions return (Duncan 2012). Xerophytic species reduce water usage during growth (Duncan 2012), have shallow-seated bulbs and has horizontal spreading roots system ensuring easy access to shallow rainfalls (Duncan 2012). About 2.3% of lachenalias are pyrophytic (*L. lutzeyeri*, *L. montana* and *L. sargeantii*) and have deep-seated bulbs, i.e. can be buried up to 100 mm deep (Duncan 2012). Pyrophytic, proteranthous, synanthous and hysteranthous are other survival traits possessed. Deep seated bulbs assure that they survive wild fires. After reproduction all *Lachenalia* species lose

leaves (deciduous) which prevent the bulb from overheating underground and loss of water in hot summer temperatures.

3.5. Disease tolerance

Lachenalia is frequently infected with *Ornithogalum* mosaic virus (OMV): a filamentous virus which causes severe mosaic symptoms and flower deformation (Burger & Von Wechmar 1988; Duncan 2012). OMV in *Lachenalia* does not result in significant deterioration of plant quality like in other members of the family *Hyacinthaceae*. Pearson *et al.* (2009) describe *Ornithogalum* virus as a harmless virus which has no physiological effects in *Lachenalia*, therefore *Lachenalia* act as a host of both this virus as well as the *Veltheimia* mosaic virus. Conversely, Duncan (2012) describes deformed or stunted leaves and florescence, discolouration patterns on the leaves and flowers in the form of mosaics, mottling or streaks of light yellow or dull red as well as necrotic spots as variety symptoms caused by *Ornithogalum* mosaic virus (OMV). *Ornithogalum* virus is mostly concentrated in bulb growth points during the post dominance period (Burger & Wechmar 1988). Like in potato tubers and in lily bulbs, *Ornithogalum* mosaic virus cannot be easily detected until the flowering phase, as it is a symptomless virus (Burger & Von Wechmar 1988). Burger & Von Wechmar (1988) described two serological assays (DAS-ELISA and DOT-ELISA) to detect OMV viral infections in *Lachenalia* and *Ornithogalum*. Horst (1988) and Veclar *et al.* (1992) described several techniques to eliminate viruses from plants including OMV.

Lachenalia is often attacked by several pathogenic insects: (1) aphids are usually found on developing flower buds and cause flower deformation and premature yellowing of the leaves (Duncan 2012). Furthermore, hemipteran secretes a sugar rich honeydew deposit on the plants' surface which affects photosynthesis (Halder *et al.* 2013). (2) *rhizoglyphus*

echinopus (transparent bulb mites) commonly feed on plant bulbs (Fan & Zhang 2003), which cause lesions and craters to develop in *Lachenalia* bulbs (Journet 2003). It also causes chlorotic deformed leaves (Duncan 2012). (3) mealy bugs are insects mostly found between the bulb scales and around the leaf bases (Duncan 2012; Halder *et al.* 2013). They produce oily white deposits and excrete honeydew that coats the plant surface and these extract is responsible for distortion (Duncan 2012; Halder *et al.* 2013). (4) caterpillars, and (5) molluscs (slugs and snails) destroy the plant by feeding on the leaves and developing flowers, in lachenalias causing a massive damage and contamination by their faeces or slime (Duncan 2012). Lachenalias might not fully recover for that season if the plants are largely destroyed.

All of these pests are transmitters of fungal or viral disease. *Lachenalia* is frequently attacked by damping-off fungi that are caused by insufficient ventilated areas during autumn and winter. Damping-off fungi attack the leaves, resulting in a total collapse and disintegration of the whole plant; rust fungi are prevalent in winter under poor ventilation and insufficient light (Duncan 2012). Its symptoms are clusters of bright orange spores that can rapidly destroy entire leaves and fungal rotting of *Lachenalia* bulbs. Most species are affected by the fungi *Botrytis cinerea* and *Fusarium oxysporum*. These fungi are most common in poor drainage soils, excessive moisture and warm climate. Symptoms of *B. cinerea* and *F. oxysporum* infection are mostly slow development of leaves, turning leaves prematurely yellow or becoming distorted.

3.6. Pollination biology and seed dispersal

Lachenalia species might prevent inbreeding depression by self incompatibility. Duncan (2012) observed that flowers of most *Lachenalia* species are self incompatible. Some of the *Lachenalia* species appear to possess heterostylous flowers (flower morphs) i.e. long

stamens with a short style or short stamens with a long style. There is no studies done on the self incompatibility of *Lachenalia* species and it is suspected that gametophytic self incompatibility might be common. Nonetheless, two species were able to breakdown self incompatibility, and various other species produced quite a number of seeds in cultivation without possible evidence of cross-pollination (Duncan 2012).

Some species are adapted for myrmecochory. These species flower closely to the ground which produce many elaiosome seeds (Duncan 2012), botanically known as diaspora. Ants are attracted to the elaiosomes: a rich proteins-lipid appendage on the *Lachenalia* seeds, which they use for feeding their larvae (Duncan 1988; Bryan 1989; Vittoz & Engler 2007; Garrido *et al.* 2009; Lengyel *et al.* 2010). The diaspora are later disposed in an area where it can germinate (Van der Pijl 1982; Vittoz & Engler 2007). Elaiosomes are therefore used as a method for seed transportation (Beattie 1985).

Dispersal of most *Lachenalia* seeds is local by means of anemochory (Duncan 2012). The scape is strongly attached to the bulb after drying of the infructescence and the seeds remain adhered to the open capsules, until strong winds blow them off (Duncan 2012). However, the wind merely blows the seeds a few millimetres from the bulbs. The infructescence of some xerophytic species, such as *L. patula* and *L. xerophila*, detach from the bulb, and seeds are scattered as the wind blows (Duncan 2012), resulting in colonial *Lachenalia* populations. A limited number of species have broadly winged capsules and the fracture is strongly blown off by wind. This results in wide range spread of these species. Seeds are also dislodged from the dry capsule by raindrops and projected up to 140 mm, but Duncan (2012) documented that *Lachenalia* seeds are non-buoyant.

The discrete nature of most populations suggests that seeds are not naturally spread easily over long distances (Bettink 2009). Therefore, in areas where new plants have established 50 m to 100 m beyond main infestations, mammals or birds are thought to be dispersers of the seed.

The diversity of floral structure within *Lachenalia* species correlates to the different dispersal strategies as well as pollination (species selection by a vector) as Schiestl & Jhonstone (2013) illustrated in some other species. Duncan (2012) recorded two pollination syndromes within the genus: melittophily and ornithophily. Certain *Lachenalia* species may have acquired favourable traits (for example flower shape, size, odour and nectar composition) that may have gradually become dormant due to natural selection enforced by different pollen vectors (Faegri & Van der Pijl 1979).

Most *Lachenalia* species are pollinated by different species of solitary bees. Some tubular and especially orange-flowering species are pollinated by sunbirds (Manning *et al.* 2004; Duncan 2012); different types of beetles; by an exotic beewolf; nectar-feeding insects; hoverflies; three types of butterflies; and moth (Duncan 2012). In addition ants frequently visit *lachenalias* but play no part in pollination (Duncan 2012). However, due to convergence evolution within *Lachenalia* species, some pollinators constantly revisit *lachenalias* specific for certain pollination syndromes. Geoflorous species may possibly be pollinated by rodents but it is not certain (Duncan 2012).

Lachenalia nervosa has a scent like a mixture of cloves and carnations (Duncan & Smith 1999) as a specialized pollination feature. Strong leaves, adequate landing facilities, a tepal surface that provide adequate foothold (Faegri & Van der Pijl 1979) and bright coloured

anthers which emits a distinctive scent (Duncan 2012) are melittophilic features of bee-pollinated species. Several *Lachenalia* species are selected for bird pollination (Faegri & Van der Pijl 1979). *Lachenalia aloides*, *L. patentissima* and *L. quadricolor* have brightly coloured tubular flowers and are usually unscented and have abundant nectar and strong scapes to allow sunbirds to cling to them. All these features may attract the sunbirds for feeding, thus resulting in cross-pollination (Faegri & Van der Pijl 1979; Proctor & Yeo 1972; Duncan 2012). Nonetheless some species are also pollinated by honey bees and/or any other possible pollinators even though they are primary ornithophily (Duncan 2012), as they might possess other floral signals not specific only to sunbirds.

3.7. Vegetation propagation

Lachenalia can be propagated through offsets, bulblets (also known as bulbils), stolons, leaf cuttings, seeds and tissue culture methods (Duncan 1988; Duncan 2012). Small underground bulbs are called bulblets but if they develop at the periphery of the mother-bulb, they are called offsets (Kapczńska 2009). Some bulbous species may occasionally produce aerial bulblets (bulbils - small bulb or bulb-shaped growth arising from the leaf axil or in the place of flowers on the floral parts or on the edge of the leaf) (De Hertogh & Le Nard 1993). *Lachenalia bifolia* can produce bulblets aboveground level, and others (i.e. *L. namaquensis*) reproduce by stolons which push the bulblets away from mother-bulb (Duncan 1988; Duncan 2012). In some species, the mother bulb divides into two or more large bulblets of similar size that would flower the following season (Duncan 2012). These methods are too slow for commercial production and are species specific (Kleynhans & Hancke 2002). Propagation through leaf cutting is the best method for commercial production (Duncan 1988). Young explant tissue produce the highest number of buds and the old tissue produced

the least (Niederwieser & Vcelar 1990). Similar, leaves (leaf cutting) propagate least when the donor plant is in full flowering stage (Ndou, *et al.* 2002; Niederwieser & Ndou 2002). The proximal region of the active growing leaf should be cut before flowering begins, placed in a slightly moist medium of vermiculite and river sand or grit, with about 1 cm of the base of the material inserted into the medium (Duncan 2012). The formed bulblets should be stored in a dry place and can be planted in the next season (Du Plessis & Duncan 1989), and can flower in one year under optimum conditions (Duncan 2012).

However, vegetative propagation reduces species integrity as it prevents gene flow within the species but preserve morphological traits. Seeds collected from the wild are more likely to have species integrity of 100% (Duncan 2012). Under natural conditions, seed of most species flowers from their third season (Duncan 1988). *Lachenalia juncifolia* and *L. reflexa* can produce floescence in their second year (Duncan 2010), *L. congesta* (and perhaps some other xerophytic species) can take up to 4 to 5 years (Duncan 2012).

3.8. Recommendation for growing *Lachenalia*

Lachenalia is a strong plant adapted to harsh South-Western African conditions. Despite its hardiness, good care should be applied in commercial production or for good plant and floescence production. Kapczńska (2009) and Duncan (2012) recommended a sunny environment for good production and Kapczńska (2009) emphasised the importance of placing the plant under the shade on the hottest part of the day. Duncan (2012) documented some species that require full morning sun and afternoon shade. *Lachenalia viridiflora* tepals become insipid under insufficient light and the prostrate leaf suberect or spread in numerous species such as *L. kliprandensis* and *L. congesta* (Duncan 2012). The best time to plant *Lachenalia* bulbs is early to late autumn in the southern hemisphere. The optimal growing

temperature for lachenalias is 15 to 20°C at daytime and 8 to 10°C at night (Duncan 2012). During active growth plants requires moderate moisture and good drainage (Kapczńska 2009). Duncan (2012) recommends a very low humid and well ventilated environment. Watering should be decreased after flowering and stopped when the leaves become yellow (Bryan 1989). Prevention of excess water when the leaves turn yellow or when the plant start losing leaves might prevent the bulb from decaying. Not all *Lachenalia* species are suitable to be grown in the garden. During the summer dormant period, summer rains or garden irrigation cause lachenalias to decay as they are subjected to fungal attacks (Duncan 2012). *Lachenalia aloides*, *L. bifolia*, *L. flava*, *L. pallida* and *L. purpureo-caerulea* are examples of very few *Lachenalia* species that withstand summer rainfalls and are therefore suitable for garden cultivation.

The nutrients required by this genus depend mostly on the genotype or species as they occur in different soil habitats, and show different rates of achieving optimal nutritional status (Roodbol *et al.* 2002; Duncan 2012). Roodbol *et al.* (2002) discussed a standard nutrient regime that will benefit most *Lachenalia* genotypes. However lachenalias adapt readily to any soil types that differ from their natural habitats (Duncan 1988; Duncan 2012). In general, *Lachenalia* flourish in any soil with a good soil drainage system (Duncan 2012), mixtures containing loam, clay loam and sand beds (Perrignon 1992), or sand and bark (Duncan 2012). Duncan (2012) provided a comprehensive description on how to plant this genus in different soil types to genotypes suitable for a particular soil type. For example, species with larger bulbs must be planted up to 4 cm deep. Species with small bulbs like *Lachenalia namibiensis* should be planted not deeper than 1 cm. For most species, a depth of 2 to 3 cm is recommended (Duncan 2012).

If seeds are grown, they should be sown 3 mm to 5 mm deep depending on particular species and should be kept in moist soil in the full sun. Seeds sown in deeper pots reach maturity faster than the one sown in shallow pots (Duncan 2012). Seeds of *L. patula* take 11 days to germinate, 58 days for *L. argillicola* and 18 days for most species (Duncan 2012). Seeds must first undergo a summer dormant phase before they can germinate in the following autumn (Duncan 2012). *Lachenalia corymbosa* and *L. campanulata* are the only species known to be ready to sown directly after harvesting.

Lachenalia has low phosphorus and magnesium requirements (Roodbol & Niederwieser, 2002) but nitrogen applied in the nursery and pot plant phases positively affects the number of florets formed per inflorescence (Engelbrecht *et al.* 2008). Nevertheless fertilization has a significant effect on the growth and flowering of the plant. Fertilisers higher in potassium but low in nitrogen content can be applied once the plant has begun active growing (Duncan 2012). High nitrogen supply cause excessive soft leaf growth, reduce florescence and cause additional leaf growth (Duncan 2012). Interaction between nutrients required by plants play an important role in sustainable crop production. Neither nitrogen nor phosphorus level substantially influenced the leaf of *Lachenalia* plants (Engelbrecht *et al.* 2010). Phosphorus substantially influence bulb firmness, however firmness of 'Ronina' bulbs decreases with an increase in phosphorus (Engelbrecht *et al.* 2010). Duncan (2012) found that species grown in slightly acidic to neutral pH grows the best. Only *Lachenalia karooica* prefers moderately alkaline medium (Duncan 2012).

3.9. Conclusion

Lachenalia has been cultivated almost all around the world. Europe and East Asia make up to 75% world production South African flower heritage. In prolonged cold climates

(i.e. northern hemisphere) lachenalias must be cultivated indoor as they cannot withstand temperatures below 0°C for very long periods. All lachenalias can be successfully grown in a container (dwarfs species requires shallow containers) but only certain species are suitable to be grown in the garden (Duncan 2012). In countries with a climate similar to the south-western parts of South Africa, *Lachenalia* has the potential to flourish and replace native plants (i.e. invasion of *Lachenalia reflexa* in Australia (Kate & Grazyna 2011)).

Factors that may insure the survival of *Lachenalia* are the annual bulb production, propagated vegetation (adventitious bud) and the rapid, profuse seeding germination of some species. Other fascinating features of this genus are ecological interactions and symbiosis with animals and vegetation respectively. Even though this genus is endemic only to a fraction of Southern Africa, it possesses adaptation plasticity over biomes, soils, soil nutrients and latitude.

Chapter 4:

4. The use of GISH techniques in phylogenetic studies of *Lachenalia pusilla* group ($x = 7, 8, 9, 10, 11$ and 13).

4.1. Abstract

Lachenalia species with basic chromosome numbers of $x = 7$ and $x = 8$ are abundant and considered to be the primitive chromosome complement of the genus *Lachenalia*. Molecular systematic and cytogenetic studies provided compelling evidence that taxa with basic chromosome numbers of $x = 9, 10$ and 13 are descendant from taxa with $x = 7$ and $x = 8$.

The aim of this study was to map and determine the chromosomal origin of taxa with $x = 9, 10$ and 13 and sort out genome constitutions of their putative parents (presumably $x = 7$ and $x = 8$ taxa) using the genomic *in situ* hybridization (GISH) technique.

Several GISH protocols were used but failed to provide results. Therefore possible reasons for this failure were discussed. Furthermore, few suggestions for optimizing the GISH protocol for further studies on the genus *Lachenalia* were provided.

Keywords: denaturation, genomic *in situ* hybridization, optimizing, preparations, probe, troubleshoot

4.2. Introduction

The application of the genomic *in situ* hybridization (GISH) technique led to chromosome mapping of parental genomic DNA, analysing of chromosome organization in interphase nuclei, detection of chromosomal aberrations and alien chromatin in plants (Devi *et al.* 2004; Chester *et al.* 2010). In plant systematics, GISH has a virtual potential of solving phylogenetic relationship between species, thus the rise of new species due to ancient hybridization, polyploidy and allopolyploid speciation events can be clarified (Dive *et al.* 2004). For example, Kono *et al.* (2012) confirmed *Begonia x chungii* ($2n = 22$) originated from *B. longifolia* and *B. palmate*. The authors sorted out the genome constitutions of its putative parents using GISH. Kono *et al.* (2012) used the entire genomic DNAs of both

parental species as probes for the *B. x chungii* GISH analysis (thus, *B. longifolia* and *B. palmate* gDNA were labelled with different probes and hybridised simultaneously). *Begonia x chungii* has 22 chromosomes consisting of six chromosomes that hybridize with a probe derived from the *B. palmate* genome, six with a probe derived from the *B. longifolia* genome and the remaining ten with genomes overlapping genomes (Kono *et al.* 2012).

Another similar example of the use of GISH as a tool for analysing phylogenetic relationship is in *Lolium x Festuca* hybrids (Pasanskiene *et al.* 1998). Pasanskiene *et al.* (1998) compared GISH bands in hybrids between *Lolium multiflorum*, *L. perenne*, *Festuca glaucescens* and *F. pratensis*, and the authors found much closer homology between *L. multiflorum* and *F. pratensis* genomes than *L. multiflorum* and *F. glaucescens*. Consequently, indicating the presence of genomic exchange events between the intergeneric species *L. multiflorum* and *F. pratensis*.

Liu & Gu (2011) used GISH to study the genome organization and evolution of *Camellia reticulata* (*Theaceae* genus *Camellia*). *Camellia reticulata* has a polyploidy series varying from $2n = 2x = 30$, $2n = 4x = 60$ to $2n = 6x = 90$, with a basic chromosome number of $x = 15$ (Liu & Gu 2011). This genus also has allopolyploidy (Parks & Griffiths 1963). Liu & Gu (2011) demonstrated that, (1) the diploid species *C. reticulata*, *C. pitardii* and *C. saluenensis* are the progenitors of polyploid *C. reticulata*, (2) hybridization between diploid *C. reticulata* and diploid *C. pitardii* resulted in the allotetraploid *C. reticulata*, and (3) subsequent hybridization between allotetraploid *C. reticulata* and diploid *C. saluenensis* formed the allohexaploid *C. reticulata*.

Certainly GISH technique seems to be a perfect approach to determine phylogenetic relationships among species with bizarre numerical and structural chromosomes as in the case of *Lachenalia Jacq. ex Murray*.

This study summarises difficulties encountered in an attempt to determine genomic phylogenetic relationship and progenitor of *Lachenalia pusilla* and *Lachenalia* species using GISH technique. Secondly, gives suggestions (wherever possible) for further GISH investigation on the genus *Lachenalia*.

The aim of this study was (1) to analyse genome origin of *Lachenalia pusilla* group, (2) to determine the evolutionary relationship of the *Lachenalia pusilla* group with taxa with basic chromosome numbers of $x = 7, 8$ and 11 by using a GISH approach and lastly (3) to give suggestions on possible modification on improving GISH application on genus *Lachenalia*.

4.3. Materials and Methods

4.3.1. Plant material

Information on specimens used in this study is given in Appendix A. Plant materials were obtained from ARC-VOPI, Summerfield's Indigenous Bulbs and Seed and SANBI (Kirstenbosch Botanical Garden). Selected accessions were used for DNA extractions and chromosome preparation. All bulbs and seeds were grown in a greenhouse.

4.3.2. Chromosome preparation, staining and screening

4.3.2.1. Pre-treating and fixation

Multiple protocols were tested to obtain good chromosome spreads (Table 4.1). At the beginning (common step for all assays), actively growing root-tip meristems were harvested.

The facilitating medium (Deco swell gel) was rinsed off twice by tap water. Then the following two pre-treatment techniques were performed:

(1) Ice cold dH₂O for 24 hour (Ma *et al.* 1996; Mirzaghaderi 2010, Protocol for GISH, 2012, www.ksu.edu/wgrc). (2) ABN (1-bromonaphthalene) for 24 to 27 hours (Mirzaghaderi 2010). In some cases roots were pre-treated for a longer time.

The roots were fixed in fresh Carnoy's fixative (EtOH: chloroform: glacial acetic acid 6:3:1) for 48 hours at 4°C and stored in 70% ethanol at 4°C until ready for use. As in the case of the pre-treatment step, root tips were macerated using 3 different techniques (Table 4.1) in the attempt to improve quality of the chromosomal spreads. At first, root tips were placed at room temperature for 15 min, thereafter macerated with:

(1). Acid hydrolysis in 1N HCl: 70% EtOH was rinsed off in dH₂O, and then roots were macerated in 1N HCl for 8 min at 60°C (HCl must be at 60° C before suspending roots). HCl was then washed twice with dH₂O before softened further in dH₂O at 4°C for 24 hours.

(2). Enzymatic digestion: After thoroughly rinsing of the roots, root tips were cut and treated with a mixture of 5% (w/v) Cellulose and 1% liquid pectolyase in 0.01 M sodium citrate buffer, pH 4.6. The enzymatic digestions were run for 4 hours at 37° C.

(3). Distilled water incubation: After fixation the root tips were soaked in dH₂O for 24 to 48 hours. However, to prevent the water becoming isotonic, dH₂O was changed after every 12 hours.

4.3.2.2. Staining and chromosome spread

Meristem tissues were stained by placing the roots in an eppendorf tube containing 1% aceto-carmine for 48 to 72 hours at room temperature. To enhance the intensity of the

aceto-carmine stain, a drop of Ferric ions (Fe^{+}) was added to the eppendorf tubes before incubation (Table. 4.1). In a few cases 2% aceto-orcein or Leuco-basic fuchsin stains were used in attempt to improve chromosome visibility. Roots were placed in an eppendorf tube with orcein at room temperature until the tips became deep dark. In the case of the fuchsin, roots were stained for 2 hours at 4° C in the dark, or until the root tip turned dark red.

Table 4.1. Summary of the materials and methods used to optimize chromosome preparations for GISH. Other minor adjustments and modifications are not discussed.

| Technique | Method | General discussion |
|-------------------|------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Pre-treatment | Ice cold dH ₂ o | <ul style="list-style-type: none"> • Very difficult to stain • No mitotic chromosomes were observed • Nucleus is visible after staining with carmine + Fe^{+} |
| | 3 to 4 drops of 1-bromonapthalene | <ul style="list-style-type: none"> • Interphase nucleic were visible after carmine + Fe^{+} stain • Poor prophase chromosomes (few) |
| Macerate | Enzymatic digestion <ul style="list-style-type: none"> • (5% cellulose + 1% pectolyase) • DAPI stain | <ul style="list-style-type: none"> • All slides were negative • Enzyme at their optimal activity can improve results |
| | Acid hydrolysis <ul style="list-style-type: none"> • Carmine, orcein & Fuchsin | <ul style="list-style-type: none"> • Carmine + Fe^{+} produced better stained chromosomes or interphase nuclei • Ice cold dH₂O roots had solid nucleus • Fuchsin stained very poorly • Chromosomes cannot be identified/ differentiated |
| | dH ₂ O incubation <ul style="list-style-type: none"> • Carmine, orcein & Fuchsin | <ul style="list-style-type: none"> • Carmine + Fe^{+} produced better stained chromosomes or interphase nuclei • Ice cold dH₂O roots had solid nucleus • Fuchsin stained very poorly • Chromosomes cannot be identified/ differentiated |
| Staining | Carmine | <ul style="list-style-type: none"> • Stain was intense when Ferric ions (Fe^{+}) were added |
| | Orcein | <ul style="list-style-type: none"> • Low signal • Fe^{+} doesn't increase the intensity of the stain |
| | Fuchsin | <ul style="list-style-type: none"> • Very poor staining property |
| | DAPI (enzymatic maceration) | <ul style="list-style-type: none"> • No genomic material present to bind to |
| Chromosome spread | squeezing of meristem cells | <ul style="list-style-type: none"> • <i>Lachenalia</i> has very small chromosomes making this technique impossible |
| | Suspending enzymatic digested cells | <ul style="list-style-type: none"> • Slides were negative • Enzyme at their optimal activity can improve results |
| | Squashing plant tissue | <ul style="list-style-type: none"> • Better than any chromosome spreading technique tried in this study • Excess pressure might break the chromosomes |

Root tips were squashed in either 1% aceto-carmine or in 2% aceto-orcein. To reduce over staining of cytoplasm, root tissues were placed in 45% acetic acid at 65°C for 10 min before they were squashed in 45% acetic acid. Preparations were screened to determine the presence of metaphase and prophase chromosomes and interphase nuclei using epifluorescence microscopy (Nikon, Germany). Cover slides were removed by freeze drying in CO₂ or in liquid nitrogen. Few drops of ice cold alcohol: acetic acid fixative (3:1) was added immediately after removal of the cover slip to reduce too much presence of cytoplasm in the cell.

4.3.2.3. Genomic DNA extraction, probe labeling and blocking DNA

Total genomic DNA (gDNA) was extracted from fresh leaves using the CTAB method by Doyle & Doyle (1987) with the CTAB modification (Doyle & Doyle 1990). DNA was then diluted in 50 µl TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) and purified with phenol/chloroform. The DNA pellet was dried and resuspended in a maximum of 50 µl TE buffer. The genomic DNA was then sheared by boiling in TE buffer for 10 min to yield 300 to 600bp fragments, and labeled by nick translation with ChromaTide Fluorescein-12-dUTP (Molecular Probes C-7604) followed by the use of 0.1 M β-mercaptoethanol (Lysak *et al.* 2006).

4.3.2.4. Genomic *in situ* hybridization and detection

The GISH procedure of Bie *et al.* (2007) was followed with some modifications: The best preparations were incubated in 100% EtOH for 5 min at room temperature after removal of the coverslip. Slides were dehydrated in an EtOH series, 70%, 90% and 100% for 2 min respectively, and air dried for 48 to 72 hours. Before probe hybridization, the slides were treated with RNase A (Sigma-Aldrich) (100 ng/µl in 2x SSC) for 60 min at 37°C. The slides were then washed 3x for 5 min in 2x SSC (20x SSC: 1.3 M NaCl, 0.015M sodium citrate) at room

temperature without proteinase K digestion, and rinsed in 1x PBS (PBS: 10mM sodium phosphate, pH 7.0, 143 mM NaCl). Preparations were fixed for 10 min in 4% paraformaldehyde (prepared in 1x PBS) at room temperature, raised in 2x SSC for 5 min and 1x PBS for 2 min at room temperature and dehydrated in 70%, 95% and 100% EtOH series for 3 min each at room temperature.

Labelled DNA probes were denatured at 95°C for 10 min. The hybridization mix (50% formamide, 10% dextran sulfate, 10 ng/μl probes DNA) were heated at 75°C for 5 min, and chilled on ice for 5 min. Total gDNA of *Lachenalia isopetala* (a diploid of $x = 7$) was used as a blocking DNA. Blocking DNA was prepared by boiling 400 mg *L. isopetala* DNA (1 mg/ml) in 0.4 mol/L NaOH for 45 min and then combined in the hybridization mixture at a ratio of 1:60 (probe:blocker). Pre-treated slides were denatured on a hot plate at 80°C for 2 min in 100 μl of 70% FA/2x SSC and dehydrated in ice cold 70%, 95% and 100% EtOH series for 5 min respectively. About 5 μl of hybridization mix was added immediately onto the preparations and immediately covered with approximately 20 x 20 mm parafilm plastic and incubated overnight at 37°C in a wet chamber.

4.3.2.5. Washing and counterstain

After overnight hybridization, slides were given a stringent wash in 1x SSC. The stringency conditions allowed the target sequences of approximately 56% homology to remain hybridized (Schwarzacher & Heslop-Harrison 2000). Post-hybridization was done by washing slides in 2x SSC at 42° C for 5 min, 50% formamide in 2x SSC for 10 min at 42° C, followed by 2 x washing in 2x SSC for 5 min at 42° C and 37° C respectively. The spreads were then counterstained for 10 min with DAPI staining in glycerol solution, containing 1 μl/ml of

4', 6-diamidino-2-phenylindole (DAPI, Sigma-Aldrich) and glycerol and immediately proceeded before glycerol damaged the probe-florescence intensity.

4.3.2.6. Image capturing and processing

Chromosome preparations were analysed using a Nikon Optiphot-2 epifluorescence microscope Fuji 800 digital camera. Photographs were taken using Fuji 800 digital camera. The images were saved, trimmed and presented in a collage (Figs. 4.1).

4.4. Results and discussion

Genomes of three *Lachenalia* accessions with each of the following basic chromosome numbers of $x = 9$, and $x = 10$ were used, as well as two accessions of each of the following groups with $x = 11$, $x = 13$, $x = 7$ and $x = 8$ were compared. All the accessions studied were diploids. For each species, no chromosome numbers from the metaphases were scored. However, genomic *in situ* hybridization requires well preserved and dispersed chromosomes with no or little cell debris for effective genomic study (Ryan & Vasu 2013). Moreover, there are limited reports dedicated to elucidate the chromosome spreading dynamics for improving plant chromosome preparation (Kirov *et al.* 2014).

Modifications of previously similar protocols on *Lachenalia*, i.e. FISH protocol (Hamatani *et al.* 2009) and karyotyping protocols (Kleynhans & Spies 1999; Hamatani *et al.* 1998, 2004, 2007, 2012, Spies *et al.* 2000; Hancke *et al.* 2001) were unsuccessful for GISH analysis for this study.

In trying to solve these problems, troubleshooting and optimizing suggestions suggested by Schwarzacher & Heslop-Harrison (2000) were followed. However, due to time constrains GISH analysis was abort for this study.

In a few accessions, poor chromosome spread was observed and better genomic stain was observed with the addition of Fe^+ (Fig. 4.1). In most cases cells observed were in a non-division phase (interphase). However, preparations prepared by pre-treating root tips in 1-bromonaphthalene showed much more mitotic chromosome at metaphase (Fig. 4.1).

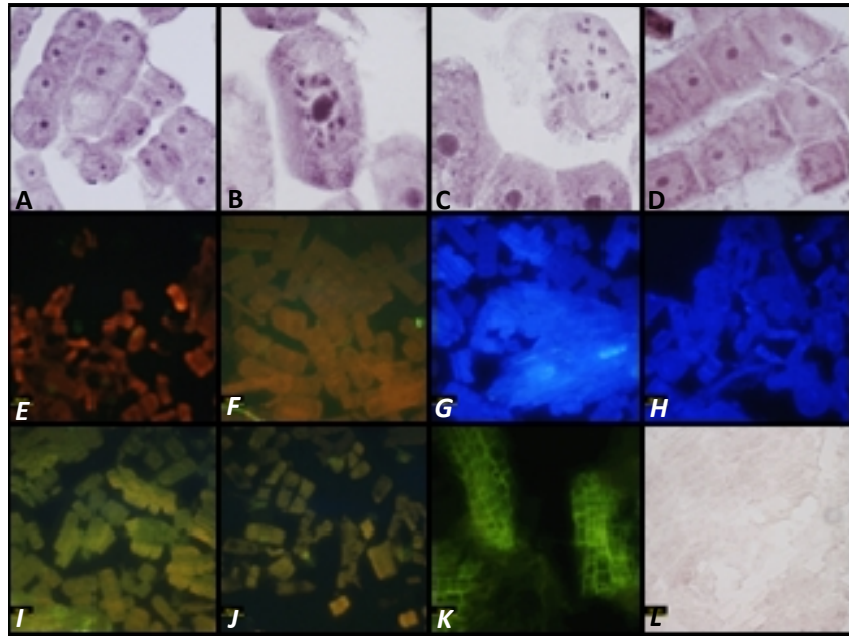


Figure 4.1: Stained genomic material of some *Lachenalia* accessions. **A-D:** For genomic screening, roots were stained in 1% Aceto-carmine + Fe^+ for 24 hours at room temperature. A, D: prophase nucleic of *L. mediana* and *L. perryae*, respectively. B, C: metaphase nuclei of *L. alba* and *L. unifolia* respectively. **E, F:** Aceto-carmine stain appears on the prophase nucleic cells of *L. unifolia* and *L. undulata*. **G, H:** *L. comptonii* and *L. alba* cells counterstained using DAPI. **I-K:** green *L. mediana*, *L. comptonii* and *L. undulata* gDNA probe appear fixed on on the cell walls of *L. unifolia*, *L. obscura* and *L. perryae* respectively. **L:** *L. punctata* cells not reacting to DAPI or gDNA probes of any species.

Genomic DNA probes were unable to penetrate and identify shared genomes due to the presences of the cell wall (Fig. 4.1). Cytoplasm, cell wall and other material present on the surface of preparations mask chromosomes and nuclei (Schwarzacher & Heslop-Harrison

2000). Treating preparations with protease increased penetration of the probe slightly (Kumar 2010). However, the presence of the cell wall and cytoplasm prevented effective chromosome denaturation; reduced penetration and effective denaturation by chemical to prevent re-annealing the chromosome DNA. Moreover, the DNA probe could not penetrate into desirable locations resulting in none probe-chromosome hybridization (Schwarzacher & Heslop-Harrison 2000). Stain present within the cells (Fig. 4.1) indicated the presence of cytoplasm and cell walls. In addition, labelled gDNA probes appeared green on the cells wall.

A high quality chromosome preparation is the most fundamental factor in obtaining good results (Schwarzacher & Heslop-Harrison 2000). Therefore, it should be acknowledged that preparations for this study were not of the best quality. Furthermore, incorrect denaturation of preparations and poor probe labelling are also common course for failure in *in situ* hybridization.

Successful labelling and fragmentation of the gDNA probe (Nick translation) was routinely checked using gel electrophoresis. However this cannot be a certain proof that the probe will bind to its complementary targets. Secondly, the direct fluorophore (fluorescein ChromaTide dUTP) method of labelled probe was selected to reduce several steps to detect the probe. Thus, minimizing the washing steps (antibody or avidin treatment) that can result in material lost. In a few preparations the probe DNA appeared to be trapped within the cells cytoplasm or cell wall. The effect of probe concentration, stringency, chromosome denaturation and blocking DNA could not be optimised for *Lachenalia* genomic *in situ* hybridization. Nonetheless, they do not have great effect on the unfeasibility of future GISH studies.

Other aspects leading to the effectiveness of target specificity were pH, salt concentration and formamide concentration in the hybridization buffer (Kumar 2010). Even though these aspects were optimized, there is however not certainty of their optimization and effect on poor probe penetration. Microscope setup, mountant and immersion oil for microscopy are also possible factors resulting in negative *in situ* hybridization (Schwarzacher & Heslop-Harrison 2000). Some slides had no or only a low mitotic index. Plant material may have consecutively washed away during the dehydration steps following slide removal (Kirov 2014). This selective loss may have been due to non-treatment of glass slides with acid (acid cleaning slides) or coating slides with poly-lysine or silane. Too high denaturation or poor preparation methods may have promoted specimen material lost (Schwarzacher & Heslop-Harrison 2000). Interestingly, no noticed differentiated plant material lost between slides denatured at 75°C to 85°C, indicating that denaturation temperature had no effect on plant material lost. Preparations were carefully handled to prevent scratches on the slides and therefore loss of material.

4.5. Conclusion

Chromosome variation exists within the genus *Lachenalia* and chromosomal evolution is evident in this unique, beautiful South African flowering genus. The success of GISH in resolving chromosome relationship of e.g. origin of triploid *Allium × cornutum* (Fredotovic *et al.* 2014) assures that an optimized GISH protocol may resolve phylogenetic chromosomal relationship in the genus *Lachenalia*. Due to sensitive nature of this technique, great precision is needed, as well as delicate treatment during the preparation of specimens. Combined cytogenetics, molecular systematics, morphological and flow cytometry should result in an improved *Lachenalia* phylogeny.

5. Evolutionary relationships between *Lachenalia* species with basic chromosome number of $x = 7, 8, 9, 10, 11$ and 13 (*Lachenalia pusilla* group) and its closest relatives.

5.1. Abstract

Lachenalia is one of the larger genera of the tribe Massonieae. The taxonomy of the genus has been studied by several authors based on basic chromosome number, morphology and systematics. In most cases, *Lachenalia* phylogeny construction included only a limited number of species from the genus, or only limited to certain taxa within the genus (i.e. $x = 7$ and $x = 8$). Species with basic chromosome number $x = 7, 8, 11$ including the subgenus *Polyxena* (the former genus *Polyxena*) are better resolved at molecular level, while other basic chromosome number species are not always resolved.

Combined molecular analysis (nucleus-plastid dataset) was used to investigate the phylogenetic relationship of *Lachenalia pusilla* group ($x = 9, 10, 11$ and 13) and *Lachenalia* species with basic chromosome number $x = 7, 8$ and the subgenus *Polyxena*. Phylogenetic and hybridization networks were performed to identify possible hybridization events.

Separate dataset analysis provided far less resolution than combined dataset with general low level of statistical support and with some incongruence. The results are largely similar to those of previous studies, conforming monophyly of $x = 7, 8, 11$ and the subgenus *Polyxena*. *Lachenalia pusilla* group and few species with $x = 7, 8$ and 11 formed paraphyletic clades in between monophyletic groups of $x = 7, 8$ and 11 . Phylogenetic networks suggested that, closely related *Massonia* species may have hybridized with *Lachenalia* species, thus introducing other chromosome numbers and the subgenus *Polyxena* may be the result of *Massonia-Lachenalia* hybridization.

Keywords: *Lachenalia*, *Massonia*, *Polyxena*, chromosome number, $x = 7$, $x = 8$, taxa, group, hybridization

5.2. Introduction

The tribe Massonieae Baker (Box 5.1) comprise of very advanced and highly diverse genera within the subfamily Hyacinthoideae. Numerous phylogenetic techniques have been used to solve the phylogenetic relationship of tribe Massonieae. However, there is still not enough phylogenetic analysis done to infer relationships among genera of the Massonieae. In addition, inventory of the species and species delimitation within tribe Massonieae and subfamily Hyacinthoideae is not yet complete. There is frequently disputes regarding the delimitation of genera in the subfamily Hyacinthoideae resulting in species from different genera being treated under the same species name. Also, taxa belonging to the same species can be treated as different species belonging to different genera (Pfosser *et al.* 2003).

These errors result in a highly polymorphic and heterogeneous genera (Pfosser *et al.* 2003) (i.e. genus *Lachenalia*). There is some taxonomic confusion within the genus *Lachenalia* due to high morphological and chromosome variation. Molecular systematic analysis based on DNA sequences suggested that the taxa with basic chromosome number of $x = 7$ and the taxa with $x = 8$ are closely related (Spies 2004; Hamatani *et al.* 2008). Groups with basic chromosome numbers of $x = 7$ and $x = 8$ are the major groups within *Lachenalia* genus. The high chromosome variation ($x = 5, 6, 7, 8, 9, 10, 11, 12, 13, 14$ and 15 , thus subgenus *Polyxena* included) within the genus and within species (i.e. *L. mutabilis*, $x = 5, 6$ and 7) indicates the possibility of speciation due to hybridization (Johnson & Brandham 1997).

Hamatani *et al.* (2008) found a high correlation between *Lachenalia* taxa with basic chromosome numbers of $x = 7$ and $x = 8$ in comparison to those with other basic chromosome numbers, based on molecular analysis of the internal transcribed spacer (*ITS*) region. In

Box 5.1: Delimitation of the tribe Massonieae Baker

Introduction

The tribe Massonieae (230 species, 19 genera) belongs to the subfamily Hyacinthoideae (Pfosser *et al.* 2003). The families Hyacinthaceae and Amaryllidaceae share very rich flora diversity.

Phylogeny in the tribe Massonieae and its chromosome evolution

Massonieae belongs to the monophyletic family Hyacinthaceae within the order Asparagales. Its close relatives are the family Themidaceae and *Aphyllanthes monspeliensis* (Pfosser and Speta 1999, Chase *et al.* 2000). Based on molecular, morphological, karyological and chemotaxonomical data the Hyacinthaceae is divided into four subfamilies, Oziroeoideae, Urigineoideae, Ornithogaloideae and Hyacinthoideae (Pfosser & Speta 1999, 2001). Massonieae is further divided into subtribe Massoniinae and Lachenaliinae (Müller-Doblies & Müller-Doblies 1997). Molecular phylogenetic analysis (Pfosser *et al.* 2003), suggested monophyletic groupings of all the genera of the subfamily Hyacinthoideae (only when the genus *Pseudoprospere* is excluded), which are divided into the tribes Massonieae and Hyacintheae (Speta 1998). Hyacintheae is a sister tribe to Massonieae (Pfosser *et al.* 2003; Wetschnig & Pfosser 2003). Similar, seed morphology permit discrimination of most genera within the tribe Massonieae (Pfosser *et al.* 2003).

However, there is a taxonomic confusion within the subfamily Hyacinthoideae as species belonging to different genera are treated under the same species name (Pfosser *et al.* 2003). For example, this error resulted in a heterogeneous genus like the genus *Scilla*, a highly polymorphic genus (Pfosser *et al.* 2003).

The genus *Massonia* is sister to the genus *Lachenalia* and *Veltheimia* is a sister genus to the *Lachenalia* and *Massonia* clade (Pfosser *et al.* 2003). The *Lachenalia* subgenus *Polyxena* may have evolved through hybridization speciation.

According to Goldblatt *et al.* (2012) the ancestral basic chromosome number for Hyacinthoideae is $x = 10$. The ancestral basic chromosome number of the tribe Massonieae is $x = 10$ (Goldblatt *et al.* 2012). The basal tribe Pseudoprosperaeae have $x = 9$ (Goldblatt *et al.* 2012). The tribe Massonieae appears to have evolved due to polyploidization events. *Merwillia*, *Schizocarphus*, *Veltheimia* have $x = 20$ (Goldblatt *et al.* 2012). The highest polypoidy genus is *Eucomis*, $x = 15$ and the genus *Lachenalia*, the most advanced species have an ancestral basic chromosome number of $x = 9$ and 10 (Goldblatt *et al.* 2012).

Distribution and adaptation

Massonieae is distributed south of the sub-Saharan Africa, Arabian Peninsula, Madagascar and India (Pfosser *et al.* 2003). According biogeographical analysis sub-Saharan Africa (place of origin) is the primary centre of diversity and radiation, followed by dispersal to the Mediterranean region (Ali *et al.* 2012). Advanced Massonieae genera are distributed in the winter rainfall area of western South Africa (Pfosser *et al.* 2003). Basal genera are better adapted to the tropical summer growing areas (Pfosser *et al.* 2003). Various species within the tribe Massonieae occur in the winter rainfall regions (*i.e.* taxa from the genus *Neobakeria*) (Müller-Doblies & Müller-Doblies 1997). *Lachenalia lutzeyeri*, *L. montana* and *L. sargeantii* are pyrophytic species (Duncan 2012).

Pollination/flower structure

There are several pollination syndromes within the family Hyacinthaceae. *Massonia depressa* (Johnson *et al.* 2001) and *Whiteheadia bifolia* (Wester 2011) show nonflying mammals floral syndrome. Mellitophily, ornithophily and phalaenophily pollination syndromes have been observed in the genus *Lachenalia* (Duncan 2012) and in *Dipcadi* (Manning *et al.* 2012) respectively..

addition, Spies (2004) used the *trnL-F* region and observed correlations among species with similar basic chromosome numbers. Additionally, there is a significant correlation between the cross-ability of *Lachenalia* species, their basic chromosome numbers (Kleynhans *et al.* 2012) and their phylogenetic relationship (Kleynhans *et al.* 2009b).

It was suggested that the *Lachenalia* species with basic chromosome number of $x = 7$ and $x = 8$ evolved from a common ancestor, while the other species of the genus with basic chromosome numbers other than $x = 7$ or $x = 8$ had evolved from a different ancestral origin (Spies 2004; Hamatani *et al.* 2008). Moreover, molecular phylogenetic studies of the *trnL-F* region (Spies 2004), *ITS* region (Hamatani *et al.* 2008) and molecular karyotyping analysis (Hamatani *et al.* 2009) placed the taxa with basic chromosome number of $x = 7$ or $x = 8$ in basal distinct clades from taxa with other basic chromosome numbers. Spies (2004) and Hamatani *et al.* (2008) found that taxa with a basic chromosome number of $x = 8$ form a small distinct clade from taxa of $x = 7$. Hamatani (2009) found similar conclusions to those of molecular phylogenetic conclusions using cytogenetics and DAPI-staining.

The relationships of $x = 9, 10, 11$ and 13 with the $x = 7$ and $= 8$ groups and with *Massonia* (outgroup) have not been established up to date. Therefore, the aim of this study is to determine the phylogenetic relationship of *Lachenalia pusilla* group ($x = 9, 10, 11$ and 13) with $x = 7$ and 8 *Lachenalia* species. To determine if the outgroup genera are involved in *Lachenalia* speciation.

5.3. Materials and methods

5.3.1. Plant material

Plants that were used in this study were collected at different locations and were supplied by ARC-VOPI (by Mrs Riana Kleynhans), Summerfield's Indigenous Bulbs and Seed (by

Mr G. Summerfield) and SANBI, Kirstenbosch Botanical Garden (by Mr G. Duncan) (Appendix A). All seeds and bulbs were grown in a greenhouse.

5.3.2. DNA extraction and PCR amplification

Total genomic DNA were extracted from young leaves of *Lachenalia* specimens using the CTAB method by Doyle & Doyle (1987), because of high amount of polysaccharides present, the CTAB concentration was adapted to 3% (Doyle & Doyle 1990). The DNA pellet was then diluted into 50 µl TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0). An additional phenol/chloroform purification step was performed to remove proteins and potentially interfering secondary metabolites as follows: 150 µl of TE was added to bring the initial volume to 200 µl. Equal volume of phenol was then added, vortex gently and spin for 2 min at 13 000 rpm at room temperature. It was followed by addition of 100 µl of Sevag (chloroform-isoamyl alcohol 96:4), centrifugation at 13 000 rpm at room temperature of the collected aqueous steps (repeated twice). DNA was precipitated by incubating with 2/3 100% EtOH-3M NaOAc (25:1) at -20°C for 24 hours, then centrifuged for 2 min at 13 000 rpm at 4°C. The DNA pellet was washed with 70% EtOH, centrifuged at 13 000 rpm for 5 min, air dried in the dark and finally re-suspend in 50 µl TE.

Three chloroplast regions, *psbA-trnH*, *trnL-F*, *rbcL* and one nuclear region *ITS* (Table 5.1) were amplified using either direct PCR (Finnzymes Direct PCR kit) or iProof™ High-Fidelity DNA Polymerase (Bio-Rad Laboratories, CA) according to the manufacturer's protocol. iProof GC buffer (Bio-Rad) was used when nuclear phylogenetic gene (*ITS*) amplification with HF buffer (Bio-Rad) did not provide satisfactory results. A final concentration of 5% DMSO was added in 20 µl PCR reaction (except for the *rbcL* region). For the direct PCR, DNA amplicons

were amplified immediately after harvesting of leaves or receiving of the seeds. In few cases direct PCR was done on dry leaves or on fresh leaves stored at -20°C.

The PCR amplifications were performed using a G storm PCR system 9700 (Perkin-Elmer) with the following thermal cycle conditions unless stated otherwise: DNA was initially denatured at 98°C for 5 min followed by 35 cycles at 98°C for 10 seconds, primer annealing at 55°C for 10 seconds excluding the nuclear region (*ITS*), and elongation at 72°C for 30 seconds with a final 1 min elongation at 72°C. Reaction conditions for the whole *ITS* region (*ITS1*, 5.8S rDNA, *ITS2*), were as follows, one cycle at 98°C for 5 min; 35 cycles at 98°C for 10 seconds, 52°C for 30 seconds; 72°C for 30 min, and one cycle at 72°C for 7 min. The PCR fragment lengths were determined on a 1% agarose gel (see Fig. 5.1)

5.3.3. DNA sequencing

PCR products were sequenced directly after 1:5 diluting with dH₂O. Amplified regions were sequenced in both directions with an automated sequencer 3730 Genetic Analyser Applied Biosystems v1.1/3.1 Cycle Sequencing Kit, according to the protocol provided with few modifications. Briefly: the component and volumes for the sequencing PCR reactions were: 1 µl of 5x sequencing buffer, 0.5 µl premix (Applied Biosystems, Life technologies), 3 µl of 10 µM primer, 3 µl dH₂O, 5% DMSO and 2 µl PCR product were used. For the internal transcribed spacer (*ITS1*, 5.8S rDNA, *ITS2*) the premix was adjusted to 1 µl due to high GC content. The final PCR reaction was set up to 10 µl. The same primers were used for sequencing except for *trnL-F*, where nested *trnL-F* primers (Table 5.1) were used. Cycle sequencing steps were as follows: initial denaturation at 96°C for 1 min, following by 25x cycles of 96°C for 10 seconds (with a ramp speed of 3°Cs⁻¹), 48°C for 15 seconds, 60°C for 4 min; 72°C for 1 min.

Table 5.1: Nucleotide sequence data of the universal primers used for the amplification of *ITS2*, *rbcl* and *psbA-trnH* and the *trnL-F* regions.

| Region | primer name | Primer sequence 5'– 3' | Taxonomic group | References |
|--------------------|-----------------|----------------------------|-----------------|-----------------------------------|
| <i>ITS</i> | <i>ITS 5</i> | TCGTAACAAGGTTTCCGTAGGTG | Fungi | White <i>et al.</i> 1990 |
| | <i>ITS 4</i> | TCCTCCGCTTATTGATATGC | Fungi | White <i>et al.</i> 1990 |
| <i>psbA-trnH</i> | <i>psbA3_f</i> | GTTATGCATGAACGTAATGCTC | Vascular plants | Tate & Simpson 2003 |
| | <i>trnHf_05</i> | CGCGCATGGTGGATTACAATCC | Vascular plants | Sang <i>et al.</i> 1997 |
| <i>trnL-F</i> | <i>trnL-F c</i> | CGAAATCGGTAGACGCTACG | Vascular plants | Pfosser & Speta 1999 |
| | <i>trnL-F f</i> | ATTTGAACTGGTGACACGAG | Vascular plants | Pfosser & Speta 1999 |
| Sequencing primers | PS1 | CTACGGACTTAATTGGATTGAGC | Vascular plants | Pfosser & Speta 1999 [#] |
| | PS4 | AGGATTTTCAGTCTCTGCTC | Vascular plants | Pfosser & Speta 1999 [#] |
| <i>rbcl</i> | <i>rbcl-a-F</i> | ATGTCACCACAAACAGAGACTAAAGC | Vascular plants | Levin 2003 |
| | <i>rbcl-a-R</i> | GTAATAATCAAGTCCACCRCG | Vascular plants | Kress & Erickson 2007 |

[#] *trnL-F* nested primers

Cleanup

Extension products were adjusted to 20 µl by adding 10 µl of dH₂O, precipitated with 5 µl of 125 mM EDTA, and 60 µl of absolute EtOH for 15 min and centrifuged for 15 min at 20 000 g. For pellet purification, 60 µl of 70% EtOH was added after removal of supernatant, centrifuged for 5 min at 20 000 g at 4°C. Tubes were then dried at 55°C for 5 min.

5.3.4. Sequence alignment and data analysis

The data sequences of *rbcl*, *trnL-F*, *psbA-trnH* and the *ITS* region were aligned using Molecular Evolutionary Genetics Analysis (MEGA) 6.0.1 (www.megasoftware.net), followed by manual adjustment and trimming at the ambiguous ends using Geneious 6.1.5 (Biomatters, Ltd., <http://www.geneious.com>) using the default alignment parameters. The post-trimmed lengths were at least 80% of the original read length. Sequences which covered more than 70% overlap between the forward and reverse sequences were considered. No data sequence was submitted online (NCBI).

Additional sequence data of *trnL-F*, *rbcl*, *ITS2* and *psbA-trnH* were obtained from Spies (2004), and from Genbank (<http://www.ncbi.nlm.nih.gov/Genbank>) (Table 5.2). Firstly, they

were converted into Nexus format, then into Mega format using MEGA 6.0.1. After readjusting and editing the data was aligned and assembled in Geneious 6.1.5. Finally all the datasets were aligned and adjusted by eye on Geneious 6.1.5.

Table 5.2: List of additional accession retrieved directly from GenBank with their GenBank ID numbers.

| Species | Accession Name |
|----------------------------------------------|-----------------------|
| ITS | |
| <i>Lachenalia algoensis</i> | AB304971.1 |
| <i>Lachenalia flava</i> | AB304975.1 |
| <i>Lachenalia arbuthnotiae</i> | AB304978.1 |
| <i>Lachenalia bachmannii</i> | AB304979.1 |
| <i>Lachenalia carnosa</i> | AB439269.1 |
| <i>Lachenalia congesta</i> | AB625560.1 |
| <i>Lachenalia hirta</i> | AB439272.1 |
| <i>Lachenalia unicolor</i> | AB439277.1 |
| <i>Lachenalia juncifolia</i> | AB304983.1 |
| <i>Lachenalia latifolia</i> | AB304984.1 |
| <i>Lachenalia latimerae</i> | AB304985.1 |
| <i>Lachenalia liliflora</i> | AB304986.1 |
| <i>Lachenalia longituba</i> | AB305011.1 |
| <i>Lachenalia orchioides var. orchioides</i> | AB304992.1 |
| <i>Lachenalia paucifolia</i> | AB305012.1 |
| <i>Lachenalia peersii</i> | AB304994.1 |
| <i>Lachenalia pusilla</i> | AB3044996.1 |
| <i>Lachenalia zeyheri</i> | AB305004.1 |
| <i>Lachenalia variegata</i> | AB305005.1 |

| | |
|-------------------------------|------------|
| <i>Lachenalia viridiflora</i> | AB305006.1 |
| <i>Massonia depressa</i> | AB305007.1 |
| <i>rbcL</i> | |
| <i>Lachenalia orchioides</i> | AM234989.1 |
| <i>trnL-F</i> | |
| <i>Lachenalia sp</i> | AJ507984.1 |
| <i>Massonia depressa</i> | AJ507980.1 |
| <i>Veltheimia bracteata</i> | JX090593.1 |
| <i>Gloriosa superba</i> | EU044686.1 |
| <i>psbA-trnH</i> | |
| <i>Gloriosa superba</i> | DQ088321.1 |

5.3.5. Phylogenetic analysis

Phylogenetic analyses were performed using three character-based methods: maximum parsimony (MP), maximum likelihood (ML) and Bayesian Inference (BI) and one distance method: Neighbour Joining (NJ). BI analysis was performed using MrBayes v.3.1.2 (Ronquist & Huelsenbeck 2003) plugin for Geneious. ML and MP were carried out using MEGA 6.0.1 (Tamura *et al.* 2013). NJ analyses were performed using Geneious Builder. All the characters were equally weighed and unordered. A sequence of *Gloriosa superba* was included as outgroup.

Phylogenetic reconstructions were done on the combined and separate datasets of the *psbA-trnH*, *trnL-F*, *rbcL* and *ITS* gene regions. Indels were coded using DnaSP 5.10.1 (Rozas 2009). The indel option was selected to be model 3: tetra-allelic. Sites with overlapping indels were excluded as well as site with missing data was excluded. Non-tree evolutionary relationships (phylogenetic networks) were determined using SplitsTree 4.13.1, NJ tree and

NeighbortNet networks (Huson & Bryant 2006). Incongruence was determined by eye comparison and by Dendroscope 3.2.9.

5.4. Results

DNA was extracted from 125 specimens representing 97 ingroup species and 10 outgroup species for all the multiple phylogenetic gene markers. All the taxa were represented by one specimen except for *L. unifolia*, *L. mediana*, *L. mutabilis*, *L. bifolia*, *L. punctata*, *L. undulata*, *L. isopetala*, and *L. juncifolia*. One hundred and eleven specimens were successfully amplified for the *rbcl* gene marker, 109 for *trnL-F*, 105 for *ITS* and 97 for *psbA-trnH* gene marker. All the successful amplified gene regions were sequenced and about 101 for *rbcl*, 99 for *trnL-F*, 97 for *ITS* and 93 for *psbA-trnH* provided good results for phylogenetic analysis.

Estimated lengths for all the phylogenetic markers included in this study (*trnL-F*, *ITS*, *psbA-trnH* and *rbcl*) are shown in Figure 5.3. Although multiple attempts were made not all accessions were sequenced successful within the genus *Lachenalia*. Attempts to obtain sequences from difficult accessions were unsuccessful due to (1) poor amplification of older samples (direct PCR), (2) poor DNA quality and or (3) complexity of the species DNA composition. Appendix B lists all the accessions and genes where sequencing was unsuccessful. Due to time constraint DNA from the accessions listed in Appendix B was not cloned in order to increase chances of obtaining their DNA sequences. Few sequences were obtained from NCBI gene bank and were included in this study (Table 5.2). Data characteristic of the single sequences plastid analysis, combined dataset of plastid (*rbcl* + *psbA-trnH* + *trnL-F*), nucleus analysis (*ITS*) and total combined dataset (nucleus-plastid) are summarized in Table 5.3.

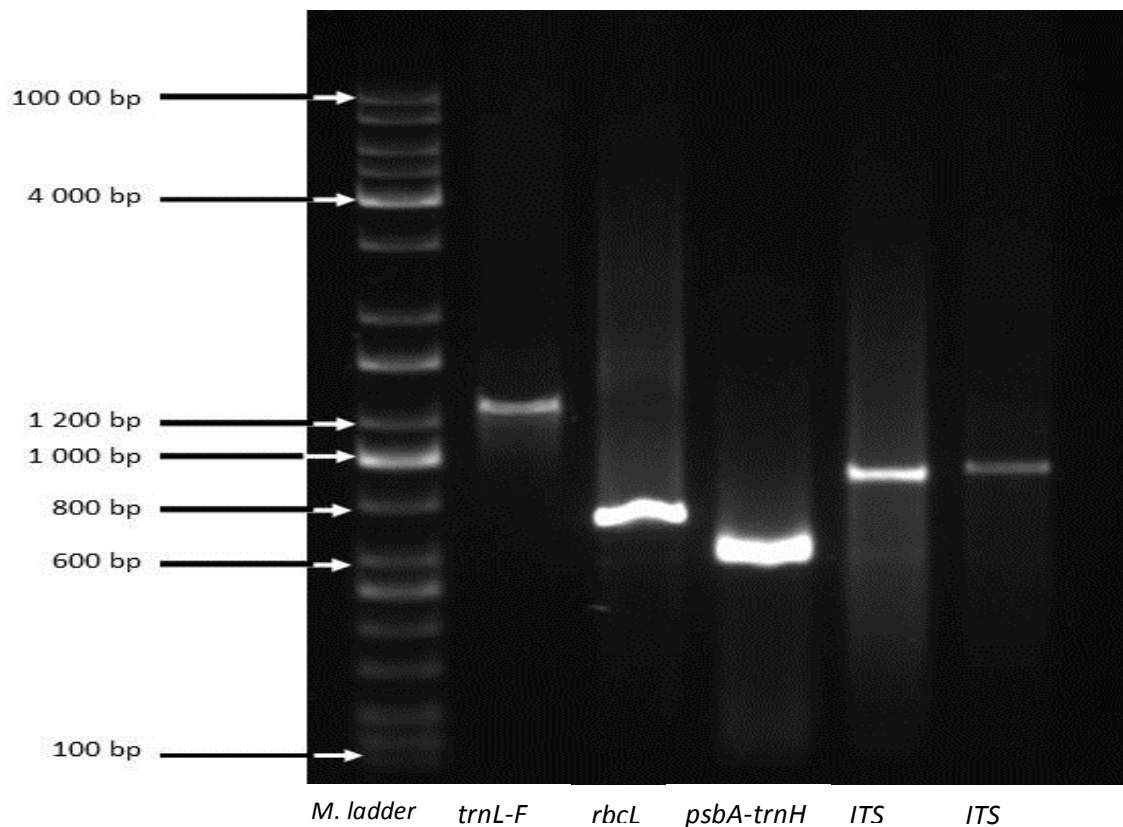


Figure 5.1. Estimated lengths of four phylogenetic genes (*trnL-F*, *ITS*, *psbA-trnH* and *rbcL*) ran for 1:30 min on 1% agarose gel. Each lane contains 6 μ l of PCR products except for the faint bands, where 4 μ l of PCR product was loaded. Molecular ladder was from KAPA Universal Ladder (KAPA Biosystems).

5.4.1. Phylogenetic analysis of individual datasets

5.4.1.1 *rbcL* dataset

The length of the unaligned *rbcL* sequences varied from 561 bp of *Eucomis pole-evansii* to 852 bp of *Lachenalia alba* (Table 5.3). The total length of aligned and trimmed *rbcL* sequences was 535 bp. The *rbcL* and *trnL-F* datasets have higher informative variation than other phylogenetic regions. However, when accessions *E. autumnalis*, *E. vandermerwei* and *Gloriosa superba* were excluded from the *rbcL* dataset, *rbcL* showed significant less informative variation (127 *rbcL* informative variation decreases to 56 when *E. autumnalis*, *E. vandermerwei* and *G. superba* were excluded). ML and BI had very low bootstrap values and a

less resolved phylogenetic relationship of the *Lachenalia pusilla* group ($x = 9, 10, 13$ plus $x = 7$ and 8 species).

5.4.1.2. *psbA-trnH* dataset

Among the datasets analysed, *psbA-trnH* had a relatively constant length (forward and reverse), making the alignment easy and unambiguous. The lengths of unaligned sequences ranges from 531 bp of ingroup species to 703 bp in the outgroup species (Table 5.3). All the accessions showed almost constant conserved variation and only 3 outgroup accessions (*Veltheimia depressa*, *Eucomis autumnalis* and *Gloriosa superba*) were very variable. In addition these three accessions were responsible for 40% of the variable characters. In comparison the *psbA-trnH* dataset had the lowest success rate of amplified DNA. Additionally, the *psbA-trnH* dataset had the lowest gap index (54) of all the phylogenetic genes.

Table 5.3. Summarised dataset Matrices used for phylogenetic inference. Combined dataset consists of concatenated *rbcL* + *trnH-psbA* + *trnL-F* (plastid) and plastid + *ITS* sequences. Sequences of combined datasets included coded indels.

| | <i>rbcL</i> | <i>psbA-trnH</i> | <i>trnL-F</i> | <i>ITS</i> | Combined data set | |
|-----------------------------|-------------|------------------|---------------|------------|-------------------|---------------|
| | | | | | Plastid | Plastid + ITS |
| Number of accessions | 104 | 84 | 94 | 89 | 231 | 275 |
| Range of length | 561-852 | 531-703 | 582-685 | 609-809 | 531-852 | 531-852 |
| Aligned length | 535 | 584 | 675 | 179* | 1867 | 2080 |
| Variable characters (%) | 220 | 78 | 324 | 59 | 324 | 1043 |
| Conserved sites | 292 | 76 | 298 | 114 | 298 | 858 |
| Parsimony informative sites | 127 | 28 | 223 | 21 | 223 | 403 |
| Indels coded | 92 | 54 | 88 | 38 | 88 | 101 |
| Best-fitting model | K2 | TN93+I | T92+G | T92+G | HKY+G | TN93+G |

*Whole *ITS* region (*ITS1*, 5.8S rDNA, *ITS2*) amplified using *ITS4* and *ITS5* was restricted only to *ITS2* region as most accessions showed better *ITS2* region sequences within the whole *ITS* region.

5.4.1.3. *trnL-F* dataset

The final length of the unaligned and untrimmed *trnL-F* sequences ranged from 501 bp to 661 bp. Sequencing of the *trnL-F* spacer was done using nested primers (*PS3* and *PS4*) with

annealing sites situated inside the *trnL-Fc* and *trnL-Ff* primer annealing sites. *trnL-F* had the highest number of variable characters and parsimony informative sites of 268 and 130 respectively. The evolutionary model was determined by MEGA 6.1 to be T92+G. The *trnL-F* dataset, had the highest parsimony informative sites of all the DNA regions included in this study.

5.4.1.4. ITS dataset

The summary statistics for the *ITS* dataset is presented in Table 5.3. Untrimmed *ITS* sequences ranged from 609 bp to 809 bp. However, the final *ITS* dataset was restricted only to *ITS2* region, which is about 179 bp (Table 5.3). The restricted *ITS2* dataset had a low number of variable characters of 59. In respect to the small *ITS* size the low number of variable characters could have been expected. The evolutionary model was tested to be T92 + G (Table 5.3).

5.4.2. Phylogenetic analysis of combined dataset

5.4.2.1. Combined plastid dataset

The combined plastid dataset included a total of 104 taxa, 1876 unambiguously aligned positions with 223 parsimony-informative characters. Species phylogenetic relationship was determined on the combined plastid dataset + indels. The combined plastid dataset + coded indels, yielded the most parsimonious tree with better supported trees than the individual plastid datasets or the non-coded indels combined plastid dataset. MP and ML of the combined dataset + indels showed tree topologies similar to the BI phylogeny (results not shown) and differed only in weakly supported clades. Additionally, the unrooted NJ tree and NeighbortNet networks (results not shown) show a similar species phylogenetic history to the MP and ML tree.

However, there is some incongruence between the plastids datasets. The most common congruence test, the Incongruence Length Difference (ILD) test also known as partition-homogeneity test (Ferris *et al.* 1994), implemented in PAUP and TNT were not executed, due to the lack of a license key (PAUP) or unsupported program format (TNT). However, ILD test can results in false positives for conflict (Sunllivan 1996; Reeves *et al.* 2001; Yoder *et al.* 2001; Pirie *et al.* 2009). Therefore, individual and combined dataset topologies were compared by eye (Pirie *et al.* 2009). None of the incongruence clades showed significances BS support as the majority of the nodes were all less than $\leq 50\%$ and therefore all the plastids spacer were combined.

5.4.2.2. Combined plastid and nuclear (nuclear-plastid) dataset

Congruence analysis (by eye) and Tanglegram construction (conflict test) suggested some incongruence between the plastid and the *ITS* datasets (Fig. 5.2). Nonetheless, the *ITS* dataset was combined with plastid dataset due to variable reliability of incongruence test (Sunllivan 1996; Reeves *et al.* 2001; Yoder *et al.* 2001; Pirie *et al.* 2009). After all, none of the strongly supported clades were mutually incompatible ($x = 7, 8$ and 11). The combined nuclear-plastid dataset consists of 2080 bp with 403 parsimony-informative sites. The combined nuclear-plastid dataset was similar to the tree topology generated from the combined plastid dataset but had relatively better resolution. Therefore, selected combined plastid-nucleus dataset (indels coded) were chosen to represent results and conclusion of this study. The evolutionary model was determined to be TN93+G (Table 5.3). BI (results not showed), ML (Fig. 5.3) and MP (Fig. 5.4) showed similar species phylogenetically relationships differ slightly on tree topologies. Combined plastid + nucleus ML tree was selected to represent explicate the outcomes of this paper. There is a correlation between groups in the

treelike phylogenetic species relationship, ML and the NJ networks phylogenetic relationships (Fig 5.5).

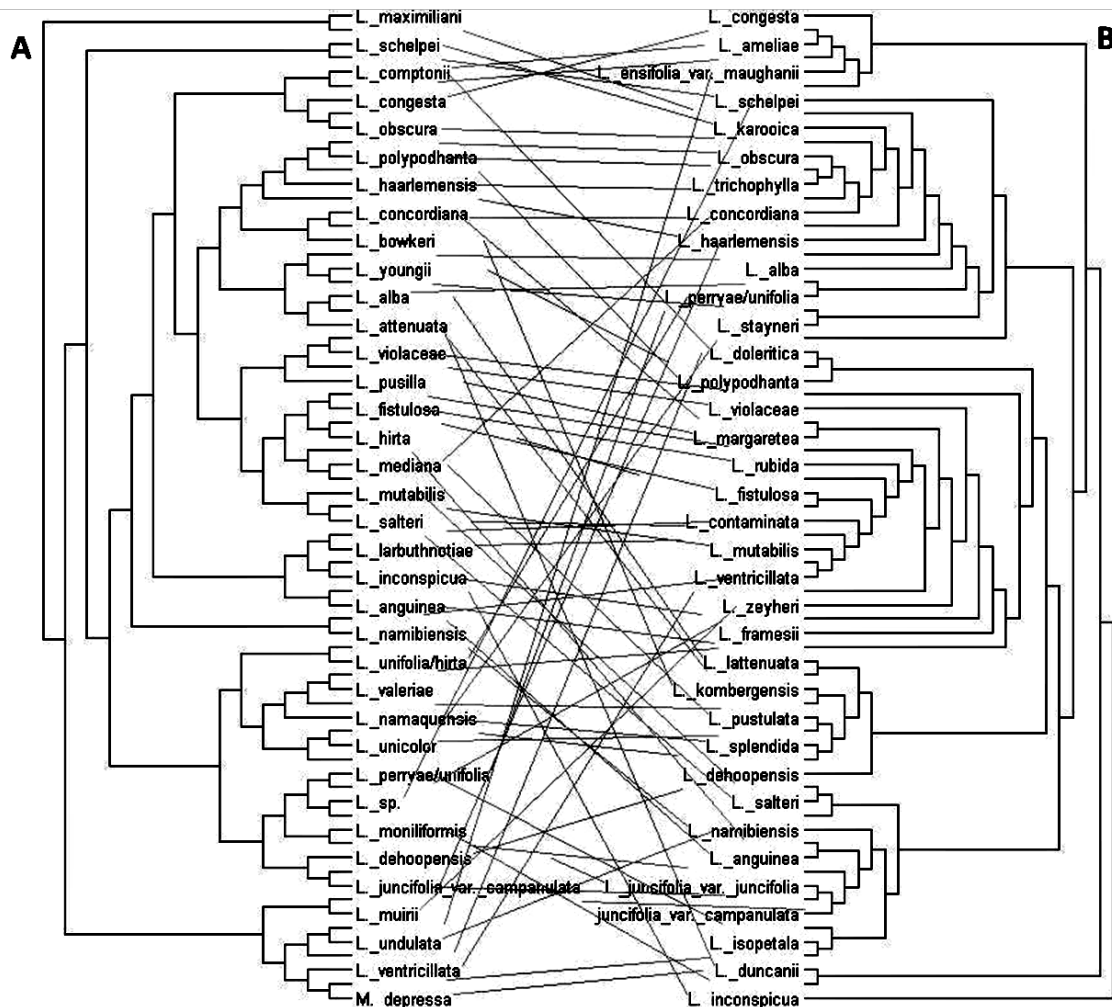


Figure. 5.2: Tanglegram of two BI trees from A, plastid *trnL-F* region and B, nuclear *ITS2* region. Incongruence between nucleus-plastid dataset is as a result of polytomies generated due to the possibility of hybridization and or autopolyploid speciation. For visual purposes, Tanglegram results were compressed, therefore not all the clades/nodes can be seen. Phylogenetic relationship tree constructed using MrBayes v.3.1.2 plugin for Geneious. Graph generated using Dendroscope 3.2.1 (Huson & Linz 2012).

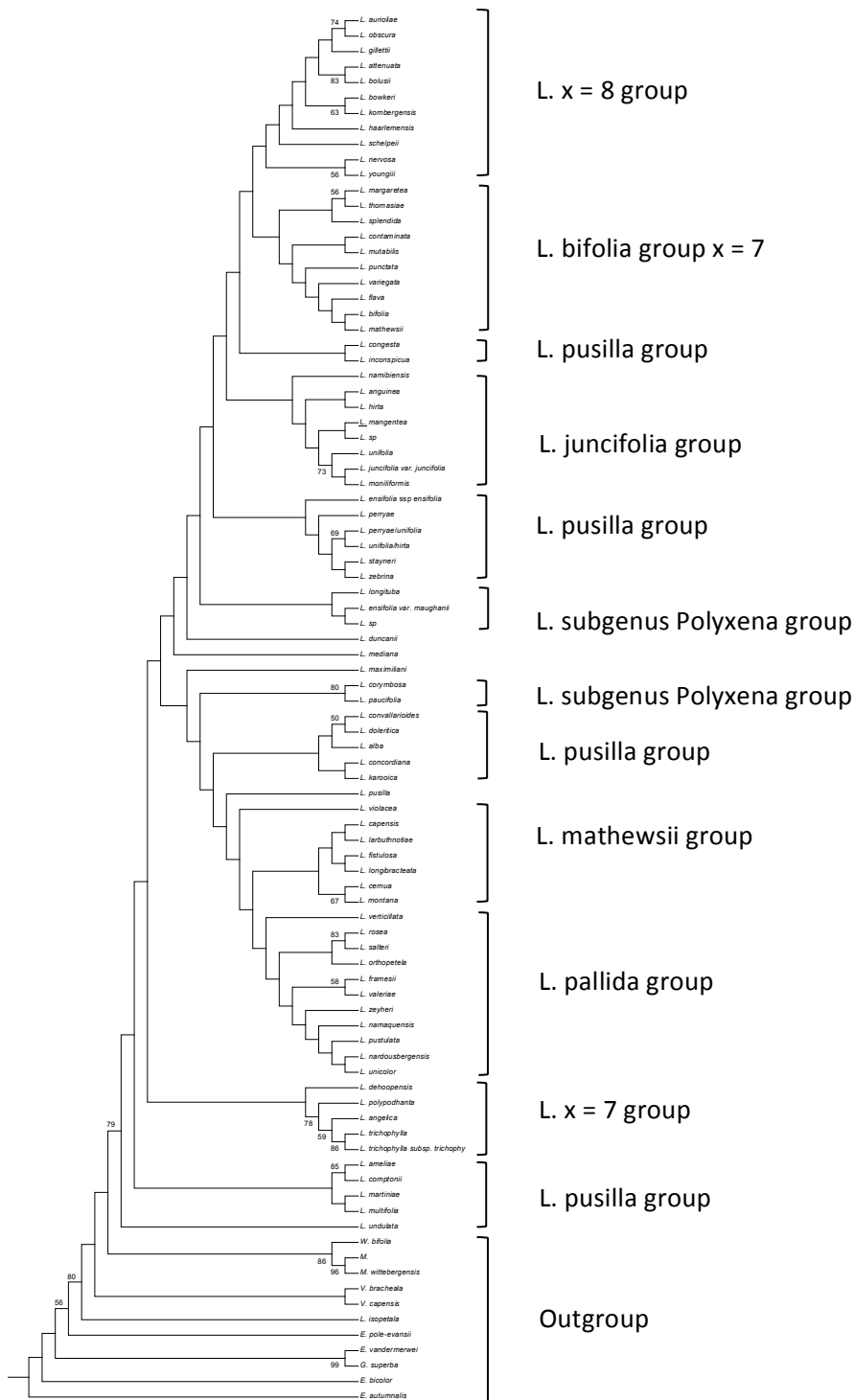


Figure 5.3. Maximum-likelihood estimate of the phylogenetic relationship of *Lachenalia* species based on the combined dataset of plastids (*rbcl* + *trnL-F* + *psbA-trnH*) and nucleus (*ITS*). For clarity purpose, multiple accessions from species show to be exclusive lineages were excluded from the tree. Bootstrap supports for clades >50% is given above branches. ***L. bifolia***, $x = 7$, ***L. juncifolia***, $x = 11$, ***L. mathewsii***, $x = 7$, **Outgroup**, closely related taxa, ***L. subgenus Polyxena***, $x = 12, 13, 14$; ***L. pusilla***, $x = 7, 8, 9, 10, 13$.

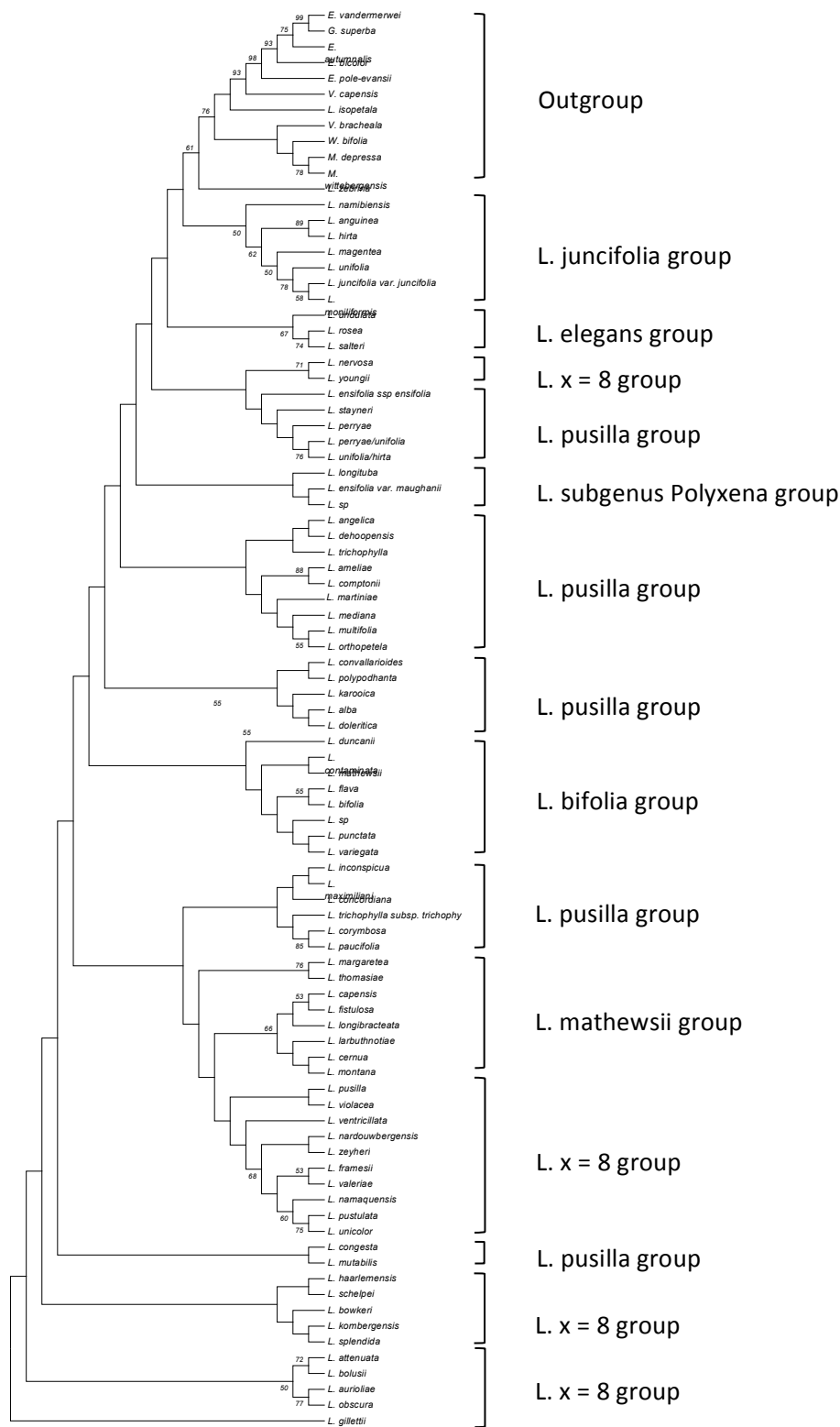


Figure 5.4. Maximum Parsimony estimate of the phylogenetic relationship of *Lachenalia* species based on the combined dataset of plastids (*rbcl* + *trnL-F* + *psbA-trnH*) and nucleus (*ITS*). For clarity purpose, multiple accessions from species show to be exclusive lineages were excluded from the tree. Bootstrap supports for clades >50% is given above branches. ***L. bifolia***, $x = 7$; ***L. elegans***, $x = 7$; ***L. juncifolia***, $x = 11$; ***L. mathewsii***, $x = 7$; **Outgroup**, closely related taxa; ***L. subgenus Polyxena***, $x = 12, 13, 14$; ***L. pusilla***, $x = 7, 8, 9, 10, 13$.

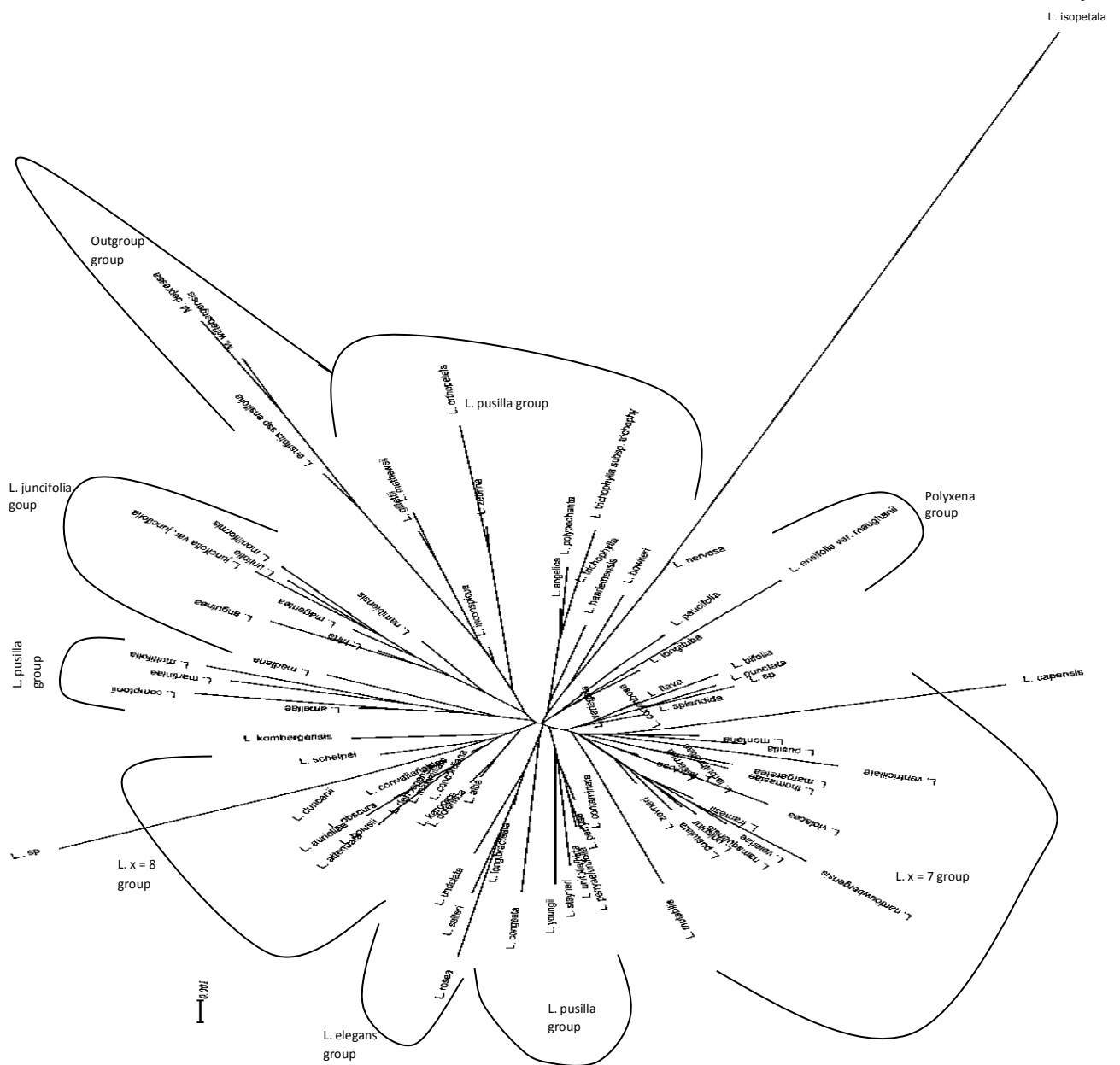


Figure 5.5. Unrooted Neighbor-Joining phylogeny network of *ITS* and combined plastids using uncorrected P distances (SplitTree4 v4.13.1) (Huson & Bryant 2006). Analysis was done on 80 *Lachenalia* accessions and two *Massonia* accessions (outgroup). Bootstrap supports analysis was excluded. ***L. elegans* group**, $x = 7$; ***L. juncifolia* group**, $x = 11$; **outgroup** = closely related taxa; ***L. pusilla* group**, $x = 7, 8, 9, 10$ and 13 ; ***L. subgenus Polyxena* group**.

5.5. Discussion

A similar pattern as Spies (2004), emerged where species with similar chromosome numbers cluster together. The most distinct clades/ groups were *L. juncifolia* group, $x = 11$, *L. pallida* group, $x = 8$ and *L. mathewsii* group $x = 7$. *Lachenalia elegans* group, $x = 7$ emerged on the MP cladogram and NJ networks. Other basic chromosome numbers $x = 9, 10, 13$ grouped together with some $x = 7$ and/or 8 taxa, and formed polyphyletic clades. This resulted in an assumption that species that belong to *L. pusilla* group ($x = 7, 8, 9, 10$ and 13) have a hybrid origin from other *Lachenalia* species with different chromosome numbers.

Similar to previous studies (i.e. Hamatani *et al.* 2008), species with $x = 7$ and $x = 8$ were found to be sister clades. Nevertheless, some species of $x = 7$ and $x = 8$ were scattered in almost all the clades beside the monophyletic clade of $x = 11$ or formed several small clusters within the *Lachenalia* cladogram. Therefore someone can conclude that taxa with $x = 7$ and 8 are polyphyletic groups on the large scale of analyses. Similar to group $x = 7$ or $x = 8$, few species belonging to the clade $x = 11$ were also found in the other clades. Species miss identification might cause this polyphyletic relationship of the genus *Lachenalia*. Therefore the genus *Lachenalia* can be considered as a heterogeneous and highly polymorphic genus.

Observation and analysis of individual datasets or combined datasets (plastid and nuclear), did not clearly resolve all taxa. These gene regions are therefore not sufficient for species level phylogeny in the genus. Poor interspecific phylogenetic resolution suggested that there is high homoplasy within *Lachenalia* species in addition to their chromosomal number differences and morphological variation.

The homoplasy on the molecular level suggest a morphological plasticity due to (1) chromosome variation on the species level (*L. mutabilis* $x = 5, 6$ and 7), (2) geographical

distribution (i.e. *L. bifolia*, Kleynhans & Spies 1999), (3) different species ploidy that may cause phenotypic variation on species level. Interspecific hybridization can significantly increase species homoplasy. Incongruence between datasets might be due to the ancient or recent hybridization within the genus (Fig. 5.2). Spies (2004), reported the possibility that the genus *Lachenalia* is a complex hybridization swarm with groups of $x = 7$ and $x = 8$ representing the primitive basic chromosome numbers.

Results indicated high homoplasy in the genus (Fig. 5.3, Fig. 5.4). Thus, the 50% consensus trees of ML, MP and BI could not well resolve the *Lachenalia* evolutionary relationship. Spies (2004), used *trnL-F* (a molecular phylogenetic gene region proven to have high species phylogenetic relationship resolution) to determined phylogenetic relationship between *Lachenalia* species. The author determined a low CI index of 0.5308 within *Lachenalia* cladogram indication high homoplasy within the genus, thus suggesting that this genus is probably a hybrid swarm. This study further illustrates this hypothesis with SplitsTree Networks (Fig. 5.3).

Based on the phylogenetic analysis, species that belong to *L. pusilla* group might have a hybrid origin. The subgenus *Polyxena* group is a sister taxon to the hybrid clades (*Lachenalia pusilla* group) (Fig. 5.3; Fig. 5.4), however the statistic support of *L. Polyxena* group and *L. pusilla* group are very low to be considered accurate. SplitsTree Networks (Fig. 5.5) analysis on the combined dataset grouped the “*Polyxena* species” (species previously belonging to the genus *Polyxena*) together forming a monophyletic clade nested between the *L. pusilla* group and *Lachenalias* closest relative, the genus *Massonia*, thus supporting the inclusion suggested by Manning *et al.* (2004).

According to the results, DNA dataset alone cannot be used to draw a robust *Lachenalia* phylogenetic relationship. Spies (2004) suggested using multiple molecular markers to determine *Lachenalia* phylogenetic relationships. In this study multiple gene regions (three plastid and one nucleus genes) were used to determine *Lachenalia* phylogenetic relationships on species level. Results suggest that single molecular markers analysis is not useful to study phylogenetic relationships among *Lachenalia* species. Similar to single gene region analysis, multiple gene region analysis are not sufficient for species level phylogeny in the genus, due to a high level of incongruence between datasets. However, combined datasets produced better support for the clades but is not reliable (BS value are lower than 50%).

Morphological and physiological dataset has also been reported not to resolve phylogenetic relationships due to high level of homoplasy, and therefore, cannot result in a robust cladogram either. Due to high chromosome variation, groupings in the genus *Lachenalia* based on chromosome karyotyping and chromosome counts result in a non-robust species classification. Therefore genomic *In Situ* hybridization studies (a tool to identify chromosomes of the parental species in hybrids studies, Chapter 4) is strongly recommended.

Neighbour-Joining network (Fig. 5.5) suggested a cluster of lachenalias with basic chromosome number of $x = 7$. Spies *et al.* (2009) and Hamatani *et al.* (2008; 2010) reported that species with basic chromosome number of $x = 7$ appear divided into sister clades within each other. Similar relationship within taxa with basic chromosome number of $x = 7$ (Fig. 5.5) was found using NJ networks. Species with basic chromosome number $x = 9, 10$ and 13 forms paraphyletic clades. Taxa with basic chromosome number of $x = 11$ form a monophyletic clade. Neighbour-Joining network results were therefore consistent with that of ML and MP.

The evolutionary original of species with $x = 11$ needs to be investigated. Goldblatt *et al.* (2012), hypothesised that the tribe Massonieae possibly evolved from a $x = 10$ ancestor and *Lachenalia* revolved from an ancestor of basic chromosomes $x = 9/10$. This may mean that taxa with $x = 9, 10, 11$, and subspecies *Polyxena* taxon are the basal taxa and an extensive descending dysploid series resulted in evolution of $x = 5, 6, 7$ and 8 groups. Asexual reproduction via bulbils may have allowed low chromosome number species (group $x = 7$ and 8) to persist. The cross-ability between $x = 11$ species is significant higher than crosses done on $x = 7$ species (Appendix C). Could this imply that group $x = 11$ species are basal species and are closest to the tribe Massonieae ancestry species ($x = 10$)?

The genus has a very complex chromosome constitution. The small chromosome within the genus supports the theory that *Lachenalia* is a more advanced genus (Spies 2004). Pfosser *et al.* (2003) reported that there is a remarkably decrease in size and weight of seeds in derived and advanced genera like *Whiteheadia*, *Massonia*, *Lachenalia*, etc. Basal taxa have brown to blackish-brown seeds and all advanced genera have black seeds (Pfosser & Speta 2003). The most advanced genus, genus *Lachenalia* is the perfect example of this correlation as it possesses both small and black seed.

Several species have more than one basic chromosome number i.e. *L. mutabilis*, $x = 5, 6$ and 7 (Crosby 1986; Johnson & Brandham 1997; Spies *et al.* 2009). Few species i.e. *L. hirta* and *L. mediana* have a variable basic chromosome number and therefore no basic chromosome number can be determined, for example, *L. mediana* has a basic chromosome number of $x = 9$ or $x = 13$ (Crosby 1986; Johnson & Brandham 1997; Spies 2004);. These species therefore cannot be grouped based on chromosome number of that particular clade.

5.6. Conclusion

Species miss identification within the tribe Massonieae lead to a high polymorphic and heterogeneous genus. Polytomies can be solved by revisiting the classification of the species in order to determine accurate species inventory and delimitation. Nonetheless, based on chromosome analysis, the genus *Lachenalia* seem to be the most complex genus within the tribe Massonieae. Based on seed morphology, Pfosser *et al.* (2003) supported this conclusion. According to Goldblatt *et al.* (2012) the genus *Lachenalia* originated from an ancestral base of $x = 9/10$ ($n = 5$). This can mean that *Lachenalia* species with $x = 9$ and 10 are the most primitive and species with other basic chromosome numbers evolved by means of an extensive descending dysploid series (often, a very uncommon phenomenon in evolution).

Observations based on molecular systematics were largely similar to those of previous *Lachenalia* phylogenetic analysis. Thus species with basic chromosome number $x = 7$ and 8 are the most primitive species and other chromosomes evolved from them. In addition, subgenus *Polyxena*, *Lachenalia* $x = 9, 10$ and 13 and $x = 7$ and 8 species (*Lachenalia pusilla* group) appear to have evolved due to hybridization speciation with other *Lachenalia* species or with very closely related genera that belong to the tribe Massonieae (genus *Massonia*).

More studies are required to determine reticulate *Lachenalia* phylogenetic relationships and to determine the chromosome evolution of the tribe Massonieae.

6. General Discussion

The morphological classification of *Lachenalia* has created taxonomic confusion, and there is no certain correlation between species chromosome number and molecular systematic when all species within the genus are analyzed. Spies (2004), hypothesised that the genus *Lachenalia* is a possible hybrid swarm. Thus, the presence of natural hybrid species further complicates the phylogenetic relationship of the genus. Nonetheless, different studies suggested similar results regarding the phylogenetic relationship within the genus *Lachenalia*.

In this study similar results of polytomy existence of some species with basic chromosome number $x = 9$, 10 and 13 , and few species with $x = 7$, $x = 8$ and 11 were observed. Similar to a study done on species diversity and reticulate evolution in the *Asplenium normale* complex (Aspleniaceae) in China and adjacent areas (Chang *et al.* 2013), significant incongruence of *Lachenalia* individual plastid and nucleus-plastid phylogenetic (Fig. 6.1) results may support reticulate speciation in the genus *Lachenalia*. Chromosome rearrangement (i.e. dysploidy, *L. latimerae* (Kleynhans *et al.* 2012)), polyploidization and hybridization are the key evolutionary processes in the genus.

It is unlikely that polyploidy acted alone forming autopolyploid species, and the more likely speciation event might be due to hybridization-polyploidization producing allopolyploids species. If the concept of allopolyploidy is correct, then *Lachenalia* subspecies (*Polyxena* species) may have arisen from hybridization between the genera *Lachenalia* and *Massonia* soon followed by polyploidization. ML (Fig. 5.3) and MP (Fig. 5.4) trees suggest a possible multiple origin of the *Polyxena* species from a common ancestor with the *Lachenalia* species

as it do not form a monophyletic clade, (rather group with other *Lachenalia* species). The other speciation scenario of the subgenus *Polyxena* (which could also account for the high morphological variation within the genus) is then hybridization of *Lachenalia* polyploids with different basic chromosome numbers forming new morphological variable species distinct from their parental species.

Interspecific hybridization ensures extensive morphological and chromosomal variation and plays a significant role in plant species diversification (Moraes *et al.* 2013). In spite of rarity per individual basis, natural hybridization is often common per species basis (Mallet 2005). The evolutionary importance of natural hybridization is the ability to increase in ecological flexibility or colonization ability (Hegarty 2012). Therefore, taxonomic problems within *Lachenalia* are probably the results of hybridization and or autopolyploidization.

Available chromosome counts in the genus *Lachenalia* indicate that polyploidy is relatively high and common. Polyploidization in some cases has been associated with stress-survival mechanisms in plants. The deep maroon petal-inflorescence colour of *L. isopetala* ($2n = 40$, $n = 10$) species may be a stress response mechanism possessed by this species. Differences in chromosome number within species are not normally reflected in external morphology and morphological variation within species is more correlated with geographic distribution than with ploidy level (i.e. *L. bifolia*). This conclusion is based only on one single study of *L. bifolia* (Kleynhans & Spies 1999) and more studies will give a better insight.

Several authors have reported on the importance of structural and numerical chromosomal changes contributing to species adaptation (Otto 2007). Success of polyploid species is associated with adaptive plasticity, thus, the ability to colonise a wider range of habitats and to survive better in an unstable climate (Hahn *et al.* 2012). Therefore,

polyploidization might not only contribute in *Lachenalia* speciation but may also have encoded advantageous survival characters (for instance, drought tolerance or pathogen resistance) that promote environmental shift of polyploid species into distinct ecological niche. The invasion of polyploids into isolated and colony selective location might have prevented gene flow enforcing population speciation. Song *et al.* (1995) stated that polyploidization can generate extensive genetic diversity in a short period of time. Therefore, polyploidization not only has a contribution to the survival success but a significant role in the diversification of many lachenalias (polyploid lineage).

Lachenalia unifolia and *L. hirta* (both with $x = 11$) differ mainly in regards to their hairiness. Van Rooyen *et al.* (2002), suggested that *L. unifolia* and *L. hirta* represent two subspecies of the same species rather than separate species. This gives an indication of phenotypic plasticity (the ability of the genotype to express different phenotypes depending on environmental conditions). Phenotypic plasticity is associated with environmental adaptation (Schlichting 1986; De Witt & Scheiner 2004) and invasiveness of species (Richards *et al.* 2006). *Lachenalia bifolia* is an appropriate example of *Lachenalia* phenotypic plasticity (Kleynhans *et al.* 1999). *Lachenalia* phenotypic plasticity may have resulted in miss identification and several unclear phylogenetical determinations. Thus giving the impression of most *Lachenalia* species are immediately descendent form their common ancestor or multiple speciation (polytomies) on the phylogenetic tree. The correlation between morphological variation in the genus *Lachenalia*, hybridization and polyploidization needs to be investigated. Is there any significant morphological variation and phenotypic plasticity with the increase in ploidy level?

Recombination of different genomes can also result in phenotypic diversity (Mallet 2005) and the origin of new hybrid taxa (Abbott 1992). Kleynhans *et al.* (2012) reported on the number of artificial inter-species crosses made at ARC over 35 years (Appendix C). As anticipated, hybridization among taxa with different ploidy levels resulted in high percentage of unsuccessful crosses. Even though the fertility of the 2.7% successful hybridization done among different ploidy level was not determined, it is expected that they have reduced fertility or some may have 0% fertility as different ploidy hybridization is often associated with hybrid sterility (Vallejo-Marin & Lye 2013). Like the hybrid, *Mimulus x robersii* (Vallejo-Marin & Lye 2013), it can be expected that sterile natural hybrids adapted through vegetative propagation resulting in population establishment.

The most dominant modes of speciation in *Lachenalia* are allopatric and ploidy changes (Duncan 2012). It is undoubtful that hybridization within species increase diversity and thus might results in speciation. The multivariate analysis using individual or the combined dataset did not clearly distinguish all the taxa from each other, indicating that *Lachenalia* species might have evolved due to ancient hybridization between the species in the subtribe Massonieae (genus *Massonia* and subgenus *Polyxena*) or they perhaps shared a recent common ancestor.

6.1. Conclusion

The most prominent basic chromosome number is $x = 7$ and $x = 8$ within the genus *Lachenalia*. The basic chromosome number has been determined in a substantial number of *Lachenalia* taxa (about 66.9% taxa with determined chromosome number). Chromosome counts and karyotype studies have been done on one or more specimens from one population.

The basic chromosome number $x = 8$ might have evolved from $x = 7$ soon after it evolved through gained aneuploidy (Spies 2004). Often plants tend to gain or lose chromosomes in responses to stressful conditions. This can result in the numerical chromosome complex. *Lachenalia bifolia* for an example, has $2n = 14, 28, 42, 49$ and 56 (Moffett 1936; Crosby 1986; Johnson & Brandham 1997; Hamatani *et al.* 1998; Kleynhans & Spies 1999; Spies *et al.* 2008). This phenomenon might enhance the feasibility of chromosomal crossover or nonhomologous recombination.

Combined molecular systematic and molecular cytogenetic studies (e.g. Spies 2004 & Hamatani *et al.* 2009 respectively) on *Lachenalia* suggested that: (1), taxa with the same basic chromosome number (x) are closely related (2), taxa with basic chromosome number of $x = 7$ and $x = 8$ are closely related to each other (3), $x = 7$ group seems to form two clusters. Hamatani *et al.* (2009), concluded that $x = 7$ taxa, have two ancestral origins and $x = 8$ taxa has a single origin. Furthermore, Hamatani *et al.* (2008, 2009) suggested that $x = 7$ species evolved from one common ancestor, diverged soon thereafter into parallel species, and $x = 8$ taxa evolved from $x = 7$ species. Species with basic chromosome number of $2n = 14$ ($x = 7$) and $2n = 16$ ($x = 8$) are abundant.

7. Summary

The geophytic genus *Lachenalia* Jacq.f.ex Murray is the largest genus (consists about 133 species, Duncan (2012)) in the tribe Massonieae. It is the most complex and advanced genus within the tribe Massonieae (sub-family Hyacinthoideae) (Pfosser *et al.* 2003). This highly morphological and chromosomal variable genus, is endemic to the south-western winter rainfall regions of Southern Africa with few exceptions and has been recorded in the literature as early as the 17th century (1787 Jacquin).

The genus *Lachenalia* has commercial value as a garden pot plant or for cut flowers. From as early as the 1990s, the ARC started a breeding programme with the aims to improve and increase the international market potential of this plant. There is a correlation between species' basic chromosome number, molecular systematics and cross-ability, thus (1) based on molecular systematics, species with the same basic chromosome number group together on the phylogenetic tree and (2) species in the same group have higher cross-ability.

Several authors revised the classification and grouping of *Lachenalia* species. However, the inventory of the species and species delimitation is not yet complete. Frequently, different species within the genus *Lachenalia* are treated under the same name (species can be mis-identified and then recognised as a different species, i.e., *L. pallida*) or one plant is distributed between different names (i.e. *L. pallida*). This error results in a highly heterogeneous and polymorphic genus that forms polymers or high homoplastic phylogenetic trees. Species with $x = 7$ are ancestral species from which other species evolved.

The evolutionary relationship of $x = 9$, 10 and 13 species, plus $x = 7$ and 8 prostrate and geoflora like species (*Lachenalia pusilla* group) was tested using molecular phylogenetic markers (*ITS*, *rbcl*, *trnL-F* and *psbA-trnH*). Species with $x = 7$ (group $x = 7$), $x = 8$ (group $x = 8$), $x = 11$ (group $x = 11$) and subspecies *Polyxena* (*Polyxena* group) formed monophyletic clades. The majority species from the *Lachenalia pusilla* group are scattered between the other groups. The *Lachenalia pusilla* group may be a hybridization product of *Lachenalia* species with different basic numbers, and/or with their closest *Lachenalia* relative (possibly with the genus *Massonia*). Incongruence test performed on two BI trees of the plastid dataset (*trnL-F*) and nucleus dataset (*ITS2*) (Tanglegram) suggested a hybrid evolution of the *Lachenalia pusilla* group. Evolution of the *Lachenalia pusilla* group (species with $x = 9$, 10 and 13) plus certain $x = 7$ and $x = 8$ species by means of hybridization was also supported by reticulate analyse (NJ tree).

This study suggested a reticulate evolution of the *Lachenalia pusilla* group ($x = 9$, 10 and 13) and few taxon with $x = 7$ and 8 . Previous studies recommended multiple gene phylogenetic analysis for better *Lachenalia* species resolution. However, due to high incongruence between datasets, multiple gene analysis does not provide better answers for *Lachenalia* classification. Genomic *in situ* hybridization, reticulate analysis and other phylogenetic methods can results in better species delimitation of *Lachenalia*.

Keywords: basic chromosome number, GISH, incongruence test, *Lachenalia*, *Lachenalia pusilla* group, *Massonia*, phenotypic plasticity, phylogenetic relationship, reticulate evolution.

8. Opsomming

Die geofietiese genus *Lachenalia* Jacq.f.ex Murray is die grootste genus (bestaan uit ongeveer 133 spesies, Duncan (2012) in die stam Massonieae. Dit is die mees komplekse en gevorderde genus binne die stam Massonieae (Hyacinthoideae sub-familie) (Pfosser *et al.* 2003). Hierdie genus wat morfologiese en chromosomaal hoogs variërend is, is meestal endemies aan die suid-westerlike winterreënvalstreek van Suid-Afrika (met 'n paar uitsonderings) en is reeds vroeg (1787 Jacquin) in die literatuur aangeteken.

Die genus *Lachenalia* het kommersiële waarde as 'n tuin plant, pot plant of snyblomme. So vroeg as die 1990's het die ARC met 'n teelprogram begin met die doelwit om die plante te verbeter en die internasionale markpotensiaal van hierdie plant verhoog. Daar is 'n korrelasie tussen spesies se basiese chromosoomgetal, molekulêre sistematiek en kruis-vermoë, dus (1) spesies met dieselfde basiese chromosoomgetal groepeer saam op die filogenetiese boom (2) spesies in dieselfde groep het 'n hoë kruis-vermoë.

Verskeie outeurs het die klassifikasie en groepering van *Lachenalia* spesies hersien. Maar die indeling van die spesies en spesies afbakening is nog nie voltooi nie. Dikwels word verskillende spesies in die genus *Lachenalia* onder dieselfde naam beskryf (spesies kan verkeerdelik geïdentifiseer word en as 'n verkeerde spesie benaam word soos in die geval van *L. pallida*). Hierdie probleem het 'n hoogs heterogene en polimorfiese genus tot gevolg wat polimere of 'n groot mate van homoplastiese filogenetiese bome tot gevolg het. Spesies met $x = 7$ is die voorvaderlike spesies waaruit ander spesies ontwikkel het.

Ons het met behulp van molekulêre filogenetiese merkers (*ITS*, *rbcl*, *trnL-F* en *psbA-trnH*) die evolusionêre verwantskap tussen spesies met $x = 9$, 10 en 13 getoets, insluitend spesies met $x = 7$ en 8 , wat die uitgestrekte en laag-groeiende spesies (*Lachenalia pusilla* groep) bevat. Spesies met $x = 7$ (groep $x = 7$), $x = 8$ (groep $x = 8$), $x = 11$ (groep $x = 11$) en subspecies *Polyxena* (*Polyxena* groep) het monofiletiese groepe gevorm. Die meerderheid spesies van die *Lachenalia pusilla* groep is tussen die ander groepe versprei. Dit wil voorkom of die *Polyxena* groep 'n gemengde produk tussen *Lachenalia* spesies en die naverwante genus (moontlik die genus *Massonia*) is. Die inkongruensietoets wat op twee BI bome van die plastied datastel (*trnL-F*) en kern datastel (*ITS2*) (Tanglegram) uitgevoer is, suggereer 'n moontlike baster evolusie van die *Lachenalia pusilla* groep. Evolusie van die *Lachenalia pusilla* groep (spesies met $x = 9$, 10 en 13) tesame met sekere $x = 7$ en $x = 8$ spesies, weens verbastering word ook deur die retikulasie-analise (NJ boom) ondersteun.

Hierdie studie suggereer dat daar retikulêre evolusie van die *Lachenalia pusilla* groep ($x = 9$, 10 en 13) en 'n paar takson met $x = 7$ en 8 plaasgevind het. Vorige studies stel voor dat veelvuldige gene vir die filogenetiese analise gebruik moet word om sodoende 'n beter resolusie vir die genus *Lachenalia* te verseker. Maar as gevolg van die hoë mate van onverenigbaarheid tussen datastelle, verskaf die analisering van veelvuldige gene nie beter antwoorde vir die klassifikasie van *Lachenalia* nie. Genomiese in situ verbastering, netwerk analise en ander filogenetiese metodes kan lei tot beter spesies afbakening van *Lachenalia*.

Sleutelwoorde: basiese chromosoomgetal, 'GISH', inkongruensietoets, *Lachenalia*, *Lachenalia pusilla* groep, *Massonia*, fenotipiese plastisiteit, filogenetiese verwantskappe, retikulêre evolusie.

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Appendix A: List of specimens used for the molecular and /or cytogenetic* studies with their localities and voucher specimen numbers. Species are alphabetically arranged and are listed according to the degree reference system (Edwards & Leistner 1971).

Eucomis autumnalis

Locality unknown, grown from seeds, *Spies 9113*.

E. bicolor

Locality unknown grown from seeds, *Spies 9114*.

E. pole-evansii

Locality unknown, grown from seeds, *Spies 9115*.

E. eucomis

Locality unknown, grown from seeds, *Spies 9116*.

Gloriosa superba

Locality unknown, grown from seeds, *Spies 9117*.

***Lachenalia alba* W.F. Barker ex G.D. Duncan**

NORTHERN CAPE.–3119 (Calvinia): Nieuwoudtville (-AD), *Spies 8897*.

***L. flava*.**

WESTERN CAPE.–3319 (Worcester): Bainskloof (-AC), *G. Duncan 363*.

***L. ameliae* W.F. Barker**

WESTERN CAPE.–3319 (Worcester): Karoopoort (-BA), *Spies 8927*.

***L. angelica* W.F. Barker**

NORTHERN CAPE.–3017 (Hondeklipbaai): Soebatsfontein (-BA), *Spies 8926*.

***L. anguinea* Sweet**

WESTERN CAPE.–3118 (Vredendal): Vredendal (-AB), *Spies 8917*.

***L. attenuata* W.F. Barker ex G.D. Duncan**

NORTHERN CAPE.–3220 (Sutherland): Sutherland (-BC), *R. Saunders s.n.*

***L. aurioliae* G.D. Duncan**

WESTERN CAPE.–3319 (Worcester): De Doorns (-BC), *Spies 8308*.

***L. bolusii* W.F. Barker**

WESTERN CAPE.–3219 (Clanwilliam): Bidouw (-AA), *Spies 7987**.

***L. bowkeri* Baker**

WESTERN CAPE.–3123 (Murraysburg): Murraysburg (-DD), *Spies 8930*.

***L. bifolia* (Cirillo) Engl.**

WESTERN CAPE.—3419 (Caledon): Pearly Beach (-CB), *Spies 7064**.

***L. capensis* W.F. Barker**

WESTERN CAPE.—3418 (Simons Town): Simons Town (-AB), *G. Duncan 139*.

L. cernua

WESTERN CAPE.—3319 (Worcester): Goudini (-BA), *Spies 8222*.

***L. comptonii* W.F. Barker**

WESTERN CAPE.—3321 (Cape Town): Kirstenbosch (-AB), *Spies 7067**.

***L. concordiana* Schltr. ex W.F. Barker**

NORTHERN CAPE.—3119 (Calvinia): Calvinia (-BD), *M. Thomas s.n.*

***L. congesta* W.F. Barker**

NORTHERN CAPE.—3220 (Sutherland): Komsberg Pass (-DB), *J. Manning s.n.*

***L. contaminata* Aiton**

WESTERN CAPE.—3318 (Cape Town): Darling (-AD), *G. Duncan 321*.

***L. convallarioides* Baker**

EASTERN CAP.— 3326 (Grahamstown): Grahamstown (-BC), *A. Dold 1018*.

***L. corymbosa* (L.) J.C.**

Locality unknown, grown from seeds, *Spies 9107*.

***L. dehoopensis* W.F. Barker**

WESTERN CAPE.—3420 (Bredasdorp): De Hoop Nature Reserve (-AD), *A. Scott s.n.*

***L. doleritica* G.D. Duncan**

NORTHERN CAPE.—3119 (Calvinia): Calvinia (-BD), *M. Thomas s.n.*

***L. duncanii* W.F. Barker**

NORTHERN CAPE.—3018 (Kamiesberg): Kliprand (-DA), *G. Duncan 189*.

***L. ensifolia* subsp. (Thunb.) J.C. Manning & Goldblatt**

Locality unknown, grown from seeds, *Spies 9108*.

***L. ensifolia* subsp. *maughanii* (W.F. Barker)**

Locality unknown, grown from seeds, *Spies 9109*.

***L. fistulosa* Baker**

WESTERN CAPE.—3318 (Cape Town): Signal Hill (-CD), *G. Duncan 3032*.

***L. framesii* W.F. Barker**

NORTHERN CAPE.–2917 (Komaggas): Komaggas (-CD), *Spies 8241*.

***L. gillettii* W.F. Barker**

WESTERN CAPE.–3218 (Clanwilliam): Piketberg (-DA), *C. Taswell-Yates s.n.*

***L. haarlemensis* Fourc.**

NAMIBIA.–2219 (Sandfontein): Haarlem (-AB), *P. Perry 3258*.

***L. hirta* (Thunb.) Thunb.**

WESTERN CAPE.–3218 (Clanwilliam): 3 km. from Elandsbaai (-AD), *Spies 6858*. 3318
(Cape Town): Jonkershoek Forestry Station (-DD), *Spies 6891*.

***L. inconspicua* G.D. Duncan**

NORTHERN CAPE.–2918 (Gamoep): Gamoep (-AD), *Spies 8906*.

***L. isopetala* Jacq.**

NORTHERN CAPE.–3120 (Calvinia): Middelpoort (DC), *Spies 8913*.

Locality unknown, grown from seeds, *Spies 9110*.

***L. juncifolia* var *juncifolia* Baker**

WESTERN CAPE.–3319 (Worcester): Rawsonville (-CD), *Spies 8245**.

L. magentea

Locality unknown, grown from seed, *Spies 8313**.

***L. karooica* W.F. Barker ex G.D. Duncan**

FREE STATE.–2925 (Jagersfontein): Fauresmith (-CB), *G. Duncan 367*.

L. kombergensis

Locality unknown, grown from seed, *Spies 8131*.

L. longibracteata

WESTERN CAPE.–3218 (Clanwilliam): Piketberg (-DA), *Spies 7768*.

***L. longituba* (A.M. van der Merwe). J.C. Manning & Goldblatt**

Locality unknown, grown from seeds, *Spies 9111*.

***L. margaretea* W.F. Barker**

WESTERN CAPE.–3218 (Clanwilliam): Pakhuis Pass (-BB), *F. Paterson s.n.*

***L. martinae* W.F. Barker**

WESTERN CAPE.–3218 (Clanwilliam): Clanwilliam (-BB), *G. Duncan 141*.

***L. mathewsii* W.F. Barker**

Locality unknown, grown from seed, *Spies 6938*.

***L. maximiliani* Schltr. ex W.F. Barker**

WESTERN CAPE.–3418 (Simons town): Wuppertal (-BB), *G. Duncan 284*.

L. mediana

EASTERN CAPE.–3325 (Port Elizabeth): Van Staadens Reserve (-CC), *Spies 9028**.

NORTHERN CAPE.–3320 (Montagu): Laingsburg (-BB), *Spies 8321*.

WESTERN CAPE.–3318 (Cape Town): Potsdam behind Killarneys Race Course (-DC), *Spies 9031**.

Locality unknown, *Spies 9027**.

***L. moniliformis* W.F. Barker**

WESTERN CAPE.–3319 (Worcester): Worcester (-CB), *P. Perry 795*.

***L. montana* W.F. Barker**

WESTERN CAPE.–3419 (Caledon): Hermanus (-AC), *Spies 8314*.

***L. multifolia* W.F. Barker**

Locality unknown, grown from seed, *Spies 8263*.

***L. mutabilis* Sweet**

WESTERN CAPE.–3219 (Wuppertal): Kouberg (-AA), *R. Saunders 188*.

***L. namaquensis* Schltr. ex W.F. Barker**

NORTHERN CAPE.–2917 (Springbok): Kosies (-BA), *N. van Berkel 273*.

***L. namibiensis* W.F. Barker**

NAMIBIA.–2616 (Aus): Numis (-AA), *P. Bruyns 7217*.

L. nardouwbergensis

WESTERN CAPE.–3119 (Vanrhynsdorp): Moedverloor (-CC), *Spies 8267*.

***L. nervosa* Ker Gawl.**

WESTERN CAPE.–3420 (Bredasdorp): Swellendam (-AB), *M. Botha s.n.*

***L. obscura* Schltr. ex G.D. Duncan**

WESTERN CAPE.–3320 (Matjiesfontein): Matjiesfontein, on a farm near the Wauchope memorial (-AB), *Spies 8898**.

***L. orthopetala* Jacq.**

WESTERN CAPE.–3318 (Cape Town): Durbanville (-DC), *G. Duncan 262*.

***L. paucifolia* (W.F. Barker) J.C. Manning & Goldblatt**

WESTERN CAPE.–3217 (Vredenburg): Vredenburg (-DD), *Spies 8915*.

Locality unknown, grown from seeds, *Spies 912*.

***L. perryae* G.D. Duncan**

WESTERN CAPE.—3218 (Clanwilliam): Paleisheuvel (-BC), *Spies 6961*.

L. perryae/unifolia

NORTHERN CAPE.—3320 (Montagu): Matjiesfontein, on the road to Whitehill Station (-BA), *Spies 9039**.

***L. polypodantha* Schltr. ex W.F. Barker**

NORTHERN CAPE.—2918 (Springbok): Varsputs, between Springbok and Pofadder (-AD), *Spies 8912*.

***L. pusilla* Jacq.**

WESTERN CAPE.—3218 (Clanwilliam): Piketberg (-DA), *Spies 8911**.

***L. pustulata* Jacq.**

Locality unknown, grown from seed, *Spies 8284*.

***L. rosea* Andrews**

WESTERN CAPE.—3419 (Caledon): Greyton (-BA), *M. Hofmeyer s.n.*

***L. punctata* Jacq.**

WESTERN CAPE.—3418 (Simons town): 60 km from Faure turn off, Cape Town area (-BB), *Spies 7093**.

***L. salteri* W.F. Barker**

WESTERN CAPE.—3420 (Bredasdorp): Bredasdorp (-CA), *Spies 8289*.

***L. schelpei* W.F. Barker**

NORTHERN CAPE.—3018 (Kamiesberg): Knersvlakte (-DA), *Spies 8904*.

***L. splendida* Diels**

Locality unknown, grown from seed, *Spies 8290*.

***L. stayneri* W.F. Barker**

WESTERN CAPE.—3319 (Worcester): Worcester (-CB), *Spies 8316*.

***L. thomasiae* W.F. Barker ex G.D. Duncan**

WESTERN CAPE.—3218 (Clanwilliam): Die Poort (-BB), *Spies 8905*.

***L. trichophylla* Baker**

NORTHERN CAPE.—3019 (Kamiesberg): Loeriesfontein (-CD), *Spies 8910*.

L. trichophylla* subsp. *trichophylla

Locality unknown, grown from seeds, *Spies 8293*

L. undulata

WESTERN CAPE.–3118 (Vredendal): Between Lutzville and Vredendal (-AC), *Spies 8317**.

***L. unicolor* Jacq.**

WESTERN CAPE.- 3318 (Cape town): Koringberg (-BA), *Spies 8294**.

***L. unifolia* Jacq.**

NORTHERN CAPE.–3017 (Hondeklipbaai): Soebatsfontein road, 5 km from Kamieskroon (-BB), *Spies 6878**.

WESTERN CAPE.–3318 (Wuppertal): Jonkershoek forestry station (-DD), *Spies 6874**.

L. unifolia/hirta

WESTERN CAPE.–3318 (Cape Town): Tienie Versveldt Nature Reserve, Darling(-AD), *Spies 6902**.

***L. valeriae* G.D. Duncan**

NORTHERN CAPE.–3017 (Hondeklipbaai): Wallekraal- Soebatsfontein road, west of Kammiesberg (-BB), *Spies 8919**.

***L. variegata* W.F. Barker**

Locality unknown, grown from seed, *Spies 7097*.

***L. verticillata* W.F. Barker**

NORTHERN CAPE.–2917 (Springbok): Springbok (-DB), *Spies 8914*; Steinkopf (-BD), *D. Müller-Doblies 88125*.

L. violacea

NORTHERN CAPE.–3017 (Hondeklipbaai): Road between Garies and Kamieskroon (-BB), *Spies 7101*.

***L. youngii* Baker**

WESTERN CAPE.–3423 (Knysna): Storms River (-BB), *R. McMaster s.n.*

L. zebrina* Baker forma *zebrina

NORTHERN CAPE.–3120 (Calvinia): Middelpoos (-DC), *Spies 8920*.

***L. zeyheri* Baker**

Locality unknown, grown from seed, *Spies 7099*.

***Lachenalia* sp**

Locality unknown, grown from seeds, *Spies 9101*.

L. sp

Locality unknown, grown from seeds, *Spies 9102*.

***Massonia depressa* Houtt**

Locality unknown, grown from seeds, *Spies 9103*.

M. wittebergensis

Locality unknown, grown from seeds, *Spies 9104*.

Veltheimia brachteata

Locality unknown, grown from seeds, *Spies 9105*.

***V. capensis* (L.) D.C.**

Locality unknown, grown from seeds, *Spies 9106*.

Whiteheadia bifolia

Locality unknown, grown from seeds. *Spies 9118*.

Appendix B. List of taxon and their phylogenetic marker unsuccessfully amplified or sequenced.

trnL-F— *Eucomis autumnalis*, *E. pole-evansii*; *Gloriosa superba*; *Massonia wittebergensis*; *Lachenalia longibracteata*, *L. nardouwbergensis*, *L. orthopetala*, *L. sp*, *L. trichophylla* subsp. *trichophylla*. **ITS**— *Eucomis autumnalis*, *E. pole-evansii*, *E. bicolor*, *E. vandermerwei*; *Gloriosa superba*; *Lachenalia ensifolia*. subsp. *ensifolia*, *L. gillettii*, *L. multifolia*, *L. nervosa*, *L. variegata*; *Veltheimia brachteata*, *V. capensis*. **psbA-trnH**— *Eucomis autumnalis*, *E. pole-evansii*, *E. bicolor*, *E. vandermerwei*; *Massonia depressa*, *M. wittebergensis* *Lachenalia concordiana*, *L. contaminate*, *L. convallarioides*, *L. dehoopensis*, *L. duncanii*, *L. fistulosa*, *L. inconspicua*, *L. arbutnotiae*, *L. mathewsii*, *L. maximiliani*, *L. namibiensis*, *L. polypodantha*, *L. salteri*, *L. trichophylla*, *L. zebrina*.

Appendix C: Number of inter-species crosses made among various different *Lachenalia* species and the results obtained from these crossing combinations. (Table copied from Kleynhans *et al.* 2012 with permission of the authors).

| Basic chromosome number of parents | No. of successful crosses | No of unsuccessful crosses | | |
|-----------------------------------------------------|---------------------------|---------------------------------|-----------------------------------|------------------------------------|
| | | No. of crosses with no seed set | No. of crosses with abnormal seed | No. of crosses with seedling death |
| 7x7 | 169(27%) | 274(44%) | 169(27%) | 10(2%) |
| 8x8 | 72(46%) | 44(28%) | 40(45%) | 1(1%) |
| 11x11 | 2(67%) | | 1(33%) | |
| 7x8 | 20(6%) | 251(79%) | 44(14%) | 3(1%) |
| 8x7 | 59(18%) | 111(34%) | 155(47%) | 6(2%) |
| 7x10 | | 17(100%) | | |
| 10x7 | 1(5%) | 5(25%) | 13(65%) | 1(5%) |
| 7x11 | 1(2%) | 54(86%) | 8(13%) | |
| 11x7 | 4(6%) | 23(33%) | 39(57%) | 3(4%) |
| 9x8 | | | 1(100%) | |
| 8x10 | | 1(33%) | 2(67%) | |
| 10x8 | 2(33%) | 1(17%) | 2(33%) | 1(17%) |
| 8x11 | 1(3%) | 23(79%) | 5(17%) | |
| 11x8 | 1(3%) | 15(39%) | 22(58%) | |
| 11x10 | | 1(100%) | | |
| 15x7 | | 2(67%) | 1(33%) | |
| Unknown basic numbers in one or both of the parents | 4(2%) | 117(59%) | 78(39%) | |
| L | 336(18%) | 939(50%) | 580(31%) | 25(1%) |

Appendix D: Aligned sequences of the comibed *ITS2* and indels dataset of all the specimens used for cladistics analysis. *M.* = *Massonia*, *L.* = *Lachenalia*, *W.* = *Whiteheadia*.

| | |
|-------------------------------|--------------------------------------------------------------|
| L. angelica | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. alba | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. flava | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. ameliae | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. anguinea | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCTTT |
| L. attenuata | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. aurioliae | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. bolusii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. bowkeri | AGTTGCTCCGGAGGATCCG-GCCGAGGGCACGCCAGCCTGGGCTTACGCATTGCGTC |
| L. bifolia | AGTTGCCCCGAGGCTATCCG-GCCGAGGGCACGCCAGCCTGGGCGTCACGCATCGCGTC |
| L. capensis | AGTTGCTCCGGAGGCTATCCG-GCCGAGGGCACGCCGACCTGGGCGTCACGCATCGCGT- |
| L. cernua | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. comptonii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. concordiana | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. congesta | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. contaminata | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. convallarioides | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. dehoopensis | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. doleritica | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. duncanii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. ensifolia var. maughanii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. fistulosa | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. framesii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. haarlemensis | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. hirta | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. inconspicua | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. isopetala | AGTTGCGCATGAGGCTATCCG-GCTGAGGGCACGCCGCTGGGCGTCACGCATCGCGTC |
| L. magentea | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. juncifolia var. juncifolia | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. karoocia | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTTACGCATCGCGTC |
| L. kombergensis | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. arbutnotiae | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. longituba | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. longibracteata | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. margaretea | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTC-CGCATCGTGT |
| L. martiniae | AGTTGCGCCCGAGGCTATCCGCGCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| L. mathewsii | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |

| | |
|--------------------------------------------|--------------------------------------------------------------|
| <i>L. maximiliani</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. mediana</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. moniliformis</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. muirii</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. mutabilis</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCATGCATCGCGTC |
| <i>L. namaquensis</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. namibiensis</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. nardousbergensis</i> | AGTTGCTCCGGAGGCTATCCG-GCCGAGGGCACGCATACCTGGGCGTCACGCATCGCGTC |
| <i>L. obscura</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. orthopetala</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. paucifolia</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. perryae</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. perryae/unifolia</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. polypodantha</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. pusilla</i> | AGTTGCTCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. pustulata</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. rosea</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. punctata</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. salteri</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. schelpei</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. sp</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. sp</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. splendida</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. stayneri</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. thomasiae</i> | AGTTGCGCCCGAGGCTATCCG-GCCAAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. trichophylla</i> | AGTTGCGCCCTAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. trichophylla subsp. trichophylla</i> | AGTTGCGCCCTAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. undulata</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. unicolor</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. unifolia</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. unifolia/hirta</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. valeriae</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. verticillata</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. violacea</i> | ?GTTGCGCCCGAGG-TATCCG-GCCGAGGGCACGCATGCCTGGGCGTCACGCATCGCGTC |
| <i>L. youngii</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. zebrina</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>L. zeyheri</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>M. depressa</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATTGCGTC |
| <i>M. wittebergensis</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |
| <i>W. bifolia</i> | AGTTGCGCCCGAGGCTATCCG-GCCGAGGGCACGCCTGCCTGGGCGTCACGCATCGCGTC |

| | |
|---------------------------------------------|--------------------------------------------------------------|
| <i>L. angelica</i> | GCTT-CGCG-TACCCC-ACGCCCCC-GCCCAGTGCGGAC-GGCGGCGCGGACGCGGACG |
| <i>L. alba</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. flava</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. ameliae</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGC-GGCGGCGCGGACGCGGACG |
| <i>L. anguinea</i> | TTTT-TCTGTACCCC-ACGTCCCC-GCACAGCGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. attenuata</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. aurioliae</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGTACGCGGACG |
| <i>L. bolusii</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. bowkeri</i> | GCTT-TGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. bifolia</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGAGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. capensis</i> | GCTC-CGGCGTACCCC-ACGTAACC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. cernua</i> | GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. comptonii</i> | GCTCGCG---TACCCC-ACGTCCCC-GCCTAGTGCGGGC-GGCGGCGCGGACGCGGACG |
| <i>L. concordiana</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. congesta</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGC-GGCGGCGGCATACGCGGACG |
| <i>L. contaminata</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGAT-GGCGGCGCGGACGCGGACG |
| <i>L. convallarioides</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. dehoopensis</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGATG |
| <i>L. doleritica</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. duncanii</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. ensifolia</i> var. <i>maughanii</i> | GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. fistulosa</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. framesii</i> | GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGGCGCGGACG |
| <i>L. haarlemensis</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGAGGAGGACGCGGACG |
| <i>L. hirta</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. inconspicua</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. isopetala</i> | GCTC-CGTG-TACTTC-GTGTCCCCGACATTTGCGGGT-GATGATGGTGGCCGTGGAAG |
| <i>L. magentea</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGATG |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. karoocia</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. kombergensis</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. arbuthnotiae</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. longituba</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. longibracteata</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. margaretea</i> | GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. martiniae</i> | GCTT-CGCG-TACCCC-ATGTCCCC-GCCCATTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. mathewsii</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. maximiliani</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. mediana</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. moniliformis</i> | GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGCGGGT-GGCGGCGCGGACGCGGACG |
| <i>L. muirii</i> | GCTC-CGCG-TACCCC-ATGTCCCC-GCCCAGTGCGGGT-GGCGGCGGTGGACGCGGACG |

L. mutabilis GCTT-CGCG-TACCCC-ACGTCTCC-ACCTAGTGC GGAT-GGCGGCGCGGACGCGGACG
L. namaquensis GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGATGCGGACG
L. namibiensis GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. nardousbergensis GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. obscura GCTT-CGCG-TACCCC-ATGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGTACGCGGACG
L. orthopetala GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. paucifolia GCTT-CGCG-TACCCC-ACGTCCC----CCATTGCGGGT-GGCGGCGCGGACGCGGACG
L. perryae GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGGTGGACGCGGACG
L. perryae/unifolia GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. polypodantha GCTT-CGCG-TACCCCCACGCCCC-GCCCAGTGC GGAC-GGCGGCGCGGACGCGGACG
L. pusilla GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCAGC---GGACGCGGACG
L. pustulata GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. rosea GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. punctata GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. salteri GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGAT-GGCGGCTGCGGACGCGGACG
L. schelpei GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. sp GCTT-CGCG-TACCCC-ACGTCCCC-GCCAAGTGC GGGC-GGCGGCGCGGACGCGGACG
L. sp GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. splendida GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. stayneri GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACACGGACG
L. thomasiae GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. trichophylla GCTC-CGCG-TACCCC-ACGCCCC-GCCCAGTGC GGAC-GGCGGCGCGGACGCGGACG
L. trichophylla subsp. trichophylla GCTC-CGCG-TACCCC-ACGCCCC-GCCCAGTGC GGAC-GGCGGCGCGGACGCGGACG
L. undulata GCTT-CGCG-TACCCC-ACGTCCCC-GCCCATTGCGGGT-GGCGGCGCGGACGCGGACG
L. unicolor GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACACGGACG
L. unifolia GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. unifolia/hirta GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. valeriae GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. verticillata GCTC-CGCG-TACCCC-ACGTCCCCCGCCGGTGC GGGT-GGCGTTGGCGGACGCGGACG
L. violacea GCTC-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. youngii GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
L. zebrina GCTC-CGGG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACACGGACG
L. zeyheri GCTT-CGCG-TACCCC-ACGTCCCC-GCCCAGTGC GGGT-GGCGGCGCGGACGCGGACG
M. depressa GCTT-CGCG-TACCCT-ATGTCTCCGCTAGTGTGGT-GGCTATG-CGGACGCGGACG
M. wittebergensis GCTT-CGTG-TACCCT-ATGTCTC-GCCTAGTGTGGT-GGCTAT-GCGGACGCGGACG
W. bifolia GCTC-CGCG-TACCCT-ATGTCTCCGCTGTTGCGGGT-GTCTATGGCGGACGCGGATG

L. angelica TGGAGATTGGCCCCC-GTG---CCTCGGCGCGGGCGGG-TCGAAGTGC GGG-CCGCCG
L. alba CGGAGATTGGCCCCC-GTG---CCTCGGCGCGGGCGGG-TCGAAGTGC GGG-CCGCCG
L. flava CGGATATTGGCCCCC-GTG---CCTCGGCGCGGGCGGG-TCGAAGTGC GGG-CCGCCG
L. ameliae CGGAGATTGGCCCCC-GTG---CCTCGGCGCGGGCGGG-TCGAAGTGC GGG-CCGCCG

| | |
|---------------------------------------------|------------------------------------------------------------|
| <i>L. anguinea</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. attenuata</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. auriloliae</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG??????? |
| <i>L. bolusii</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. bowkeri</i> | CGGAGATTGGCCCCC-GTG---CCTCGGGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. bifolia</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TTGAAGTGTTGG-CCGCCG |
| <i>L. capensis</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TTGAAGTTCGGG-CTGCCG |
| <i>L. cernua</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. comptonii</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. concordiana</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-TCGCCG |
| <i>L. congesta</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TTGAAGTGCGGG-CCGCCG |
| <i>L. contaminata</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. convallarioides</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. dehoopensis</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. doleritica</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. duncanii</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. ensifolia</i> var. <i>maughanii</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. fistulosa</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCTGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. framesii</i> | CGGATATTGGCCCTC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. haarlemensis</i> | TGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. hirta</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. inconspicua</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TTGAAGTTCGGTCCGCCG |
| <i>L. isopetala</i> | CGGAGATTGGCCCCC-ATG---CCTGGGGGCGGCGGG-TGGAAGTGCTGG-CCGCCG |
| <i>L. magentea</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. karoocica</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. kombergensis</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. arbutnotiae</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TTGAAGTTCGGG-CTGCCG |
| <i>L. longituba</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. longibracteata</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. margaretea</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. martiniae</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGTTGG-CCGCCG |
| <i>L. mathewsii</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGTTGGTATGAAAGTGCGGGCCGCCG |
| <i>L. maximiliani</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. mediana</i> | CGGATATTGGCCCCC-GTG---CCTTGGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. moniliformis</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. muirii</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. mutabilis</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGAGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. namaquensis</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. namibiensis</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. nardousbergensis</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGGCGGG-TCGAAGTGTTGG-CCTCCG |

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|--------------------------------------------|--------------------------------------------------------------|
| <i>L. obscura</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. orthopetala</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TTGAAGTCTGG-CTGCCG |
| <i>L. paucifolia</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. perryae</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. perryae/unifolia</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. polypodantha</i> | TGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. pusilla</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. pustulata</i> | CGGATATTGGCCCCC-GTG---CCTTGC GGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. rosea</i> | CGGATATTGGCCCCC-GTG---CCTTGC GGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. punctata</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGTGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. salteri</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. schelpei</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. sp</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. sp</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. splendida</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. stayneri</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. thomasiae</i> | TGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TTGAAGTGCGGG-CCGCCG |
| <i>L. trichophylla</i> | TGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. trichophylla subsp. trichophylla</i> | TGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. undulata</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. unicolor</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. unifolia</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. unifolia/hirta</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. valeriae</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>L. verticillata</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. violacea</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. youngii</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCGGG-CCGCCG |
| <i>L. zebrina</i> | CGGATATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TTGAAGTCTGG-CCGCCG |
| <i>L. zeyheri</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGCTGG-CCGCCG |
| <i>M. depressa</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGTGGG-CCGCCG |
| <i>M. wittebergensis</i> | CGGAGATTGGCCCCC-GTG---CCTCGCGGCGCGGCGGG-TCGAAGTGTGGG-CCGCCG |
| <i>W. bifolia</i> | CGGAGATTGGCCCCC-GTC---CCTCGCGGCGCGGCGGG-TCGAAGTGTGGG-CCGCCG |
| | |
| <i>L. angelica</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. alba</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. flava</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. ameliae</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. anguinea</i> | GCCGAAGAAGAAGAGGAAAGGGAG |
| <i>L. attenuata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. aurioliae</i> | ????AAGAAGAGGAGGAAAGGGGA |
| <i>L. bolusii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |

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|---------------------------------------------|---------------------------|
| <i>L. bowkeri</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. bifolia</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. capensis</i> | GCCGAAGAGGAAGAGGAAAGGGAG |
| <i>L. cernua</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. comptonii</i> | GCCGAAGAAAGAGGAGGAAAGGGAG |
| <i>L. concordiana</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. congesta</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. contaminata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. convallarioides</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. dehoopensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. doleritica</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. duncanii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. ensifolia</i> var. <i>maughanii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. fistulosa</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. framesii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. haarlemensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. hirta</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. inconspicua</i> | GCCGAAGAAGAGGAGGAAAGGGAA |
| <i>L. isopetala</i> | ACTGAAGAAGAGGAAGAAAGGGAG |
| <i>L. magentea</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. karooica</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. kombergensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. arbutnotiae</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. longituba</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. longibracteata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. margaretea</i> | GCCGAAGGAGAGGAGGAAAGGGAG |
| <i>L. martiniae</i> | GCCGAAAAAGAGGAGGAAAGGGAG |
| <i>L. mathewsii</i> | GCCGAAGAAGAGGAGGAAAGGAAA |
| <i>L. maximiliani</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. mediana</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. moniliformis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. muirii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. mutabilis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. namaquensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. namibiensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. nardousbergensis</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. obscura</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. orthopetala</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. paucifolia</i> | GCCGAAGAAGAGGGAGGAAAGGGAG |
| <i>L. perryae</i> | GCCGAAGAAGAGGAGGAAAGGGAG |

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|--------------------------------------------|--------------------------|
| <i>L. perryae/unifolia</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. polypodantha</i> | GCCGAAGAAGAGAAGGAAAGGGAG |
| <i>L. pusilla</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. pustulata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. rosea</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. punctata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. salteri</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. schelpei</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. sp</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. sp</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. splendida</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. stayneri</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. thomasiae</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. trichophylla</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. trichophylla subsp. trichophylla</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. undulata</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. unicolor</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. unifolia</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. unifolia/hirta</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. valeriae</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. verticillata</i> | GCCGAAGAAGAGGAAGAAAGGGAG |
| <i>L. violacea</i> | GCCGGGGAAGAGGAGGAAAGGGAG |
| <i>L. youngii</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. zebrina</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>L. zeyheri</i> | GCCGAAGAAGAGGAGGAAAGGGAG |
| <i>M. depressa</i> | GCCGAAGAAGAGGAAGAAGGGGAG |
| <i>M. wittebergensis</i> | GCCGAAGAAGAGGAGGAGAGGGAG |
| <i>W. bifolia</i> | GCCGAAGAAGAGGAAGAAAGGGAG |

Appendix E: Aligned sequences of the combined *psbA-trnH* and indels dataset of all the specimens used for cladistics analysis. *E.* = *Eucomis*, *G.* = *Gloriosa*, *M.* = *Massonia*, *L.* = *Lachenalia*, *V.* = *Veltheimia*, *W* = *Whiteheadia bifolia*.

| | |
|-------------------------------|--------------------------------------------------------------|
| G. superba | ??????????G---AAGCTCC-GTCCA----CAAAGGGA-TAAG-----ACATC |
| L. angelica | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. alba | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATTC-----CCAAT |
| L. flava | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. ameliae | TAATGTA-TTAAG----AATCCTT-GAATTCAAGGCAAGGAGC-TATCC-----CCAAT |
| L. anguinea | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. attenuata | TAATGTA-TTAAGAAAGAATCTTT-GAATT-----CAAGGA-C----CC-----CAAT |
| L. aurioliae | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. bolusii | TAATGTA-TTAAGAAAGAATCTTT-GAATT-----CAAGGA-C----CC-----CAAT |
| L. bowkeri | TAATGTA-TTAAGAAAGAATCTTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. bifolia | ???G-----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. capensis | TAGTGTA-TTAATC---AATTCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. cernua | TAATGTA-TTAAG----AATTCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. comptonii | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. congesta | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. corymbosa | TAATGTA-TTAAG----AATCGTT-GAATT-----CAAAGAGC-TATCC-----CCAAT |
| L. doleritica | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATTC-----CCAAT |
| L. ensifolia subsp. ensifolia | TA-TG-A-TCAC----AA-C-TT-CC-----CCGGGAGC-TATCC-----CCCTT |
| L. ensifolia var. maughanii | TAATGTA-TTAAG----AATCGTT-GAATT-----CAAAGAGC-TATGGGCCCC-CCAAT |
| L. framesii | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. gillettii | TATTAAC-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. haarlemensis | TA---T--TTAAGAAAGAATCTTT-GAATT-----CAAGGAGC-TATCC-----CCA-- |
| L. hirta | TAATGTA-TTAAT----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. isopetala | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. magentea | TAATGTA-TTAAG----AATTCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. juncifolia var. juncifolia | TATT-AAGATAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. karooica | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATTC-----CCAAT |
| L. kombergensis | TA-T-T--TTAAGAATGAATCTTT-GAAAT-----TCAAGAGC-TATCC-----CCAAT |
| L. longituba | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. longibracteata | TAATGTA-TTAAG----AATTCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. margaretea | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. martiniae | TAATGTATTTAAG----AATCTTT-GAATT-----CAAGGAGC-TATCCCCAC--CCAAT |
| L. mediana | ???G-----AATCCTT-GAATT-----CAAGGAGC-TATCCCCCACCACCAAT |
| L. moniliformis | TAATGTC-TTACG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. muirii | TAATGTA-TTAAG----AATTCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. multifolia | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TGTCC-----CCAAT |
| L. mutabilis | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCCCCC--CCAAT |

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| L. namaquensis | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCA-- |
| L. nardousbergensis | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. nervosa | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. obscura | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. orthopetala | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TGTCC-----CCAAT |
| L. paucifolia | TAATGTA-TTAAG----AATCGTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. perryae | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. perryae/unifolia | TAATGTA-TTAAG----ATCCTT-GAATT-----CAAGGAGC-TATCCC-----CCAAT |
| L. pusilla | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. pustulata | ???????????G----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. rosea | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. punctata | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. schelpei | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. sp | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. sp | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. splendida | TA---T--TTAAGAAAGAATCTTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. stayneri | TATT-AAGATAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. thomasiae | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. trichophylla subsp. trichophylla | TAATGTA-TTAAG----AATCGTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. undulata | TAATGTA-TTAAG----AATCTTT-GAATT-----CAAGGAGC-TATCCCCAC--CCAAT |
| L. unicolor | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. unifolia | TATT-AAGATAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. unifolia/hirta | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. valeriae | TAGTGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCA-- |
| L. variegata | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. verticillata | TAGTGTA-TTAAA----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. violacea | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. youngii | TAATGTA-TTAAG----AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| L. zeyheri | TAGTGTA-TTAAGA---AATCCTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| V. bracheata | TATTGTA-TTAAG----AATCGTT-GAATT-----CAAGGAGC-TATCC-----CCAAT |
| V. capensis | ?????????????????CCT-GGAACTAA---AAATGGGC-AATCCTGAG--CCAAA |
| W. bifolia | TATTGTA-TTAAG----AATCCTT-GAATT-----CAAGGTGC-TTTCC-----CCAAT |
| | |
| G. superba | -----TGTCTTAGTGTATACGAA----TCGTTGA-----AGG-AGCA--- |
| L. angelica | ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--- |
| L. alba | ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--- |
| L. flava | ----ATTTTG--TTTACGAAAACAAGCTATTCTATTGG-----GGACAGCT--- |
| L. ameliae | ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--- |
| L. anguinea | ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--- |
| L. attenuata | ----AGCTTG--TTTTCGTAAACAAG----CTATTGG-----GG--T----- |
| L. aurioliae | ----AGCTTG--TTTTCGTAAACAAA----ATATTGG-----GGACAGCT--- |

L. bolusii ---AGCTTG--TTTTCGTAAACAAG----CTATTGG-----GG--T-----
L. bowkeri ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGC---
L. bifolia ---ATTTTG--TTTACGAAAACAAGCTATTCTATTGG-----GGACAGCT--
L. capensis ---ATTTTG--TTTACGAGAACAAA----CTATTGG-----GGACAGCT--
L. cernua ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. comptonii ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. congesta ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGAGAGCC--
L. corymbosa ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. doleritica ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. ensifolia subsp. *ensifolia* ---CCCTTG--TTTGCGAAAACCAG----CTATTGG-----GGACAGCC--
L. ensifolia var. *maughanii* ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. framesii ---ATTTTGTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. gillettii ---AGCTTG--TTTTCGTAAACAAG----CTATTGG-----GGACAGCTAAG
L. haarlemensis ----TT--G--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. hirta ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. isopetala ATATTTGTTT--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. magentea ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. juncifolia var. *juncifolia* ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. karooica ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. kombergensis ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGAT--
L. longituba ---GTTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. longibracteata ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. margaretea ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. martiniae ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GTGGGGATAGCT--
L. mediana ---AGCTTG--TTTACGAAAACAAG----CTATTGGT---GGGGATAGATAGCC---
L. moniliformis ---ATTTTG--TTGACGAAAACAAG----CTATTGG-----GGACAGCC--
L. muirii ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. multifolia ---AGCTTG--TTTTCGTAAACAAG----CTATTGG-----GGTATTTTAT
L. mutabilis ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGTGGGGT--
L. namaquensis ----TT--GTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. nardousbergensis ---ATTTTGTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. nervosa AAATATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. obscura ---AGCTTG--TTTTCGTAAACAAG----ATATTGG-----GGACAGCT--
L. orthopetala ---AGCTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGTC--
L. paucifolia ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. perryae ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. perryae/unifolia ----TTTTG---TTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. pusilla ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. pustulata ---ATTTTGTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT--
L. rosea -ATTTTTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC--
L. punctata ---ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT--

L. schelpei ----ATTTTG--TTTACGAGAAACAAG----CTATTGG-----GGACAGCT---
L. sp ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. sp ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC---
L. splendida ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. stayneri ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC---
L. thomasiae ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. trichophylla subsp. *trichophylla* ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC---
L. undulata ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GTGGGGATAGCT---
L. unicolor ----ATTTTGTTTTTACGAAAACAAG----CTATTGG-----GGATAGCT---
L. unifolia ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC---
L. unifolia/hirta ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCC---
L. valeriae ----TT--GTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. variegata ----ATTTTG--TTTACGAGAAACAAG----CTATTGG-----GGACAGCT---
L. verticillata ----AGCTTG--TTTACGAAAACAAG----CTATTGG-----G-----
L. violacea ----ATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. youngii -ATTATTTTG--TTTACGAAAACAAG----CTATTGG-----GGACAGCT---
L. zeyheri ----ATTTTGTTTTTACGAAAACAAG----CTATTGG-----GGACAGCT---
V. bracheata ----ATTTTG--TTTACGAAAACAGG----CTATTGG-----GGACAGCC---
V. capensis ----TCTTTATTTTTAGAAAACAAG----GGTTTAA-----AAACTAGA---
W. bifolia ----GTTTTG--TTTACAAAACAAG----CTATTGG-----GGACAGCC---

G. superba ----A-----TACC-----AA-----ACCTACTCC----
L. angelica ----C-----C---TGCC-AATGAATGATTAA-----CTATTTATTCC----
L. alba ----C-----C---TGCC-AATGAATGATTAG-ATTAAGTATCTATTTATTCC----
L. flava ----C-----C---TGCC-AATGAATGATTAAATTAATTAATTAATTTATTCC----
L. ameliae ----C-----C---TGCC-AATGAATGATTAA-----G---TATTTATTCC----
L. anguinea ----C-----T---CGCC-AATGAATGATTAA-----CTATTTATTCC----
L. attenuata ----C-----C---TGCC-AATGAATGATTAA----GTTAAGTATTTATTCC----
L. auriloliae ----C-----C---TGCC-AATGAATGATTAA-----TTAAGTATTTATTCC----
L. bolusii ----C-----C---TGCC-AATGAATGATTAA----GTTAAGTATTTATTCC----
L. bowkeri ----C-----C---TGCC----AATGATTAA-----GTATTTATTCC----
L. bifolia ----C-----C---TGCC-AATGAATGATTAAATTAATTAATTAATTTATTCC----
L. capensis ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC----
L. cernua ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC----
L. comptonii ----C-----C---TGCC-----
L. congesta ----C-----C---TGCC-AATGAATGATTAA-----GTATTTATTCC----
L. corymbosa ----CCTCC--C---TGCC-AATGAATGATTAA-----GTATCTATTTATTCC----
L. doleritica ----C-----C---TGCC-AATGAATGATTAG-ATTAAGTATCTATTTATTCC----
L. ensifolia subsp. *ensifolia* ----C-----C---TGCC-CATGAATGATTAA----ATTAATTTAATCT----
L. ensifolia var. *maughanii* ----C-----C---TGCC-AATGAATGATTAA-----CTATT---CC----
L. framesii ----C-----C---TGCC-AATGAATGATTAA-----GTATCTTCC----

L. gillettii ATTTAT-----TTTATCCCAATGAATGATAAT-----GATTAACCA-----
L. haarlemensis ----C-----C---TGCC-----TTAATTAA-----GTATTTATTCC-----
L. hirta ----C-----C---TGCC-AATGAATGATTAA-----CTATTTATTCC-----
L. isopetala ----CCTTC--C---TGCC-AATCAATGAATGA-----AATTTATTTATTCC-----
L. magentea ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC-----
L. juncifolia var. *juncifolia* ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. karooica ----C-----C---TGCC-AATGAATGATTAG-ATTAAGTATCTATTTATTCC-----
L. kombergensis ----CAA-----TGCC-----AATGATTAA-----GTATTTATTCC-----
L. longituba ----C-----C---TGCC-AATGAATGATTAA-----GTATCTATTTATTCC-----
L. longibracteata ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC-----
L. margaretea ----C-----C---TGCC-AATGAATGATTAA----ATTAAGTATTTATTCC-----
L. martiniae ----C-----C---TGCC-AATGAATGATTAA-----TATTCC-----
L. mediana ----C-----C---TGCC-AATGAATGATTAA-----GTATCTATTTATTCCAAACC
L. moniliformis ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. muirii ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC-----
L. multifolia T---C-----C---TGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC-----
L. mutabilis ----C-----C---TGCC-AATGAATGATTAA----ATTAAGTATTTATTCC-----
L. namaquensis ----C-----C---TGCC-AATGAATGATTAA-----GTAT--CTTCCAAATC
L. nardousbergensis ----C-----C---TGCC-AATGAATGATTAA-----GTAT--CTTCCAAATT
L. nervosa ----C-----C---TGCC-GATTAAT-----
L. obscura ----C-----C---TGCC-AATGAATGATTAA-----TTAAGTATTTATTCC-----
L. orthopetala ----C-----TGCC-AATGAATGATTAA-ATTAAGTATGTATTTATTCC-----
L. paucifolia ----CCTCC--C---TGCC-AATGAATGATTAA-----GTATCTATTTATTCC-----
L. perryae ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. perryae/unifolia ----C-----C---TGCCAAATGAATGATTAA-----CTATTTAATTT-----
L. pusilla ----C-----CAATTGCC-AATGAATGATTAA-ATTAAGTATCTATTTATTCC-----
L. pustulata ----C-----C---TGCC-AATGAATGATTAA-----GTAT--CTTCC-----
L. rosea ----C-----C---TGCC-----
L. punctata ----C-----C---TGCC-AATGAATGATTAA-----ATTAAGTATTTATTCC-----
L. schelpei ----C-----C---T-----TAATTAA-----TTATTCC-----
L. sp ----C-----C---TGCC-AATGAATGATTAA-----ATTAAGTATTTATTCC-----
L. sp ----C-----C---TGCC-AATGAATGATTAA-----AATTTATTCC-----
L. splendida ----C-----C---TGCC-----AATGATTAA-----GTATTTATTCC-----
L. stayneri ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. thomasiae ----C-----C---TGCC-AATGAATGATTAA-ATTAAGTAT----TTATTCC-----
L. trichophylla subsp. *trichophylla* ----C-----C---TGCC-AATGAATGATTAA-----CTATTTATTCC-----
L. undulata ----C-----C---TGCC-AATGAATGATTAA-----TATTCC-----
L. unicolor ----C-----C---TGCC-AATGAATGATTAA-----GTAT--CTTCC-----
L. unifolia ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. unifolia/hirta ----C-----C---TGCC-AATGAATGATTAA-----CTATTTAATTT-----
L. valeriae ----C-----C---TGCC-AATGAATGATTAA-----GTATCTTCC-----

L. variegata ----C-----C---TGCC-AATGAATGATTAA----ATTAAC TATTATTCC----
L. verticillata -----GCC-AATGAATGATTAA-ATTAAGTATCTATTATTCC----
L. violacea ----CCTCCCTCTCTGCC-AATGAATGATTAA-----ATTAAC TTTCC----
L. youngii ----C-----C---TGCC-AATGAATGATAAT---GATTAAGTATT--AT-----
L. zeyheri ----C-----C---TGCC-AATGAATGATTAA-----GTAT--CTCC----
V. bracheata ----C-----C---TGCC-AATGAATGAATGA--TTAAGTATCTATTATTCC----
V. capensis ----C-----T---AGAATAAAAAGGGATAGG----TGCAGAGACTCAATGG----
W. bifolia ----C-----C---TGCC-----GTATCTATTATTCC----

G. superba ---T----TTAGCAGTAGGTTTGGTATTGCTCCCT-----T
L. angelica A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. alba A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. flava A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. ameliae A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. anguinea A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. attenuata A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. aurioliae A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. bolusii A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. bowkeri A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. bifolia A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. capensis A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. cernua A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. comptonii A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. congesta A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. corymbosa A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. doleritica A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. ensifolia subsp. ensifolia A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. ensifolia var. maughanii T-AAC----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. framesii A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. gillettii --AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. haarlemensis A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. hirta G-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. isopetala A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. magentea A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. juncifolia var. juncifolia CTAAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. karooica A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. kombergensis A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. longituba A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. longibracteata A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. margaretea A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T
L. martiniae A-AAT----TAACGACGAGATTATTATCGTTTCTCGCATG-----T

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| L. mediana | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. moniliformis | AGAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. muirii | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. multifolia | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. mutabilis | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. namaquensis | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. nardousbergensis | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. nervosa | --AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. obscura | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. orthopetala | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. paucifolia | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. perryae | ATAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. perryae/unifolia | AGAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. pusilla | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. pustulata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. rosea | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. punctata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. schelpei | A-AAT-----TAACGAGGAGATTTATTATCGTTTCTCGCATG-----T |
| L. sp | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. sp | A-AAT-----TAACGACGAGATTTATTATCTTTTCTCGCATC-----T |
| L. splendida | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. stayneri | CTAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. thomasiae | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. trichophylla subsp. trichophylla | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. undulata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. unicolor | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. unifolia | CTAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. unifolia/hirta | ATAAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. valeriae | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. variegata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. verticillata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. violacea | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. youngii | --AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| L. zeyheri | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| V. bracheata | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| V. capensis | A-AGCTGTTCTAACGAATAGAGTTGACTACGTTGCGTTAATAACAGGAACAGGAATCCTT |
| W. bifolia | A-AAT-----TAACGACGAGATTTATTATCGTTTCTCGCATG-----T |
| | |
| G. superba | TTTGTTGGATTTCAGTCATATG---GTGT-----TTTG----- |
| L. angelica | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTG----- |
| L. alba | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTG----- |

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| <i>L. flava</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. ameliae</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. anguinea</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. attenuata</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. aurioliae</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. bolusii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. bowkeri</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. bifolia</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. capensis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. cernua</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. comptonii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. congesta</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. corymbosa</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. doleritica</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. ensifolia</i> var. <i>maughanii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. framesii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. gillettii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. haarlemensis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. hirta</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. isopetala</i> | CTCGCGAAAGTCAG--AGTCG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. magentea</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. karooica</i> | CTCGAGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. kombergensis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. longituba</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. longibracteata</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. margaretea</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. martiniae</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. mediana</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. moniliformis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. muirii</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. multifolia</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. mutabilis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. namaquensis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. nardousbergensis</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. nervosa</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. obscura</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. orthopetala</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. paucifolia</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| <i>L. perryae</i> | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |

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| L. perryae/unifolia | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. pusilla | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. pustulata | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. rosea | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. punctata | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. schelpei | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. sp | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. sp | CTCGCGAAACGCAG--AGTAG---GCACGAATTCT---CCCAACTTG----- |
| L. splendida | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. stayneri | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. thomasiae | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. trichophylla subsp. trichophylla | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. undulata | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. unicolor | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. unifolia | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. unifolia/hirta | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. valeriae | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. variegata | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. verticillata | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. violacea | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. youngii | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| L. zeyheri | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| V. bracheata | CTCGCGAAAGTAAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| V. capensis | CTATCGAAATTAAGAAAAAGAAAGGGTGACCTATATATCTAATACGTACGTATACATAC |
| W. bifolia | CTCGCGAAAGTCAG--AGTAG---GCGCGAATTCT---CCCAATTTG----- |
| | |
| G. superba | CGCACTAAACACAC-----ATATAAATATAAACACAC |
| L. angelica | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. alba | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. flava | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. ameliae | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. anguinea | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. attenuata | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. aurioliae | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. bolusii | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. bowkeri | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. bifolia | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. capensis | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. cernua | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. comptonii | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| L. congesta | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |

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| <i>L. corymbosa</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. doleritica</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAAGAAAATGT |
| <i>L. ensifolia</i> var. <i>maughanii</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. framesii</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. gillettii</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. haarlemensis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. hirta</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. isopetala</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. magentea</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. karooica</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. kombergensis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. longituba</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. longibracteata</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. margaretea</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. martiniae</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. mediana</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. moniliformis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. muirii</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. multifolia</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. mutabilis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. namaquensis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. nardousbergensis</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. nervosa</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. obscura</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. orthopetala</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. paucifolia</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. perryae</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. perryae/unifolia</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. pusilla</i> | TGACCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. pustulata</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. rosea</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. punctata</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. schelpei</i> | CGA-CCCACCATAGGATG-----TGTTATATAAATAGGAAAATGT |
| <i>L. sp</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. sp</i> | TGA-CCCACCATACAATC-----TGTTATATAAATAAGAAAAAGT |
| <i>L. splendida</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. stayneri</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. thomasiae</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |
| <i>L. trichophylla</i> subsp. <i>trichophylla</i> | TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT |

L. undulata TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. unicolor TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. unifolia TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. unifolia/hirta TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. valeriae TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. variegata TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. verticillata TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. violacea TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. youngii TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
L. zeyheri TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
V. bracheata TGA-CCCACCATACGATC-----TGTTATATAAATAGGAAAATGT
V. capensis TGA-CATATCAAACGATTAATCATGACCCGAATCCATATATTATATACATGTATATGC
W. bifolia TGA-CCCACCATACGATC-----CGTTATATAAATAGGAAAATGT

G. superba ---ATA-TATC-----TTTTAGGTGT---
L. angelica ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. alba ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. flava ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. ameliae ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. anguinea ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. attenuata ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. aurioliae ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. bolusii ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. bowkeri ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. bifolia ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. capensis ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. cernua ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. comptonii ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. congesta ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. corymbosa ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. doleritica ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. ensifolia subsp. ensifolia ---TCC-TTTCATTATGAATAGGGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. ensifolia var. maughanii ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. framesii ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. gillettii ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. haarlemensis ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. hirta ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. isopetala ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. magentea ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. juncifolia var. juncifolia ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---
L. karooica ---TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT---

L. kombergensis ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. longituba ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. longibracteata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. margaretea ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. martiniae ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. mediana ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. moniliformis ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. muirii ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. multifolia ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. mutabilis ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. namaquensis ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. nardousbergensis ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. nervosa ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. obscura ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. orthopetala ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. paucifolia ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCT-TTTTGGGTAT----
L. perryae ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. perryae/unifolia ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. pusilla ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. pustulata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. rosea ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. punctata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. schelpei ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCAGTTTGGGTAT----
L. sp ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. sp ----TCC-TTTCATTATGAATAACGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. splendida ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. stayneri ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. thomasiae ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. trichophylla subsp. *trichophylla* ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. undulata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. unicolor ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. unifolia ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. unifolia/hirta ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. valeriae ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. variegata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. verticillata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. violacea ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. youngii ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
L. zeyheri ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
V. bracheata ----TCC-TTTCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----
V. capensis AATATTA-TATGTAATATGCGCAATATATGCAAAATTCAGAGTTA-TTGTGGATCTATTC

W. bifolia ----TCC-TTCCATTATGAATAGCGATTGTATGGCC---AATCA-TTTTGGGTAT----

G. superba -A---TAAATG-----AAATT-----

L. angelica -AACGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. alba -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. flava -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. ameliae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. anguinea -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. attenuata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. aurioliae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. bolusii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. bowkeri -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. bifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. capensis -AATGGTATATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. cernua -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. comptonii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. congesta -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. corymbosa -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. doleritica -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. ensifolia subsp. ensifolia -AATGGGAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCCCTCCCTCATGTTGA

L. ensifolia var. maughanii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. framesii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. gillettii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. haarlemensis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. hirta -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. isopetala -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. magentea -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. juncifolia var. juncifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. karooica -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. kombergensis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. longituba -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. longibracteata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. margaretea -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. martiniae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. mediana -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. moniliformis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. muirii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. multifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. mutabilis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. namaquensis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. nardousbergensis -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

L. nervosa -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. obscura -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. orthopetala -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. paucifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. perryae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. perryae/unifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. pusilla -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. pustulata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. rosea -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. punctata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. schelpei -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. sp -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. sp -AATGGAAAATGCCCGAGGACACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGCGGA
L. splendida -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. stayneri -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. thomasiae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. trichophylla subsp. *trichophylla* -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. undulata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. unicolor -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. unifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. unifolia/hirta -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. valeriae -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. variegata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. verticillata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. violacea -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. youngii -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
L. zeyheri -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
V. bracheata -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA
V. capensis CAATCGAAGTTGACGGAAGAATCGAATATTCAGTGATCAAATCATTTATCCAGAGTTTA
W. bifolia -AATGGTAGATGCCCGAG---ACCAAGTTACTATTATTTCTTTCTCCTCCCTCATGTTGA

G. superba -----AAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. angelica GTTTTTCATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. alba GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. flava GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. ameliae GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. anguinea GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. attenuata GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. aurioliae GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. bolusii GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT
L. bowkeri GTTTTTCAATTTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT

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|---------------------------------------------|------------------------------------------------------|
| <i>L. bifolia</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. capensis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. cernua</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. comptonii</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. congesta</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. corymbosa</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. doleritica</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | GTTTTCAATTTTCTAGAAAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. ensifolia</i> var. <i>maughanii</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. framesii</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. gillettii</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. haarlemensis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. hirta</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. isopetala</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. magentea</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. karooica</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. kombergensis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. longituba</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. longibracteata</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. margaretea</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. martiniae</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. mediana</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. moniliformis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. muirii</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. multifolia</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. mutabilis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. namaquensis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. nardousbergensis</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. nervosa</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. obscura</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. orthopetala</i> | TTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. paucifolia</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. perryae</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. perryae/unifolia</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. pusilla</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. pustulata</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. rosea</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. punctata</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. schelpei</i> | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| <i>L. sp</i> | GTTTTCAATTTTCTAGAAAAATGATTAGCT-----ACAAAAGGATTTTTTTT |

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| L. sp | TTTTTCAATTTTCTATATAAAAGATTAGCT-----ACTAAAGGATTTTTTTT |
| L. splendida | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. stayneri | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. thomasiae | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. trichophylla subsp. trichophylla | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. undulata | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. unicolor | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. unifolia | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. unifolia/hirta | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. valeriae | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. variegata | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. verticillata | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. violacea | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. youngii | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| L. zeyheri | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| V. bracheata | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| V. capensis | ATAGACCCCTTTTTTGA AAAACTGATTAATCGGACGAGAATAAAGAGAGAGTCCCCTTC |
| W. bifolia | GTTTTCAATTTTCTAGATAAATGATTAGCT-----ACAAAAGGATTTTTTTT |
| | |
| G. superba | TAGTGAAC-GTGTCACGGCGGATTACTCC--TTTTTTTACATTAT-TAAAATG-GGT-A |
| L. angelica | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. alba | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. flava | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. ameliae | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. anguinea | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. attenuata | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. aurioliae | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. bolusii | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. bowkeri | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. bifolia | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. capensis | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. cernua | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. comptonii | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. congesta | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. corymbosa | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. doleritica | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. ensifolia subsp. ensifolia | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. ensifolia var. maughanii | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. framesii | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. gillettii | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| L. haarlemensis | TAGTGAAC-GTGTCACAGCCGATTACTCC--TTTTTTTACATTTG-AAAGATT-GGC-A |

L. hirta TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. isopetala TAGTGAAC-GTGTACACA---GATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. magentea TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. juncifolia var. *juncifolia* TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. karooica TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. kombergensis TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. longituba TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. longibracteata TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. margaretea TAGTGAAC-GTGTACAGCCGATTACTCC---TTTTTTTACATTTT-AAAGATT-GGC-A
L. martiniae TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. mediana TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTCCATTTT-AAAGATT-GGC-A
L. moniliformis TAGTGAAC-GTGTACAGCCGATTACTCC---TTTTTTTACATTGT-AAAGATT-GGC-A
L. muirii TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. multifolia TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. mutabilis TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. namaquensis TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. nardousbergensis TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. nervosa TAGTGAAC-GTGTACAGCC--TACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. obscura TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. orthopetala TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTG-AAAGATT-GCC-A
L. paucifolia TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. perryae TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. perryae/unifolia TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. pusilla TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. pustulata TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. rosea TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. punctata TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. schelpei TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. sp TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. sp TTTTGTACGGTGGCACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. splendida TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. stayneri TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. thomasiae TAGTGAAC-GTGTACAGCCGATTACTCC---TTTTTTTACATTTT-AAAGATT-GGC-A
L. trichophylla subsp. *trichophylla* TAGTGAAC-GTGTACAGCCGATTACTCC---TTTTTTTACATTTT-AAAGATT-GGC-A
L. undulata TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. unicolor TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. unifolia TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. unifolia/hirta TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. valeriae TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A
L. variegata TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTTACATTTT-AAAGATT-GGC-A
L. verticillata TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTTACATTTT-AAAGATT-GGC-A

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|--------------------------------------|--------------------------------------------------------------|
| <i>L. violacea</i> | TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTACATTTT-AAAGATT-GGC-A |
| <i>L. youngii</i> | TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTACATTTT-AAAGATT-GGC-A |
| <i>L. zeyheri</i> | TAGTGAAC-GTGTACAGCCGATTACTCC-TTTTTTTTACATTTT-AAAGATT-GGC-A |
| <i>V. bracheata</i> | TAGTGAAC-GTGTACAGCCGATTACTCC--TTTTTTTACATTTT-AAAGATT-GGC-A |
| <i>V. capensis</i> | T---ACA-TGTCAATACCGACAACAATGAAATTTATAGTAAGAGG-AAAATCCGTCGACT |
| <i>W. bifolia</i> | TAGTGAAC-GTGTACAGCCGATTACTCCTTTTTTTTTTCTTTTA-AAAGATT-GGA-A |
| <i>G. superba</i> | TTCTATG-CCCAATA??????AAGAAGAAGGGAAGGAGAGAGGGAAGGGAGGAAGAAGA |
| <i>L. angelica</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. alba</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. flava</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. ameliae</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. anguinea</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. attenuata</i> | TTCTATG-TCCAATAGAATATAAAGGGAGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. aurioliae</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. bolusii</i> | TTCTATG-TCCAATAGAATATAAAGGGAGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. bowkeri</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. bifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. capensis</i> | TTCTATG-TCCAATAAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. cernua</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. comptonii</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. congesta</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. corymbosa</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. doleritica</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. ensifolia subsp. ensifolia</i> | TTCTATG-TCCAATAGAATATAGAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. ensifolia var. maughanii</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>L. framesii</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. gillettii</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. haarlemensis</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. hirta</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. isopetala</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGGAGA |
| <i>L. magentea</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. juncifolia var. juncifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. karooica</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>L. kombergensis</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. longituba</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. longibracteata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. margaretea</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>L. martiniae</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. mediana</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. moniliformis</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |

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|---------------------------------------------------|----------------------------------------------------------------|
| <i>L. muirii</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. multifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. mutabilis</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. namaquensis</i> | TTATATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. nardousbergensis</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. nervosa</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAGGA |
| <i>L. obscura</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. orthopetala</i> | TTGTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. paucifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. perryae</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. perryae/unifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. pusilla</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGAAAGGGAGGGAAGAGAAGAAAG |
| <i>L. pustulata</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. rosea</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. punctata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. schelpei</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGAGAAGAGAAGAAGA |
| <i>L. sp</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. sp</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAGGAAAAGAAAAGA |
| <i>L. splendida</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. stayneri</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. thomasiae</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>L. trichophylla</i> subsp. <i>trichophylla</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>L. undulata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. unicolor</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. unifolia</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. unifolia/hirta</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. valeriae</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. variegata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>L. verticillata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. violacea</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAAGGGAAGAGAAGAAAG |
| <i>L. youngii</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>L. zeyheri</i> | TTCTATG-TCCAATAGAATATAAAGAAGAAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAG |
| <i>V. bracheata</i> | TTCTATG-TCCAATAGAATATAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAGA |
| <i>V. capensis</i> | TTAGAAATCGTGAGGGTTCAAGAGAAAGAGAAAAAAGAAAAAGAAAAAGAAAAAGAAAA |
| <i>W. bifolia</i> | TTCTATGCCCAATAAAAATTAAAGAAGGAGGAGGGAGGGAGGGAGGGAAGAGAAGAAAA |
| <i>G. superba</i> | AAGGGGGA |
| <i>L. angelica</i> | AAGGGGAG |
| <i>L. alba</i> | AAGGGGAG |
| <i>L. flava</i> | AAGGGGAG |
| <i>L. ameliae</i> | AAGGGGAG |

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|---------------------------------------------|----------|
| <i>L. anguinea</i> | AAGGGGAG |
| <i>L. attenuata</i> | AAGGGGAG |
| <i>L. aurioliae</i> | AAGGGGAG |
| <i>L. bolusii</i> | AAGGGGAG |
| <i>L. bowkeri</i> | AAGGGGAG |
| <i>L. bifolia</i> | AAGGGGAG |
| <i>L. capensis</i> | AAGGGGAG |
| <i>L. cernua</i> | AAGGGGAG |
| <i>L. comptonii</i> | AAGGGGAG |
| <i>L. corymbosa</i> | AAGGGGAG |
| <i>L. doleritica</i> | AAGGGGAG |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | AAGGGGAG |
| <i>L. ensifolia</i> var. <i>maughanii</i> | AAGGGGAG |
| <i>L. framesii</i> | AAGGGGAG |
| <i>L. gillettii</i> | AAGGGGAG |
| <i>L. haarlemensis</i> | AAGGGGAG |
| <i>L. hirta</i> | AAGGGGAG |
| <i>L. isopetala</i> | AAGGGGAG |
| <i>L. magentea</i> | AAGGGGAG |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | AAGGGGAG |
| <i>L. karooica</i> | AAGGGGAG |
| <i>L. kombergensis</i> | AAGGGGAG |
| <i>L. longituba</i> | AAGGGGAG |
| <i>L. longibracteata</i> | AAGGGGAG |
| <i>L. margaretea</i> | GAGGGGAG |
| <i>L. martiniae</i> | AAGGGGAG |
| <i>L. mediana</i> | AAGGGGAG |
| <i>L. moniliformis</i> | GAGGGGAG |
| <i>L. muirii</i> | AAGGGGAG |
| <i>L. multifolia</i> | AAGGGGAG |
| <i>L. mutabilis</i> | AAGGGGAG |
| <i>L. namaquensis</i> | AAGGGGAG |
| <i>L. nardousbergensis</i> | AAGGGGAG |
| <i>L. nervosa</i> | AAGGGGAG |
| <i>L. obscura</i> | AAGGGGAG |
| <i>L. orthopetala</i> | AAGGGGAG |
| <i>L. paucifolia</i> | AAGGGGAG |
| <i>L. perryae</i> | AAGGGGAG |
| <i>L. perryae/unifolia</i> | AAGGGGAG |
| <i>L. pusilla</i> | AAGGGGAG |
| <i>L. pustulata</i> | AAGGGGAG |

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|-------------------------------------|----------|
| L. rosea | AAGGGGAG |
| L. punctata | AAGGGGAG |
| L. schelpei | AAGGGGAG |
| L. sp | AAGGGGAG |
| L. sp | AAGGGGAG |
| L. splendida | AAGGGGAG |
| L. stayneri | AAGGGGAG |
| L. thomasiae | GAGGGGAG |
| L. trichophylla subsp. trichophylla | GAGGGGAG |
| L. undulata | AAGGGGAG |
| L. unicolor | AAGGGGAG |
| L. unifolia | AAGGGGAG |
| L. unifolia/hirta | AAGGGGAG |
| L. valeriae | AAGGGGAG |
| L. variegata | AAGGGGAG |
| L. verticillata | AAGGGGAG |
| L. violacea | AAGGGGAG |
| L. youngii | AAGGGGAG |
| L. zeyheri | AAGGGGAG |
| V. bracheata | AAGGGGAG |
| V. capensis | AGAAAAAG |
| W. bifolia | AAGGGAAG |

Appendix F: Aligned sequences of the combined *TrnL-F* and indels dataset of all the specimens used for cladistics analysis. *E.* = *Eucomis*, *G.* = *Gloriosa*, *M.* = *Massonia*, *L.* = *Lachenalia*, *V.* = *Veltheimia*. *W.* = *Whiteheadia bifolia*.

| | |
|---------------------------------------------|---------------------------------------------------------------|
| <i>E. bicolor</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>E. vandermerwei</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. alba</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. flava</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTGTTAGAAAAACAA---G |
| <i>L. ameliae</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. angelica</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. anguinea</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. attenuata</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. aurioliae</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. bolusii</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. bowkeri</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. bifolia</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. capensis</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. cernua</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. comptonii</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. concordiana</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. congesta</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. contaminata</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. convallarioides</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. corymbosa</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. dehoopensis</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. doleritica</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. duncanii</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAACAAG |
| <i>L. ensifolia</i> var. <i>maughanii</i> | CCCTGGAACTAAAAATGGGCAATCCCTGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. fistulosa</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. framesii</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. gillettii</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. haarlemensis</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. hirta</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. inconspicua</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. isopetala</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. magentea</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAA-CAA---G |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAA-CAA---G |
| <i>L. karooica</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. kombergensis</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |
| <i>L. arbuthnotiae</i> | CCCTGGAACTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAGAAAAACAA---G |

L. longituba CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. margaretea CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. martiniae CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. mathewsii CCCGGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. maximiliani CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. mediana CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. moniliformis CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAA-CAA---G
L. muirii CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. multifolia CCCAGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. mutabilis CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. namaquensis CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. namibiensis CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. nervosa CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. obscura CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. paucifolia CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. perryae CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. perryae/unifolia CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. polypodantha CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAA-----
L. pusilla CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. pustulata CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. rosea CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. punctata CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. salteri CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. schelpei CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. sp CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. splendida CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. stayneri CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. thomasiae CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. trichophylla CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. undulata CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. unicolor CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. unifolia CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAA-CAA---G
L. unifolia/hirta CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. valeriae CCCCGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. variegata CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. verticillata CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. violacea CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. youngii CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. zebrina CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
L. zeyheri CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G
M. depressa CCCTGGAAGTAAAAATGGGCAATCC-TGAGCCAAATCTTTATTTTGTAAAAACAA---G

V. bracheata CCCTGGAACATAAAAATGGGCAATCC-TGAGCCAATCTTTATTTTTAGAAAAACAA---G
V. capensis CCCTGGAACATAAAAATGGGCAATCC-TGAGCCAATCTTTATTTTTAGAAAAACAA---G
W. bifolia CCCTGGAACATAAAAATGGGCAATCC-TGAGCCAATCTTTATTTTTAGAAAAACAA---G

E. bicolor GGTTTA---AAAT----ACTAGAATAAAAAATAAAAA-----GGGATAGGTGCAGAGACT
E. vandermerwei GGTTTA---AAAT----ACTAGAATAAAAAATAAAAA-----GGGATAGGTGCAGAGACT
L. alba GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. flava GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. ameliae GGTTTA---AAAG----ACTAGAATAAAAAA-----G---GGGATAGGTGCAGAGACT
L. angelica GGTTTA---AAA-----ACTAGACTAGAATAAAAA-----GGGATAGGTGCAGAGACT
L. anguinea GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. attenuata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. aurioliae GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. bolusii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. bowkeri GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. bifolia GGTAA---AAAAAAA---ACTAGACTAGAATAAAAAAAA---GGGATAGGTGCAGAGACT
L. capensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. cernua GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. comptonii GGTTTA---AAA-----CTAGAATAAAAAA-----GGGATAGGTGCAGAGACT
L. concordiana GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. congesta GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. contaminata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. convallarioides GGTTTA---AAA-----ACTAGACTAGAATAAAAAA-----GGGATAGGTGCAGAGACT
L. corymbosa GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. dehoopensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. doleritica GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGATAGGTGCAGAGACT
L. duncanii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. ensifolia subsp. ensifolia GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. ensifolia var. maughanii GGTTTA---AAAAA---AAAAAATAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. fistulosa GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. framesii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. gillettii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. haarlemensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. hirta GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. inconspicua GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. isopetala GGTTCATAAAA---ACTAGACTAGAATAAAAA-----GGGATAGGTGCAGAGACT
L. magentea GGTTTA---AAA-----ACTAGACTAGAATAAAAAA-----GGGATAGGTGCAGAGACT
L. juncifolia var. juncifolia GGTTTA---AAA-----ACTAGACTAGAATAAAAAA-----GGGATAGGTGCAGAGACT
L. karooica GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. kombergensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA-----GGGATAGGTGCAGAGACT
L. arbutnotiae GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAA---GGGATAGGTGCAGAGACT

L. longituba GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---AGGATAGGTGCAGAGACT
L. margaretea GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. martiniae GGTTTA---AAAG---ACTAGAATAAAAAAGGGATA---GGGATAGGTGCAGAGACT
L. mathewsii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. maximiliani GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. mediana GGTTTA---AAA-----ACTAGACTAGAATTAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. moniliformis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. muirii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. multifolia GGTTTA---AAAG---ACTAGAATAAAAAA-----GGGATAGGTGCAGAGACT
L. mutabilis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. namaquensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. namibiensis GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. nervosa GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. obscura GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. paucifolia GGTTTA---AAA-----ACTAGACTAGAATAAAA-AAAA---GGGATAGGTGCAGAGACT
L. perryae GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. perryae/unifolia GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. polypodantha --TTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. pusilla GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. pustulata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. rosea GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. punctata GGTTTA---AAA-----ACTAGA-----ATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. salteri GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAGGGGGATAGGTGCAGAGACT
L. schelpei GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. sp GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAAA---GGGATAGGTGCAGAGACT
L. splendida GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. stayneri GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. thomasiae GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. trichophylla GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAGGGGGATAGGTGCAGAGACT
L. undulata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. unicolor GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. unifolia GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. unifolia/hirta GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. valeriae GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. variegata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. verticillata GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. violacea GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. youngii GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAGGGGGATAGGTGCAGAGACT
L. zebrina GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
L. zeyheri GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT
M. depressa GGTTTA---AAA-----ACTAGACTAGAATAAAAAAAG---GGGATAGGTGCAGAGACT

| | |
|--------------------------------------|---------------------------------------------------------------|
| <i>V. bracheata</i> | GGTTTA---AAA-----ACTAGACTAGAATAAAAA-----GGGATAGGTGCAGAGACT |
| <i>V. capensis</i> | GGTTTA---AAA-----ACTAGACTAGAATAAAAA-----GGGATAGGTGCAGAGACT |
| <i>W. bifolia</i> | GGTTTA---AAA-----ACTAGACTAGAATAAAAA-----GGGATAGGTGCAGAGACT |
| <i>E. bicolor</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTG-----ATAA-CAGG- |
| <i>E. vandermerwei</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTG-----ATAA-CAGG- |
| <i>L. alba</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. flava</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATA-----GA |
| <i>L. ameliae</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. angelica</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. anguinea</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. attenuata</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA-----ATAA-CAGGA |
| <i>L. aurioliae</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA |
| <i>L. bolusii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. bowkeri</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA |
| <i>L. bifolia</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. capensis</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. cernua</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. comptonii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. concordiana</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. congesta</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. contaminata</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. convallarioides</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. corymbosa</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. dehoopensis</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. doleritica</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. duncanii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. ensifolia subsp. ensifolia</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. ensifolia var. maughanii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. fistulosa</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. framesii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |
| <i>L. gillettii</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA-----ATAA-CAG-- |
| <i>L. haarlemensis</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA |
| <i>L. hirta</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA |
| <i>L. inconspicua</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGAA |
| <i>L. isopetala</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTG-----ATAA--A--- |
| <i>L. magentea</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. juncifolia var. juncifolia</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. karooica</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA |
| <i>L. kombergensis</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA-----ATAA-CAGGA |
| <i>L. arbutnotiae</i> | CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA |

L. longituba CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. margaretea CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATATATAATAA-CAGGA
L. martiniae CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. mathewsii CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. maximiliani CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. mediana CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. moniliformis CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA
L. muirii CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. multifolia CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. mutabilis CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. namaquensis CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA----ATAA-CAGGA
L. namibiensis CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA
L. nervosa CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. obscura CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. paucifolia CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. perryae CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAAACAGGA
L. perryae/unifolia CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. polypodantha CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATT----ATAA-CAGGA
L. pusilla CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. pustulata CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA----ATAA-CAAGA
L. rosea CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. punctata CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTATAATA-A----ATAA-CAGGA
L. salteri CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. schelpei CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. sp CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. splendida CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA----ATAA-CAGGA
L. stayneri CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. thomasiae CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATATATAATAA-CAGGA
L. trichophylla CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. undulata CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. unicolor CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA----ATAA-CAGGA
L. unifolia CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. unifolia/hirta CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. valeriae CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTT-----ATA----ATAA-CAGGA
L. variegata CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. verticillata CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
L. violacea CAATGGAAGCTGTTCTAACGAATGGAGTTGATTACGTTGTGTTATA----ATAA-CAGGA
L. youngii CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTATA----ATAA-CAGGA
L. zebrina CAATGGAAGTTGTTCTAACGAATGGAGTTGACTACGTTGCGTTA-----ATAA-CA---
L. zeyheri CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGTGTTATA----ATAA-CAGGA
M. depressa CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTA-----ATAA-A---

V. bracheata CAATGGAAGCTGTTCTAACGAATAGAGTTGACTACGTTGCGTTA-----ATAA-CAGGA
V. capensis CAATGGAAGCTGTTCTAACGAATAGAGTTGACTACGTTGCGTTA-----ATAA-CAGGA
W. bifolia CAATGGAAGCTGTTCTAACGAATGGAGTTGACTACGTTGCGTTA-----ATAA-CAGG-

E. bicolor -----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
E. vandermerwei -----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. alba ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. flava ATA -----ATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. ameliae ATAGAATAAATAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. angelica ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. anguinea AT-GAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. attenuata ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. aurioliae ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. bolusii ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. bowkeri ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. bifolia ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA----ATATATCTA
L. capensis ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. cernua ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. comptonii ATAGAATAAATAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATTTA
L. concordiana ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. congesta AT-----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. contaminata ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. convallarioides ATA-----ATCAATCCTTCTATCGAAATTAAGAAAGGGTGGCCTA-----TATATCTA
L. corymbosa ATATAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. dehoopensis ATAGAATAATTAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. doleritica ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. duncanii ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. ensifolia subsp. *ensifolia* ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. ensifolia var. *maughanii* ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. fistulosa ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. framesii ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. gillettii ---GAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. haarlemensis ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. hirta AT-----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. inconspicua ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. isopetala -CAGG-----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. magentea AT-----AATCCTTCTATCGAAATTAAGAAAGGGGGACCTA-----TATATCTA
L. juncifolia var. *juncifolia* AT-----AATCCTTCTATCGAAATTAAGAAAGGGGGACCTA-----TATATCTA
L. karooica ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. kombergensis ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. arbutnotiae ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA

L. longituba ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. margaretea ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. martiniae ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. mathewsii ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACGTA-----TATATCTA
L. maximiliani ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. mediana ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. moniliformis AT-GAAT----AATCCTTCTATCGAAATTAAGAAAGGGGGACCTA-----TATATCTA
L. muirii ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. multifolia ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. mutabilis ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTATA--TATCTATCTA
L. namaquensis ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. namibiensis AT-----AATCCTTCTATCGAAATTAAGAAAGGGTGACGTA-----TATATCTA
L. nervosa ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGCGACCTA-----TATATCTA
L. obscura ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. paucifolia ATATAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. perryae ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. perryae/unifolia ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. polypodantha ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. pusilla ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. pustulata ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. rosea ATAGAATAAATAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. punctata ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA----ATATATCTA
L. salteri ATAGAATAAATAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. schelpei ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. sp ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. splendida ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. stayneri ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGCGACCTA-----TATATCTA
L. thomasiae ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. trichophylla ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. undulata ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. unicolor ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. unifolia AT-----AATCCTTCTATCGAAATTAAGAAAGGGGGACCTA-----TATATCTA
L. unifolia/hirta ATAGAAT----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. valeriae ATAGAAT----ATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. variegata ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. verticillata ATAGAAT-----TCCTTCTATTGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. violacea ATAGAAT----ATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. youngii ATAGAAT----AATCCTTCTATCGAAATGAAAGAAAGGGCGACCTA-----TATATCTA
L. zebrina --AGG-----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
L. zeyheri ATAGAATAATCAATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA
M. depressa -CAGG-----AATCCTTCTATCGAAATTAAGAAAGGGTGACCTA-----TATATCTA

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|---------------------------------------------|--------------------------------------------------------------|
| <i>V. bracheata</i> | ACAGG-----AATCCTTCTATCGAAATTAAGAAAAGGGTGACCTA-TATTATATATCTA |
| <i>V. capensis</i> | ACAGG-----AATCCTTCTATCGAAATTAAGAAAAAGAAAGGGTGACCTATATATCTA |
| <i>W. bifolia</i> | -----AATCCTTCTATCGAAATTAAGAAAAGGGTGACCTA-----TATATCTA |
| <i>E. bicolor</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>E. vandermerwei</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. alba</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. flava</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. ameliae</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATGATGACCCGAATCCATATAT-T |
| <i>L. angelica</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. anguinea</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. attenuata</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. aurioliae</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. bolusii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. bowkeri</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. bifolia</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. capensis</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATTATGACCCGAATCCATATAT-T |
| <i>L. cernua</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. comptonii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATGATGACCCGAATCCATATAT-T |
| <i>L. concordiana</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. congesta</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. contaminata</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. convallarioides</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. corymbosa</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. dehoopensis</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. doleritica</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. duncanii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. ensifolia</i> var. <i>maughanii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. fistulosa</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATTATGACCCGAATCCATATAT-T |
| <i>L. framesii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. gillettii</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. haarlemensis</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. hirta</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. inconspicua</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. isopetala</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATT-ATATATAT |
| <i>L. magentea</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. karooica</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. kombergensis</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>L. arbutnotiae</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |

L. longituba ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. margaretea ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. martiniae ATACGTACGTATACATACTGACATATCAAACAATTAATCATGACCCGAATCCATATAT-T
L. mathewsii ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. maximiliani ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. mediana ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. moniliformis ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. muirii ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. multifolia ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. mutabilis ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. namaquensis ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. namibiensis ATACGTACGTATACATACT-----ATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. nervosa ATACGTACGTATACATACTGACATATCAAACAATTAATCATGACCCGAATCCATATAT-T
L. obscura ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. paucifolia ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. perryae ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. perryae/unifolia ATACGTACATATACATACTGACATATCAAACGATTAATCATGATCCGAATCCATATAT-T
L. polypodantha ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. pusilla ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. pustulata ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. rosea ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. punctata ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. salteri ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. schelpei ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. sp ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. splendida ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. stayneri ATACGTACGTATACATACTGACATATCAAACAATTAATCATGACCCGAATCCATATAT-T
L. thomasiae ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. trichophylla ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. undulata ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. unicolor ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. unifolia ATACGTACGTATAC-----TGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. unifolia/hirta ATACGTACATATACATACTGACATATCAAACGATTAATCATGATCCGAATCCATATAT-T
L. valeriae ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. variegata ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. verticillata ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. violacea ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
L. youngii ATACGTACGTATACATACTGACATATCAAACAATTAATCATGACCCGAATCCATATAT-T
L. zebrina ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCC-----CCATATAT-T
L. zeyheri ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T
M. depressa ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T

| | |
|--------------------------------------|--------------------------------------------------------------|
| <i>V. bracheata</i> | ATACGTACGT-----TGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>V. capensis</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>W. bifolia</i> | ATACGTACGTATACATACTGACATATCAAACGATTAATCATGACCCGAATCCATATAT-T |
| <i>E. bicolor</i> | ATA--TATA---CATGTATATGCAATATTATATGCAATATGCGCAATATATGCAAAAT |
| <i>E. vandermerwei</i> | ATA--TATA---CATGTATATGCAATATTATATGCAATATGCGCAATATATGCAAAAT |
| <i>L. alba</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. flava</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. ameliae</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. angelica</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. anguinea</i> | ATA--TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. attenuata</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. aurioliae</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. bolusii</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. bowkeri</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. bifolia</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. cernua</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. comptonii</i> | -TA--TATA---CACGTATATGCAATAT-----ATGCAATATATGCAAAAT |
| <i>L. concordiana</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. congesta</i> | ATA--TATA---CACGTATATACAATAT-----ATGC-----AAAAT |
| <i>L. contaminata</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. convallarioides</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. corymbosa</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. dehoopensis</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. doleritica</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. duncanii</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. ensifolia subsp. ensifolia</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. ensifolia var. maughanii</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. fistulosa</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. framesii</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. gillettii</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. haarlemensis</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. hirta</i> | ATA--TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. inconspicua</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. isopetala</i> | ATA--TATA---CATGTATATGCAATAT-----GCGCAATATATGCAAAAT |
| <i>L. magentea</i> | ATA--TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. juncifolia var. juncifolia</i> | ATA--TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. karooica</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. kombergensis</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. arbutnotiae</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| <i>L. longituba</i> | ATA--TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |

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| L. margaretea | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. martiniae | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. mathewsii | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. maximiliani | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. mediana | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. moniliformis | ATA---TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| L. muirii | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. multifolia | ATA---TATA---CACGTATATGCGATAT-----ATGC-----AAAAT |
| L. mutabilis | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. namaquensis | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. namibiensis | ATA---TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| L. nervosa | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. obscura | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. paucifolia | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. perryae | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. perryae/unifolia | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. polypodantha | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. pusilla | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. pustulata | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. rosea | GCA---TATA---CGTGTGTATATATAATAT-----ATGC-----AAAAT |
| L. punctata | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. salteri | GCA---TATA---CGTGTATATATAATAT-----ATGC-----AAAAT |
| L. schelpei | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. sp | ATAATATATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. splendida | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. stayneri | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. thomasiae | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. trichophylla | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. undulata | GCA---TATA---CGTGTATATATAATAT-----ATGC-----AAAAT |
| L. unicolor | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. unifolia | ATA---TATACACGTATGTATATGCAATAT-----ATGC-----AAAAT |
| L. unifolia/hirta | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. valeriae | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. variegata | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. verticillata | ATA---TATA---CACGTATATACAATAT-----ATGC-----AAAAT |
| L. violacea | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. youngii | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| L. zebrina | ATA---TATA---CATGTATATGCAATAT-----ATGC-----AAAAT |
| L. zeyheri | ATA---TATA---CACGTATATGCAATAT-----ATGC-----AAAAT |
| M. depressa | ATA---TATA---CATGTATATGCAA-----AAT |
| V. bracheata | ATA---TATA---CATGTATATGCAATATTATATGTAATATGCGCAATATATGCAAAAT |

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| <i>V. capensis</i> | ATA---TATA---CATGTATATGCAATATTATATGTAATATGCGCAATATATGCAAAAAT |
| <i>W. bifolia</i> | ATA---TATA---CATGTATATGCAATAT-----GCGCAATATATGCAAAAAT |
| <i>E. bicolor</i> | TCGGAGTTATTGTGGATCTATTCCAATCGAAATTGAC---GGAAGAATCGAAT---AG |
| <i>E. vandermerwei</i> | TCGGAGTTATTGTGGATCTATTCCAATCGAAATTGAC---GGAAGAATCGAAT---AG |
| <i>L. alba</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. flava</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. ameliae</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. angelica</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. anguinea</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATC-AATATTCAG |
| <i>L. attenuata</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. aurioliae</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. bolusii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. bowkeri</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. bifolia</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. capensis</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. cernua</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. comptonii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. concordiana</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. congesta</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGATCA---AA |
| <i>L. contaminata</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. convallarioides</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. corymbosa</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. dehoopensis</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. doleritica</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. duncanii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. ensifolia subsp. ensifolia</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. ensifolia var. maughanii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. fistulosa</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. framesii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. gillettii</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. haarlemensis</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. hirta</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATC-AATATTCAG |
| <i>L. inconspicua</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. isopetala</i> | TCAGAGTTATTGTGGATCTATTCCACCAATCGAAGAC---GGAAGAATCTAATATTCAG |
| <i>L. magentea</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATC-AATATTCAG |
| <i>L. juncifolia var. juncifolia</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATC-AATATTCAG |
| <i>L. karooica</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. kombergensis</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. arbutnotiae</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |
| <i>L. longituba</i> | TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC---GGAAGAATCGAATATTCAG |

L. margaretea TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. martiniae TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. mathewsii TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. maximiliani TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. mediana TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. moniliformis TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATC-AATATTCAG
L. muirii TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. multifolia TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. mutabilis TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. namaquensis TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. namibiensis TCAGAG----ATGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. nervosa TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. obscura TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. paucifolia TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. perryae TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. perryae/unifolia TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. pusilla TC-----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. pustulata TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. rosea TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. punctata TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. salteri TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. schelpei TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. sp TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. splendida TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. stayneri TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. thomasiae TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. trichophylla TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. undulata TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. unicolor TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. unifolia TCAGAG----GTGGATCTATTCTATCGAAGTTGAC----GGAAGAATC-AATATTCAG
L. unifolia/hirta TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. valeriae TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. variegata TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. verticillata TCAGAG----GTGGATCTATTCCAATCGAAGTTGATTGATGGAAGAATCGAATATTCAG
L. violacea TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. youngii TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. zebrina TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
L. zeyheri TCAGAG----GTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
M. depressa TCAGAATTATTGTGGATCTATTCCAATCGAAGTTGAT----GGAAGAATCGAATATTCAG
V. bracheata TCAGAGTTATTGTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG
V. capensis TCAGAGTTATTGTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTCAG

W. bifolia TCAGAGTTATTGTGGATCTATTCCAATCGAAGTTGAC----GGAAGAATCGAATATTGAG

E. bicolor TGATCAAATCATT-----CCAG----AGTT----TAAT----AAACCC----TTT---
E. vandermerwei TGATCAAATCATT-----CCAG----AGTT----TAAT----AAACCC----TTT---
L. alba TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T
L. flava TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. ameliae TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. angelica TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T
L. anguinea TGATCAAATTATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. attenuata TGGTCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. aurioliae TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. bolusii TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. bowkeri TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. bifolia TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. capensis TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. cernua TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. comptonii TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---CTT--T
L. concordiana TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T
L. congesta TCAT-----TC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. contaminata TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. convallarioides TGATCAAAA---TC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. corymbosa TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. dehoopensis TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. doleritica TGATCAAAA---TC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. duncanii TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T
L. ensifolia subsp. ensifolia TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. ensifolia var. maughanii TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---CTT--T
L. fistulosa TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. framesii TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. gillettii TGATCAAATCATTC--ATTCCAG----AGTT----TAATAGATAGACCCC---TTT--T
L. haarlemensis TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. hirta TGATCAAATTATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. inconspicua TGATCAAATCATTC---T--AG----AGTT----TAAT----AGACCCC---TTT--T
L. isopetala TGATCAAATCATTC--ATTCCAG----AGTT--GTTTAAT----AGACCC---TTT--T
L. magentea TGATCAAATTATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. juncifolia var. juncifolia TGATCAAATTATTC--ATTCCAG----AGTT----TAAT----AGACCC---TTT--TT
L. karooica TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T
L. kombergensis TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. arbuthnotiae TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTT--T
L. longituba TGATCAAATCATTC--ATTCCAG----AGTT----TAAT----AGACCCC---TTTTTT
L. margaretea TGATCAAATCATTC-----CAG----AGTT----TAAT----AGACCCC---TTT--T

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| L. martiniae | TGATCAAATCATTCC--ATTCCAGCCAGAGTT-----TAAT----AGACCCC---TTT--T |
| L. mathewsii | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. maximiliani | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. mediana | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. moniliformis | TGATCAAATTATTT--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. muirii | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. multifolia | TGATCAAATCATTCC--ATTCC-----TT-----TAAT----AGACCCC---TTT--T |
| L. mutabilis | TGATCAAATCATTCTCATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. namaquensis | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAATAGATAGACCCC---TTT--T |
| L. namibiensis | TGATCAAATCATTCC--ATT-CAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. nervosa | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCCAG-TTT--T |
| L. obscura | TGATCAAATCATTCC--ATT-CAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. paucifolia | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. perryae | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. perryae/unifolia | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. polypodantha | TGATCAAAA---TC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. pusilla | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. pustulata | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAATAGATAGACCCC---TTT--T |
| L. rosea | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. punctata | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. salteri | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. schelpei | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. sp | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---CTT--T |
| L. splendida | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAATAGATAGACCCC---TTT--T |
| L. stayneri | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. thomasiae | TGATCAAATCATTCC-----CAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. trichophylla | TGATCAAATCATTCC-----CAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. undulata | TGATCAAATCAT-----TCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. unicolor | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAATAGATAGACCCC---TTT--T |
| L. unifolia | TGATCAAATTATTC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. unifolia/hirta | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. valeriae | TGATCAAATCATTCC--ATTCCAG----A--T-----AGAT----AGACCCC---TTT--T |
| L. variegata | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. verticillata | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. violacea | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. youngii | GGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| L. zebrina | TGATCAAAA---TC--ATTCCAG----AGTT-----TAAT----AGACCC---TTT--T |
| L. zeyheri | TGATCAAATCATTCC--ATTCCAG----AGTT-----TAAT----AGACCCC---TTT--T |
| M. depressa | TGATCAAATCATTCC--ATTCCAG----AGTTTAGTTTAAT----AGACCC---TTT--TT |
| V. bracheata | TGATCAAATCATTT--ATTCCAG----AGTT-----TAAT----AGACCC---TTTTTTT |
| V. capensis | TGATCAAATCATTT--ATTCCAG----AGTT-----TAAT----AGACCC---TTT--T |

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| <i>W. bifolia</i> | TGATCAAATCATTCC--ATTCCAG----AGTT----TAAT----AGACCC----TTT-TT |
| <i>E. bicolor</i> | TTGAAAAACGGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>E. vandermerwei</i> | TTGAAAAACGGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. alba</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. flava</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. ameliae</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. angelica</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. anguinea</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. attenuata</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. aurioliae</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. bolusii</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. bowkeri</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. bifolia</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. capensis</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. cernua</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. comptonii</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. concordiana</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. congesta</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. contaminata</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. convallarioides</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. corymbosa</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. dehoopensis</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. doleritica</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. duncanii</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. ensifolia subsp. ensifolia</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L._ensifolia var. maughanii</i> | TTGATAATCTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. fistulosa</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. framesii</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. gillettii</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. haarlemensis</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. hirta</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. inconspicua</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. isopetala</i> | TTGAAAAACTGATTAATCGAACGAGAATCGAGAATAAAGAGAGAGTCCCGTTCTACATGT |
| <i>L. magentea</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. juncifolia var. juncifolia</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. karooica</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. kombergensis</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. arbuthnotiae</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. longituba</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>L. margaretea</i> | TTGAAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |

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|---------------------|------------------------------------------------------------|
| L. martiniae | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. mathewsii | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. maximiliani | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. mediana | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. moniliformis | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. muirii | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. multifolia | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. mutabilis | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. namaquensis | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. namibiensis | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. nervosa | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. obscura | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. paucifolia | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. perryae | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. perryae/unifolia | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. polypodantha | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. pusilla | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. pustulata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. rosea | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. punctata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. salteri | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. schelpei | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. sp | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. splendida | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. stayneri | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. thomasiae | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. trichophylla | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. undulata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. unicolor | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. unifolia | TTGAAAACTGATTAATCGGACGAGAATA-----AAAGAGAGAGTCCCGTTCTACATGT |
| L. unifolia/hirta | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. valeriae | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. variegata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. verticillata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. violacea | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. youngii | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. zebrina | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| L. zeyheri | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| M. depressa | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| V. bracheata | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| V. capensis | TTGAAAACTGATTAATCGGACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |

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|--------------------------------------|--------------------------------------------------------------|
| <i>W. bifolia</i> | TTGAAAACTGATTAATCGTACGAGAATA-----AA-GAGAGAGTCCCGTTCTACATGT |
| <i>E. bicolor</i> | CAATATGTCAATACCGACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAGAAAAAAA |
| <i>E. vandermerwei</i> | CAATATGTCAATACCGACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAGAAAAAAA |
| <i>L. alba</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. flava</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. ameliae</i> | CAATAT-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. angelica</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. anguinea</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. attenuata</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| <i>L. aurioliae</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. bolusii</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. bowkeri</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. bifolia</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. capensis</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. cernua</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. comptonii</i> | CAATAT-----C-GACAACAA-TGAA-TTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. concordiana</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. congesta</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. contaminata</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. convallarioides</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. corymbosa</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. dehoopensis</i> | CAATAC-----C-GACAACAA-TGAA-TTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. doleritica</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. duncanii</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. ensifolia subsp. ensifolia</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. ensifolia var. maughanii</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. fistulosa</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. framesii</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. gillettii</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| <i>L. haarlemensis</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. hirta</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. inconspicua</i> | CAATAC-----C-GACACCAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. isopetala</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGAAAAGAAAAAAG |
| <i>L. magentea</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGGAGGAAAAGAAAAAA |
| <i>L. juncifolia var. juncifolia</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGGAGGAAAAGAAAAAA |
| <i>L. karoocia</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. kombergensis</i> | CAATAC-----C-GACAACAAATGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| <i>L. arbuthnotiae</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. longituba</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| <i>L. margaretea</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAAAGAAAAA |

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|---------------------|-------------------------------------------------------------|
| L. martiniae | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. mathewsii | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. maximiliani | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. mediana | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. moniliformis | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGGAGGAAAAGAAAAAA |
| L. muirii | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. multifolia | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. mutabilis | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. namaquensis | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| L. namibiensis | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAGAA |
| L. nervosa | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. obscura | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. paucifolia | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. perryae | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. perryae/unifolia | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. polypodantha | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAGAGAAAAGAAAAAA |
| L. pusilla | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. pustulata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| L. rosea | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. punctata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGGAAGAAAAAA |
| L. salteri | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. schelpei | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. sp | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. splendida | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| L. stayneri | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. thomasiae | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAAAAAAAAA |
| L. trichophylla | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. undulata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. unicolor | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| L. unifolia | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGGAGGAAAAGAAGAAA |
| L. unifolia/hirta | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. valeriae | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| L. variegata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. verticillata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. violacea | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. youngii | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| L. zebrina | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAGA |
| L. zeyheri | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| M. depressa | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |
| V. bracheata | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAGAAGAAAAAA |
| V. capensis | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAAAGAAAAAA |

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|--------------------------------------|-----------------------------------------------------------|
| <i>W. bifolia</i> | CAATAC-----C-GACAACAA-TGAAATTT-ATAGTAAGAGGGAAGGAAGAAAAAAA |
| <i>E. bicolor</i> | GAGGAAGAGAAAAGAGAGAAGGGAAGAAG |
| <i>E. vandermerwei</i> | GAGGAAGAGAAAAGAGAGAAGGGAAGAAG |
| <i>L. alba</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. flava</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. ameliae</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. angelica</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. anguinea</i> | GAGAAGGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. attenuata</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. aurioliae</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. bolusii</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. bowkeri</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. bifolia</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. capensis</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. cernua</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. comptonii</i> | GGGGAGGAAAAAGAGAGGAAGGGGGGAG |
| <i>L. concordiana</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. congesta</i> | GAGGAGGAAGGAAGAGAGGAAGGGGGAAG |
| <i>L. contaminata</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. convallarioides</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. corymbosa</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. dehoopensis</i> | GAGGAGGAAAAAGAGAGGAAGGGGGGAG |
| <i>L. doleritica</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. duncanii</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. ensifolia subsp. ensifolia</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. ensifolia var. maughanii</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. fistulosa</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. framesii</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. gillettii</i> | GAGGAGGAAAAAGAGAAGAGGGGGAAG |
| <i>L. haarlemensis</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. hirta</i> | GAGAAGGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. inconspicua</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. isopetala</i> | AAGGAAGAAAAAGAAGGAAAAGGGAAG |
| <i>L. magentea</i> | GAGAAGGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. juncifolia var. juncifolia</i> | GAGAAGGGAAAAAGAGAGAGAGGGGGAAG |
| <i>L. karooica</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. kombergensis</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. arbutnotiae</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |
| <i>L. longituba</i> | GAGGAGGAAAAAGAGAGAAAGGGGGAAG |
| <i>L. margaretea</i> | GAGGAGGAAAAAGAGAGGAAGGGGGAAG |

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|---------------------|------------------------------|
| L. martiniae | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. mathewsii | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. maximiliani | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. mediana | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. moniliformis | GAGAAGGGAAAAAGAGGAAGGGGAAG |
| L. muirii | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. multifolia | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. mutabilis | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. namaquensis | GAGGAGGAAAAAAGAGAAGGGGAAG |
| L. namibiensis | GAGAAGGAAAAAAGAGGAAGGGGAAG |
| L. nervosa | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. obscura | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. paucifolia | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. perryae | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. perryae/unifolia | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. polypodantha | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. pusilla | GAGGGAGAAAAAAGAGGAAGGGGAAG |
| L. pustulata | GAGGAGGAAAAAAGAGAAGGGGAAG |
| L. rosea | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. punctata | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. salteri | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. schelpei | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. sp. | GAAGAGGAAAAAAGAGGAAGGGGAAG |
| L. splendida | GAGGAGGAAAAAAGAGAAGGGGAAG |
| L. stayneri | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. thomasiae | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. trichophylla | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. undulata | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. unicolor | GAGGAGGAAAAAAGAGAAGGGGAAG |
| L. unifolia | GAGAAGGGAAAAAGAGGAAGAGGAAG |
| L. unifolia/hirta | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. valeriae | GAGGAGGAAAAAAGGGAGGAAGGGGAAG |
| L. variegata | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. verticillata | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. violacea | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. youngii | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. zebrina | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| L. zeyheri | GAGGAGGAAAAAAGAGGAAGGGGAAG |
| M. depressa | GAGGAAGAAAAAAGAAAGAGGGGAAG |
| V. bracheata | GAGGAAGAAAAAAGAGAAAAGGGGAAG |
| V. capensis | GAGGAAGAAAAAAGAGGAAGGGGAAG |

W. bifolia

GAGGAAGAAAAAGAGAGAGAGGGGAAG

Appendix G: Aligned sequences of the combined *rbcl* and indels dataset of all the specimens used for cladistics analysis. *E.* = *Eucomis*, *G.* = *Gloriosa*, *M.* = *Massonia*, *L.* = *Lachenalia*, *V.* = *Veltheimia*, *Whiteheadia bifolia*.

| | |
|--------------------------------------|--------------------------------------------------------------|
| <i>E. autumnalis</i> | GATTGACTT--ATTATAC-----TCGTATTATTACGAAACCAAAGATACTGATATT-- |
| <i>E. bicolor</i> | TATTTTGAC--GCCGTGG-----TCTTGTTT-TTACG--CG-GTGAATTGATTTT-- |
| <i>E. pole-evansii</i> | GATTGACTTGTATTATACTCGGTGGACCTGATT-ACGAAA-CCAAAGATACTGATATTAC |
| <i>E. vandermerwei</i> | GATTGACTTT-ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>G. superba</i> | AATTGACTT--ATTATAC-----TCCTGAAT-ACAAAACCCAAGATACTGATATC-- |
| <i>L. angelica</i> | ??????????ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. alba</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. flava</i> | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. ameliae</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. anguinea</i> | GATTAACCT--ATTATAC-----TCCTGATTTACGAAA-CCAAAGATACTGATATT-- |
| <i>L. attenuata</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. aurioliae</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. bolusii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. bowkeri</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. bifolia</i> | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. capensis</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. cernua</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. comptonii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. concordiana</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. congesta</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. contaminata</i> | ??TGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. convallarioides</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. cornymabosa</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. corymbosa</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. dehoopensis</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. doleritica</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. duncanii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. ensifolia subsp. ensifolia</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. ensifolia var. maughanii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. fistulosa</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. framesii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. gillettii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. haarlemensis</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. hirta</i> | ?ATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. inconspicua</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. isopetala</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. magentea</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |

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| L. juncifolia var. juncifolia | GATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. karooica | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. kombergensis | ??????????ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. arbuthnotiae | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. longituba | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. longibracteata | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. margaretea | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. martiniae | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. mathewsii | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. maximiliani | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. mediana | TATTTA-AC--GG-GTGG-----TATTGTTT--TTACA-GCGGAGGCCGTGATTTT-- |
| L. moniliformis | ?ATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. muirii | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. multifolia | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. mutabilis | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. namaquensis | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. namibiensis | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. nardousbergensis | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. nervosa | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CTAAAGATACTGATATT-- |
| L. obscura | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. orthopetala | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. paucifolia | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. perryae | GATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. perryae/unifolia | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. polypodantha | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. pusilla | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. pustulata | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. rosea | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. punctata | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. salteri | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. schelpei | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. sp | ???????????????????????????????????????????????????????????? |
| L. sp | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. splendida | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. stayneri | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. thomasiae | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. trichophylla | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. trichophylla subsp. trichophylla | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. undulata | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. unicolor | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| L. unifolia | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |

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| <i>L. unifolia/hirta</i> | GATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. valeriae</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. variegata</i> | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. verticillata</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. violacea</i> | ?ATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. youngii</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CTAAAGATACTGATATT-- |
| <i>L. zebrina</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>L. zeyheri</i> | GATTAACCT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>M. depressa</i> | GATTGACTT--ATTATAC-----TCCGGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>M. wittebergensis</i> | GATTGACTT--ATTATAC-----TCCGGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>V. bracheata</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>V. capensis</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| <i>W. bifolia</i> | GATTGACTT--ATTATAC-----TCCTGATT-ACGAAA-CCAAAGATACTGATATT-- |
| | |
| <i>E. autumnalis</i> | AATATTG-AGCAG-CATTCCGA-GTAACTCCTCAACC-TGGAGTTCCC-GCTGAAGAAGC |
| <i>E. bicolor</i> | GAGGGTGAATTCATTTTACAC-GGCGCCTCAAATTG-GAGTTCCCG-GTGAAGAATCAG |
| <i>E. pole-evansii</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCAA-GAGTTCCCG-CTGAAGAAGCAG |
| <i>E. vandermerwei</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>G. superba</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. alba</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. flava</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. ameliae</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. anguinea</i> | TTAGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCGCTGAAGAAGCAG |
| <i>L. attenuata</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. auriloliae</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. bolusii</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. bowkeri</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. bifolia</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. capensis</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. cernua</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. comptonii</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. concordiana</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. congesta</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. contaminata</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. convallarioides</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCCG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. cornymabosa</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. corymbosa</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. dehoopensis</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. doleritica</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. duncanii</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. ensifolia subsp. ensifolia</i> | TTGGCAG----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |

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| L. ensifolia var. maughanii | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. fistulosa | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. framesii | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. gillettii | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. haarlemensis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. hirta | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. inconspicua | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. isopetala | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. magentea | TGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. juncifolia var. juncifolia | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. karooica | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. kombergensis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. arbutnotiae | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. longituba | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. longibracteata | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. margaretea | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CGGAAGAAGCAG |
| L. martiniae | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. mathewsii | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. maximiliani | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. mediana | GCAATTTGGGGGATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. moniliformis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. muirii | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. multifolia | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. mutabilis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. namaquensis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. namibiensis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. nardousbergensis | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. nervosa | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. obscura | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. orthopetala | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. paucifolia | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. perryae | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. perryae/unifolia | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. polypodantha | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. pusilla | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. pustulata | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. rosea | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. punctata | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. salteri | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. schelpei | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| L. sp | ???????????????????????????????????????????????????????????? |

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| <i>L. sp</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. splendida</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. stayneri</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. thomasiae</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. trichophylla</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. trichophylla subsp. trichophylla</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GGGTTCCCG-CTGAAGAAGCAG |
| <i>L. undulata</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. unicolor</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. unifolia</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. unifolia/hirta</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. valeriae</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. variegata</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. verticillata</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. violacea</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. youngii</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. zebrina</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>L. zeyheri</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>M. depressa</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>M. wittebergensis</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>V. brachatea</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>V. capensis</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| <i>W. bifolia</i> | TTGGCAG-----CATTCCGAGT-AACTCCTCAACCTG-GAGTTCCCG-CTGAAGAAGCAG |
| | |
| <i>E. autumnalis</i> | AGGGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGG |
| <i>E. bicolor</i> | GGGCTGGGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>E. pole-evansii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>E. vandermerwei</i> | GGGCTGCGGTAGCTGCCGAATCTTCTAGTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>G. superba</i> | GGGCTGCAGTAGCTGCCGAGTCTTCTACTGGTACATGGACAACCTGTGTGGACTGATGGAC |
| <i>L. angelica</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. alba</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. flava</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. ameliae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. anguinea</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. attenuata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. aurioliae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. bolusii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. bowkeri</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. bifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. capensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. cernua</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. comptonii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |

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|---------------------------------------------|---------------------------------------------------------------|
| <i>L. concordiana</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. congesta</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. contaminata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. convallarioides</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. cornymabosa</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. corymbosa</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. dehoopensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. doleritica</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. duncanii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. ensifolia</i> var. <i>maughanii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. fistulosa</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. framesii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. gillettii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. haarlemensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. hirta</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. inconspicua</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. isopetala</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. magentea</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. karooica</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. kombergensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. arbutnotiae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. longituba</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. longibracteata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. margaretea</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. martiniae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. mathewsii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. maximiliani</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. mediana</i> | GAGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. moniliformis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. muirii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. multifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. mutabilis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. namaquensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. namibiensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. nardousbergensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. nervosa</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. obscura</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. orthopetala</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. paucifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |

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|--------------------------------------------|------------------------------------------------------------------------|
| <i>L. perryae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. perryae/unifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. polypodantha</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. pusilla</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. pustulata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. rosea</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. punctata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. salteri</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. schelpei</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. sp</i> | ????????????????????????????????????????????????????????????ACCGATGGAC |
| <i>L. sp</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. splendida</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. stayneri</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. thomasiae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. trichophylla</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. trichophylla subsp. trichophylla</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. undulata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. unicolor</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. unifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. unifolia/hirta</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. valeriae</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. variegata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. verticillata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. violacea</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. youngii</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. zebrina</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>L. zeyheri</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>M. depressa</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>M. wittebergensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>V. bracheata</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>V. capensis</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>W. bifolia</i> | GGGCTGCGGTAGCTGCCGAATCTTCTACTGGTACATGGACAACCTGTGTGGACCGATGGAC |
| <i>E. autumnalis</i> | ACTTACCAGTCTTGATCGTTACAAAGGACGATGCTACCACATTGAGGCCGTTGTTGGG-G |
| <i>E. bicolor</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>E. pole-evansii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>E. vandermerwei</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGTTAAACCACATTGAGGGGATTGTTTTGAG |
| <i>G. superba</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACAGCATCGAGAAAGTTATTGGG-G |
| <i>L. angelica</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. alba</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. flava</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |

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|---------------------------------------------|--------------------------------------------------------------|
| <i>L. ameliae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>L. anguinea</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. attenuata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. aurioliae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. bolusii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. bowkeri</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. bifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. capensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. cernua</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. comptonii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>L. concordiana</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. congesta</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. contaminata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. convallarioides</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. cornymabosa</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. corymbosa</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCATATTGAGGCCGTTGTTGGA-G |
| <i>L. dehoopensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. doleritica</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. duncanii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. ensifolia</i> var. <i>maughanii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. fistulosa</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. framesii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. gillettii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. haarlemensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. hirta</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. inconspicua</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. isopetala</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>L. magentea</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. karooica</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. kombergensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. arbuthnotiae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. longituba</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. longibracteata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. margaretea</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. martiniae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. mathewsii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. maximiliani</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. mediana</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. monilliformis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |

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|--------------------------------------------|--------------------------------------------------------------|
| <i>L. muirii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. multifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. mutabilis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. namaquensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. namibiensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. nardousbergensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. nervosa</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. obscura</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. orthopetala</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. paucifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. perryae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. perryae/unifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. polypodantha</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. pusilla</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. pustulata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. rosea</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCATATTGAGGCCGTTGTTGGA-G |
| <i>L. punctata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. salteri</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCATATTGAGGCCGTTGTTGGA-G |
| <i>L. schelpei</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. sp</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. sp</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. splendida</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. stayneri</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. thomasiae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. trichophylla</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. trichophylla subsp. trichophylla</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCTGTTGTTGGA-G |
| <i>L. undulata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. unicolor</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. unifolia</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. unifolia/hirta</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. valeriae</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. variegata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. verticillata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. violacea</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. youngii</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. zebrina</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>L. zeyheri</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>M. depressa</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>M. wittebergensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G |
| <i>V. bracheata</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |
| <i>V. capensis</i> | TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGG-G |

W. bifolia TTACCAGTCTTGATCGTTACAAAGGACGATGCT--ACCACATTGAGGCCGTTGTTGGA-G

E. autumnalis AAGAAAATCAATTT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
E. bicolor AAGAAAATCAATTT--ATTGCTTGGGAAGAATGTAGCTTATGACCCAGTTTAGATAACCTT
E. pole-evansii AAGAAAATCAATTT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
E. vandermerwei AAGAAAAGGAATTGGAATTTTACGCG---GTGGACTTGATTTTACGCGGTGGACTTGATT
G. superba AAGATAATCAATAT--ATTGCTTACG---GTGGACTCCTTTAGACGCGGTGGAAG-AATT
L. angelica AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. alba AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. flava AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. ameliae AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. anguinea AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. attenuata AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. aurioliae AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. bolusii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. bowkeri AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. bifolia AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. capensis AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. cernua AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. comptonii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. concordiana AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. congesta AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. contaminata AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. convallarioides AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. cornymabosa AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. corymbosa AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. dehoopensis AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. doleritica AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. duncanii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. ensifolia subsp. ensifolia AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. ensifolia var. maughanii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. fistulosa AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. framesii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. gillettii AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. haarlemensis AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. hirta AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. inconspicua AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. isopetala AAGAAAATCAATTT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. magentea AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. juncifolia var. juncifolia AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT
L. karooica AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT

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|-------------------------------------|-------------------------------------------------------------|
| L. kombergensis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. arbuthnotiae | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. longituba | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. longibracteata | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. margaretea | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. martiniae | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. mathewsii | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. maximiliani | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. mediana | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. moniliformis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. muirii | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. multifolia | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. mutabilis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. namaquensis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. namibiensis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. nardousbergensis | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. nervosa | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. obscura | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. orthopetala | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. paucifolia | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. perryae | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. perryae/unifolia | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. polypodantha | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. pusilla | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. pustulata | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. rosea | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. punctata | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. salteri | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. schelpei | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. sp | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. sp | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. splendida | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. stayneri | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. thomasiae | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. trichophylla | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. trichophylla subsp. trichophylla | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. undulata | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. unicolor | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. unifolia | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. unifolia/hirta | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| L. valeriae | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |

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|--------------------------------------|--------------------------------------------------------------|
| <i>L. variegata</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>L. verticillata</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>L. violacea</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>L. youngii</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>L. zebrina</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>L. zeyheri</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>M. depressa</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>M. wittebergensis</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>V. bracheata</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>V. capensis</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| <i>W. bifolia</i> | AAGAAAATCAATAT--ATTGCTT-----ATGTAGCTTAT---CC--TTTAGA---CCTT |
| | |
| <i>E. autumnalis</i> | T-----TTGAAGAGGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>E. bicolor</i> | TGAGCTGTTGAAGAGGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>E. pole-evansii</i> | T-----TTGAAGAGGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>E. vandermerwei</i> | T-----TACGCGGTGGACTTGAT-TTTACGCGGTGGACTTGATTTACACG-GTGGACT |
| <i>G. superba</i> | T-----TACGCGGTGGACATGAT-TTTACGCGGTGGACTTGATTTACGCG-GTGGACT |
| <i>L. angelica</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. alba</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. flava</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. ameliae</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. anguinea</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. aurioliae</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. bolusii</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. bowkeri</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. bifolia</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. capensis</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. cernua</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. comptonii</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. concordiana</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. congesta</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. contaminata</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. convallarioides</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. cornymabosa</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. corymbosa</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. dehoopensis</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. doleritica</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. duncanii</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. ensifolia subsp. ensifolia</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. ensifolia var. maughanii</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. fistulosa</i> | T-----TTGAAGAAGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |

L. framesii T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. gillettii T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. haarlemensis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. hirta T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. inconspicua T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. isopetala T-----TTGAAGAGGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. magentea T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. juncifolia var. *juncifolia* T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. karooica T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. kombergensis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. arbutnotiae T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. longituba T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. longibracteata T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. margaretea T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. martiniae T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. mathewsii T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. maximiliani T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. mediana T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. moniliformis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. muirii T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. multifolia T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. mutabilis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. namaquensis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. namibiensis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. nardousbergensis T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. nervosa T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGCA-ATGTATT
L. obscura T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. paucifolia T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. perryae T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. perryae/unifolia T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. polypodantha T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. pusilla T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. pustulata T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. rosea T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. punctata T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. salteri T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. schelpei T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. sp T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. sp T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. splendid T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT
L. stayneri T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT

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|---------------------------------------------------|-------------------------------------------------------------|
| <i>L. thomasiae</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. trichophylla</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. trichophylla</i> subsp. <i>trichophylla</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. undulata</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. unicolor</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. unifolia</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. unifolia/hirta</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. valeriae</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. variegata</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. verticillata</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. violacea</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. youngii</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGCA-ATGTATT |
| <i>L. zebrine</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>L. zeyheri</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>M. depressa</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>M. wittebergensis</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>V. bracheata</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>V. capensis</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTGGGTA-ATGTATT |
| <i>W. bifolia</i> | T-----TTGAAGAAGGTTCTGTTACTAACATGTTTACTTCCATTGTAGGTA-ATGTATT |
| | |
| <i>E. autumnalis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTGGGAGGACTCGAATTCC |
| <i>E. bicolor</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>E. pole-evansii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>E. vandermerwei</i> | TGATTTTA---CGCGTGGACTTGATTTTACGCGGTGGACTTG-----ATTTTAC |
| <i>G. superba</i> | TGATTTTA---CGCGTGGACTTGATTTTACGCGGTGGACTTG-----ATTTTAC |
| <i>L. angelica</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. alba</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. flava</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. ameliae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. anguinea</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. attenuata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. aurioliae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. bolusii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. bowkeri</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. bifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. capensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCCT |
| <i>L. cernua</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCCT |
| <i>L. comptonii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. concordiana</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. congesta</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. contaminata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |

| | |
|---------------------------------------------|-----------------------------------------------------------|
| <i>L. convallarioides</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. cornymabosa</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. corymbosa</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. dehoopensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. doleritica</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. duncanii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. ensifolia</i> var. <i>maughanii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. fistulosa</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. framesii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. gillettii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. haarlemensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. hirta</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. inconspicua</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. isopetala</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. magentea</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. karooica</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. kombergensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. arbutnotiae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. longituba</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. longibracteata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. margaretea</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. martiniae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. mathewsii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. maximiliani</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. mediana</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. moniliformis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. muirii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. multifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. mutabilis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. namaquensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. namibiensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. nardousbergensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. nervosa</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. obscura</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. orthopetala</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. paucifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. perryae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. perryae/unifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. polypodantha</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |

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| <i>L. pusilla</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. pustulata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. rosea</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. punctata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. salteri</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. schelpei</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. sp</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. sp</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. splendida</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. stayneri</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. thomasiae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. trichophylla</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. trichophylla subsp. trichophylla</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. undulata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. unicolor</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. unifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. unifolia/hirta</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. valeriae</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. variegata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. verticillata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. violacea</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. youngii</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. zebrina</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>L. zeyheri</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>M. depressa</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>M. wittebergensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>V. bracheata</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>V. capensis</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| <i>W. bifolia</i> | TGGTTTCAAAGCCCTACGAGCTCTACGTC---TGGAGGATCTG-----CGAATTCCC |
| | |
| <i>E. autumnalis</i> | CCGGTGGACTTGCTTATTCCCGGAAAACCTGATTCTACG--CGGTGGACTTGCCATCCAA |
| <i>E. bicolor</i> | CC-----TTCTTATTC-C--AAAACCTTGATCCAAGGCCCGCCGCA-TTGC-ATCCAA |
| <i>E. pole-evansii</i> | CC-----TTCTTATTCG-C--AAAACCT--TCCAAGGCCCGCCGCACTGGC-ATCCAA |
| <i>E. vandermerwei</i> | GCGGTGGACTTGATTTTACCCGGTGGACTTGATTTTA-----CGCGGTGG--ACTTGA |
| <i>G. superba</i> | GCGGTGGACTTGATTTTACCGCGGACTTGATTTTA-----CGCGGTGG--ACTTGA |
| <i>L. angelica</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |
| <i>L. alba</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |
| <i>L. flava</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |
| <i>L. ameliae</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |
| <i>L. anguinea</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |
| <i>L. attenuata</i> | CC-----TTCTTATTC-C--AAAACCT--TCCAAGGCCCGCCCA-TGGC-ATCCAA |

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| <i>L. aurioliae</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. bolusii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. bowkeri</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. bifolia</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. capensis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. cernua</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. comptonii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. concordiana</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. congesta</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. contaminata</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. convallarioides</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. cornymabosa</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. corymbosa</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. dehoopensis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. doleritica</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. duncanii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. ensifolia</i> subsp. <i>ensifolia</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. ensifolia</i> var. <i>maughanii</i> | CC-----TGCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. fistulosa</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. framesii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. gillettii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. haarlemensis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. hirta</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. inconspicua</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. isopetala</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. magentea</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. karooica</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. kombergensis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. arbutnotiae</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. longituba</i> | CC-----TGCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. longibracteata</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. margaretea</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. martiniae</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. mathewsii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. maximiliani</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. mediana</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCCT-TGGC-ATCCAA |
| <i>L. moniliformis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. muirii</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. multifolia</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |
| <i>L. mutabilis</i> | CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA |

L. namaquensis CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. namibiensis CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. nardousbergensis CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. nervosa CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. obscura CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. orthopetala CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. paucifolia CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. perryae CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. perryae/unifolia CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. polypodantha CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. pusilla CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. pustulata CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. rosea CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. punctata CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. salteri CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. schelpei CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. sp CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. sp CC-----TGCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. splendida CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. stayneri CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. thomasiae CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. trichophylla CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. trichophylla subsp. trichophylla CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. undulata CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. unicolor CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. unifolia CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. unifolia/hirta CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. valeriae CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. variegata CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. verticillata CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. violacea CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. youngii CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. zebrina CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
L. zeyheri CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
M. depressa CC-----TGCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
M. wittebergensis CC-----TGCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
V. brachatea CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
V. capensis CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA
W. bifolia CC-----TTCTTATTC-C--AAAACCTT--TCCAAGGCCCGCCCCA-TGGC-ATCCAA

E. autumnalis TTTTGAAAGAGATAAA-TTGTACT-----TTTACCCGGTGGACCCCTATT----G

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| <i>E. bicolour</i> | GTT-GAAAGAGATAAA-TTGAACAGTGAAGGTGATTTACGCGGTCGTCCCCTATTAC--G |
| <i>E. pole-evansii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATTTTACG |
| <i>E. vandermerwei</i> | TTT--TACGCGGTGGACTTGATTT-----TACGCGGTGGACTTGATTTTA-CG |
| <i>G. superba</i> | TTT--TACGCGGTGGACTTGATTT-----TACGCGGTGGACT??????????? |
| <i>L. angelica</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. alba</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. flava</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. ameliae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. anguinea</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. attenuata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. aurioliae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. bolusii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. bowkeri</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. bifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. capensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. cernua</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. comptonii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. concordiana</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. congesta</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. contaminata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. convallarioides</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. cornymabosa</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGCCGTCCCCTATT----G |
| <i>L. corymbosa</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. dehoopensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. doleritica</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. duncanii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. ensifolia subsp. ensifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. ensifolia var. maughanii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. fistulosa</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. framesii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. gillettii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. haarlemensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. hirta</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. inconspicua</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. isopetala</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTATT----G |
| <i>L. magentea</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. juncifolia var. juncifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. karooica</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. kombergensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. arbutnotiae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |
| <i>L. longituba</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCCTATT----G |

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| <i>L. longibracteata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. margaretea</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCATTATT----G |
| <i>L. martiniae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. mathewsii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. maximiliani</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. mediana</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. moniliformis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. muirii</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. multifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGCCGTCCCTTATT----G |
| <i>L. mutabilis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. namaquensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. namibiensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. nardousbergensis</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. nervosa</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. obscura</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. orthopetala</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGCCGTCCCTTATT----G |
| <i>L. paucifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. perryae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. perryae/unifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. polypodantha</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. pusilla</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. pustulata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. rosea</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. punctata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. salteri</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. schelpei</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. sp</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. sp.</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. splendida</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. stayneri</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. thomasiae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCATTATT----G |
| <i>L. trichophylla</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. trichophylla subsp. trichophylla</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. undulata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. unicolor</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. unifolia</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. unifolia/hirta</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. valeriae</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. variegata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. verticillata</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |
| <i>L. violacea</i> | GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G |

L. youngii GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
L. zebrine GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
L. zeyheri GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
M. depressa GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
M. wittebergensis GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
V. bracheata GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
V. capensis GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTTATT----G
W. bifolia GTT-GAAAGAGATAAA-TTGAACA-----AGTACGGTCGTCCCTGTT----G

E. autumnalis GGATGTACTAGGGAGGGAGAAGAAAAAAGAAAAAGAG
E. bicolor GGGTGGACTGGGGAAAAAAGGAGGGAAAGAGGAGAA
E. pole-evansii GGGTGGACTGGGGAGGGAGAAGGAGAGGAAAGGAGGA
E. vandermerwei CGGTGGACCGAAAAGAAAAGGGAAGAAAAGAAGAAGAGA
G. superba ?????????GGGAGAAAGGGGAAGAAAAGAAGAAGAGA
L. angelica GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. alba GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. flava GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. ameliae GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. anguinea GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. attenuata GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. aurioliae GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. bolusii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. bowkeri GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. bifolia GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. capensis GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. cernua GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. comptonii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. concordiana GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. congesta GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. contaminata GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. convallarioides GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. cornymabosa GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. corymbosa GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. dehoopensis GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. doleritica GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. duncanii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. ensifolia subsp. ensifolia GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. ensifolia var. maughanii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. fistulosa GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. framesii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA
L. gillettii GGATGTACTAGGGAGGGAGAAGGAGGGGAAGAGGAGGA

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|---------------------------------------------|-----------------------------------------|
| <i>L. haarlemensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. hirta</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. inconspicua</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. isopetala</i> | GGATGCACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. magentea</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. juncifolia</i> var. <i>juncifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. karooica</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. kombergensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. arbuthnotiae</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. longituba</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. longibracteata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. margaretea</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. martiniae</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. mathewsii</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. maximiliani</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. mediana</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. moniliformis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. muirii</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. multifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. mutabilis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. namaquensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. namibiensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. nardousbergensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. nervosa</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. obscura</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. orthopetala</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. paucifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. perryae</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. perryae/unifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. polypodantha</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. pusilla</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. pustulata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. rosea</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. punctata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. salteri</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. schelpei</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. sp</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. sp</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. splendida</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. stayneri</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. thomasiae</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |

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|---------------------------------------------------|-----------------------------------------|
| <i>L. trichophylla</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. trichophylla</i> subsp. <i>trichophylla</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. undulata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. unicolor</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. unifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. unifolia/hirta</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. valeriae</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. variegata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. verticillata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. violacea</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. youngii</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. zebrina</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>L. zeyheri</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>M. depressa</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>M. wittebergensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>V. bracheata</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>V. capensis</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |
| <i>W. bifolia</i> | GGATGTACTAGGGGAGGGAGAAGGAGGGGAAGAGGAGGA |