J Plant Biotechnol (2012) 39:219-224
DOI:http://dx.doi.org/10.5010/JPB.2012.39.4.219

Review

Alstroemeria plants and its biotechnological applications

Sung-Soo Lim · Sang-Il Lee · Se-Chan Kang · Jong-Bo Kim

Received: 16 October 2012 / Accepted: 23 October 2012

Abstract *Alstroemeria* plants are widely cultivated in many countries especially in Western Europe and North America and popularity has increased in recently due to its long-base life, large variety of colors and low energy requirement during cultivation period. So far, more than 60 species have been released on the commercial market in the world. To meet the demand of consumer and develop the elite *Alstroemeria* cultivars, conventional breeding including cross-hybridization and selection as well as mutation breeding were used. However, as other important ornamental plants such as lily, rose, carnation and orchids accepted the biotechnological methods, this newly-born approach should be applied and developed an optimized the genetic transformation system. Then, this biotechnological approach can be fused with the conventional breeding methods and thus can be contributed to the production of elite *Alstroemeria* plants containing agriculturally good genetic traits which are useful for the both farmers and consumers in the future. In this paper, we reviewed the botanical and genetical features of *Alstroemeria* plants and its biotechnological approaches in the last decades.

Keywords *Alstroemeria*, Calli, Cut-flower, Regeneration, Transformation

Introduction

Recently, the popularity of *Alstroemeria* plants increased in many countries including Canada, Japan, UK and USA. It is because *Alstroemeria* flowers have a lot of beautiful colors, long-vase life and low energy requirement (Park et al. 2010). An overview of production volume and auction turnover since the past decade in the Netherlands is shown in Table 1. During the 2000s, there has been a slight change in the top 10 of famous cut flowers at the auction in Aalsmeer, The Netherlands. The consumers purchased more tulip, gerbera, and rose, whereas they lost more and more their interests for carnation, lily and chrysanthemum. The other cut flowers have remained stable. Although reduction in cultivation area and turnover in *Alstroemeria* were observed in the last 5 years, it might be due to the global economic crisis. As shown in Table 2, ranking changes in turnover of cut-flowers in Aalsmeer auction in the Netherlands which is the biggest one in the world can be observed for the last 10 years. Though *Alstroemeria* is not included top 10 cut-flower at this moment in auction, this flower is still popular cut-flower in many countries including Canada, Japan, Germany, the Netherlands, UK and USA. In Korea, this flower imported in the middle of 1990's. Since then, the popularity and demand on *Alstroemeria* cultivation have been increased. On the contrary, *Alstroemeria* is on the list of top 10 cut flowers in Korea as compared to the case of the Netherlands and finally ranked in the 10th position of the annual turnover in Korea (Table 1).

Discovery, geographical feature, and growth habit of *Alstroemeria*

In 1714, Feuillee discovered *Alstroemeria* in Chile, and he registered it under the genus *Hemerocallus*. The name *Alstroemeria* was given by Linnaeus in 1762 (Aker and Healy 1990). Linnaeus combined the information of Feuillee and Alstroemer, and named the genus *Alstroemeria* and described three species (Buitendijk 1998). Herbert (1837) reported 29 species, while Kunth described 40 species in 1850 (Uphof 1952). Later, Baker reported a total of 44 species, which he divided into two groups, the Chilean

S.-S. Lim
Department of Intenational Trade, Future College, Konkuk University, Choong-Ju, 380-701, Korea

S.-I. Lee · J.-B. Kim (✉)
Department of Biotechnology, College of Biomedical & Health Sciences, Konkuk University, Choong-Ju, 380-701, Korea
e-mail: jbhee1011@kku.ac.kr

S.-C. Kang
Dept. of Life Science, College of Bionano Technology, Gachon University, Seongnam 461-701, South Korea

Table 1 Supply volume and auction turnover of *Alstroemeria* in the Netherlands (Centraal Bureau Voor de Statistiek)

	2003	2004	2005	2011
Auction supply (no. stems million)	268.7	257.9	262.7	186
Auction turnover (Million Euro)	40	38.4	39.1	30

Table 2 Top 10 cut flowers in turnover in Aalsmeer flower auction in 2001 and 2011 (Bloemenbureau Holland, 2011, http://www.floraholland.com)

ranking	2001		2011	
	Flower	Turnover*	Flower	Turnover*
1	Rose	653.0	Rose	761
2	Chrysanthemum	289.1	Chrysanthemum	267
3	Tulip	177.3	Tulip	223
4	Lily	155.9	Lily	146
5	Gerbera	103.8	Gerbera	124
6	Cymbidium	66.6	Cymbidium	59
7	Freesia	61.7	Freesia	47
8	Carnation	56.2	Lisianthus	41
9	Alstroemeria	44.6	Amaryllis	36
10	Gypsophilia	42.0	Anthrurium	32

*: Euro × 1,000,000

Table 3 Top 10 cut flowers in turnover in flower auction in 2009 and 2010 in Korea (2010 Ministry for Food, Agriculture, Forestry and Fisheries)

ranking	2009		2010	
	Flower	Turnover*	Flower	Turnover*
1	Rose	116,960	Rose	99,575
2	Chrysanthemum	85,913	Chrysanthemum	77,390
3	Lily	28,539	Lily	29,335
4	Carnation	17,575	Carnation	19,656
5	Gerbera	16,106	Gerbera	15,388
6	Freesia	7,146	Gypsophilia	6,023
7	Gypsophilia	6,419	Freesia	5,599
8	Antirrhinum	6,069	Antirrhinum	5,551
9	Lisianthus	2,985	Lisianthus	2,903
10	Gladiolus	2,399	Alstroemeria	2,256

*: Korean Won × 1,000,000

species and the Brazilian species based on geographical distribution, with 24 and 20 species, respectively (Buidentijk 1998). Generally, the Chilean and Brazilian species can be discriminated by evaluating the morphological differences such as leaf shape, color and shapes of flower, the fragrance, and their year-round production (De Jeu et al. 1995).

Alstroemeria species predominantly have their natural habit in South-America, mainly in Chile and Brazil, as mentioned above, but also in Argentina, Bolivia, Paraguay, Peru, and Venezuela, species are found (Ravenna 1988). The center of distribution appears to be in central Chile (Bayer 1987). Some species such as *A. pelegrina*, *A. ligtu* and *A. aurea* are widely distributed, whereas others, like *A. patagonica*, are found in more restricted areas (Aker and Healy 1990). In general, soil temperature seems to be a crucial factor for the growth of *Alstroemeria* species. In many *Alstroemeria* species flowering is highly dependent on a period of cool soil temperature (Healy and Wilkins 1982, 1986). Cool

temperatures are also important for seed germination of many species of *Alstroemeria* (Hannibal 1942), especially the ones that grow high up in the mountains or in coastal areas. *A. campaniflora* is adapted to tropical marshy areas, whereas, *A. parvula* is found in an alpine area. Surprisingly, *A. polyphilla* and *A. graminea* are found in the desert (Aker and Healy 1990).

Botanical features of the *Alstroemeria* species

Alstroemeria plants are multiplied by splitting of fleshy rhizomes. The roots vary from thick and tuberous to thin and fibrous and produce thickened cylindrical storage roots that mainly contain starch and are edible (Bridgen et al. 1989). The *Alstroemeria* species have vegetative and generative shoots, which are initiated on the subterranean rhizomes that branch sympodically. The rhizome apex is the axillary bud of the first scale of the previous shoot. Then, each successive aerial shoot growing from the rhizome is a shoot grown from an axillary bud of the preceding aerial shoot. The axillary bud, which is also subterranean located, is the second scale leaf of the aerial shoot, and has the potential to produce growth as another rhizome. The other leaves do not have the special meristem for growing rhizome.

In the flower structure, the perianth consists of two whorls of three petals. The petal of the outer whorl has a different size and shape compared to the petal of the inner whorl. In some genotypes, the nectaries produce abundant drops of nectar. Moreover, spots and streaks are associated with the signaling of pollen vectors (De Jeu et al. 1992). *Alstroemeria* is predominantly an insect-pollinated crop (Dahlgren and Clifford 1982). The ovary is pseduo-epigyn (Buxbaum 1951), with three carpels forming a tripartite ovary in which an axial placenta is present. In each cavity of the ovary, two rows of ovules are located next to each other along the central placenta. The total number of ovules varies from 24 to 36 depending on the genotype (De Jeu et al. 1992).

The *Alstroemeria* has a protandrous flowering, which means that the anthers dehisce before the stigma is receptive. Therefore, self-pollination within a flower is difficult. In total, six anthers are situated in two whorls. Two days after anthesis, the first anther dehisces; at that moment, the style is still short and undeveloped. Four days after anther dehiscence, the anthers become dried and the filaments curl towards the lowest petal, at a distance of the developing style. Two days after all six anthers have wilted, the stigma becomes receptive, producing droplets of exudate on the papillae. This is the exact moment for pollination of the stigma. The pollen grains are only able to germinate a wet stigma. The pollen tubes grow between the papillae and within 24 hours grow through the cavity into the ovary. In general, after compatible fertilization, it takes about two months before the round seeds are scattered with force out of the ripe fruits.

Taxonomy and chromosome studies of *Alstroemeria*

Alstroemeria is a member of the monocotyledonous family *Alstroemeriaceae*, order Liliales, superorder Liliflorae, division Monocotyledon (Dahlgren et al. 1985). *Alstroemeria* first belonged to the Liliaceae, and later it was included in the Amaryllidaceae (Herbert 1837). Hutchinson (1957) proposed to separate the genus Alstroemeria from the Amaryllidaceae into a new family of Alstroemeriaceae, comprising four genera *Alstroemeria*, *Bomarea*, *Schickendantzia* and *Leontochir*. The classification of the genus *Alstroemeria* as described in Dahlgren et al. (1985) is generally accepted, although some *Alstroemeria* species are still considered as members of the *Amaryllidaceae*.

The chromosome number and genome composition of Alstroemeriaceae is well documented by Buitendijk (1998). The species of *Alstroemeria*, *Bomarea* and *Leontochir* are mainly diploid with a basic chromosome number of n=8 for *Alstroemeria* species and n=9 for *Bomarea* (Whyte 1929). However, the commercial cultivars are not only diploid, but also triploid (2n=3X=24), tetraploid (2n=4X=32), and even aneuploid (Hang and Tsuchyia 1988; Tsuchyia et al. 1987). Interestingly, the most attractive cultivars are triploid and tetraploid with big-sized flowers and a variety of colors.

Somatic embryogenesis and transformation in *Alstroemeria*

Based on the above-mentioned knowledgements in botanical and genetic studies in *Alstroemeria* species, breeders need to develop good *Alstroemeria* cultivars to meet consumer's demand. For this, conventional breeding and mutation techniques were employed so far. Brief history and development of *Alstroemeria* breeding was reviewed by Park et al. (2010).

However, as other ornamental crops combined biotechnological methods with conventional ways to make crop improvement efficiently, this kind of effort is supposed to be needed for *Alstroemeria* breeding program as well. To achieve this goal, the availability of efficient regeneration systems is essential for the application of genetic modification. As compared to dicotyledonous ornamentals, monocotyledonous ornamentals seem to be rather recalcitrant. *Alstroemeria* is not an exception. In the past decades, multiplication of *Alstroemeria* via tissue culture was carried out mainly by

rhizome splitting (Lin 1998). Once rhizomes have been produced from shoot clusters (Fig. 1A), plants will form healthy roots and will be established in the greenhouse within 3-4 months (Fig. 1B). Breeding companies and farmers use rhizome splitting in commercial propagation. Some *Alstroemeria* species, especially the "Butterfly type" have shown significantly low propagation efficiency (Buitendijk 1992). Therefore, a "Butterfly type" was used in this study. Another important *in vitro* technique is the embryo rescue system. It was developed to solve crossing-barriers and produced hybrids in *Alstroemeria* (De Jeu 1992). A large number of new cultivars have been developed through embryo rescue. However, neither embryo rescue techniques nor rhizome division is suitable for genetic modification due to the low efficiency and the non-adventitious character of the regeneration system. Adventitious regeneration is a precondition for genetic engineering.

Regeneration procedures have to be developed and to be able to produce genetically modified plants. The regeneration system might also be used for plant propagation. Adventitious regeneration was first reported by Ziv et al. (1973). They obtained plants from apical inflorescences via direct plant regeneration. Since then, a large number of publications have appeared.

In the late 1980s, Bridgen et al. (1989) reported on somatic embryogenesis and Gonzales-Benito and Alderson (1990, 1992) presented plant regeneration via callus tissues, which were induced from mature zygotic embryos. However, the regeneration efficiency of the system described was too low to be used for genetic transformation. A few years later, Hutchinson et al. (1994) described callus induction and plant regeneration from mature zygotic embryos with a 40% rate of regeneration frequency. In addition, they reported another regeneration system from callus using liquid culture (Hutchinson et al. 1997). Furthermore, Van Schaik et al. (1996) obtained plants from callus that was induced on immature zygotic embryos with 41-54% regeneration rates, depending on the used cultivars. Also, *Alstroemeria* plants were regenerated from seedling-derived friable embryogenic calli (FEC) (Lin et al. 2000a) and ovary-derived FEC (Akutsu et al. 2002) via somatic embryogenesis. In both cases FEC was derived from generative tissue and does lead to loss of the original genotype, since *Alstroemeria* is a vegetative propagated crop. Sage et al. (2000) also supported the view that it is necessary to develop an efficient embryogenic culture system from vegetative tissues in order to propagate elite new Narcissus genotypes. Thus, the two systems described by Lin et al. (2000a) and Akutsu et al. (2002) would be difficult to immediately combine with a genetic transformation protocol system in *Alstroemeria* due to their low efficiency in regeneration system and explant source problem. After the first production of transgenic *Alstroemeria* plants by using particle bombardment (Lin et al. 2000b), another two transformation studies via *agrobacterium tumefaciens* were reported by Akutsu et al. (2004a, b). However, it needs to be optimized to obtain a high efficiency and stable transgene integration as well as its gene expression in the further generations.

More recently, Kim et al. (2005) isolated protoplasts from FEC and regenerated protoplast-derived plants for the first time. Although the protoplast culture system showed a low efficiency as compared to the efficiency in potato and rice, it would be helpful to extend the genetic variation and develop new elite breeding lines within the current germplasm in *Alstroemeria* species. In fact, although this protoplast culture system is very important in terms of the extention of new genetic variation in floriculture abd vegetable breeding program, not many research groups are working on it especialyy in Korea. After that, CEC (compact type embryogenic calli) and FEC (friable embryogenic calli) were isolated leaves with axil tissues as vegetative explant and

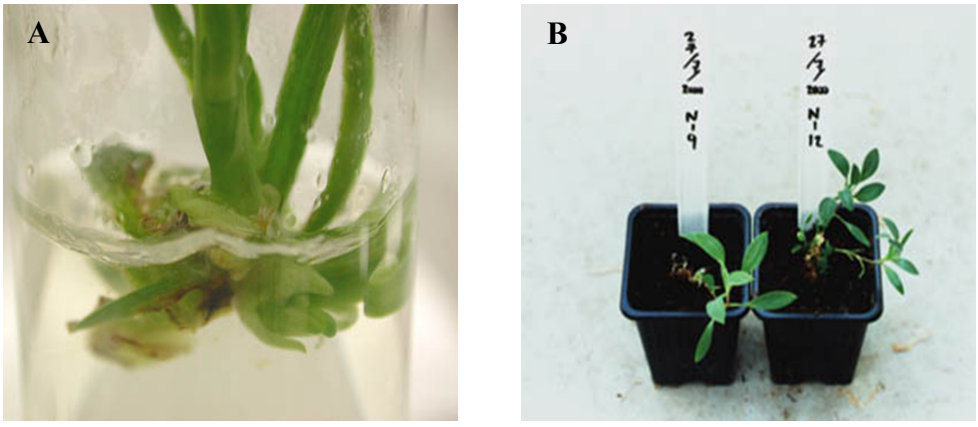

Fig. 1 Rhizome formation and regeneration in *Alstroemeria* A) Rhizome formation, B) 3 months after rhizome culture in the greenhouse

were used for regeneration of *Alstroemeria* plants with a high efficiency (Kim et al. 2006). These two different types of calli showed the different morphology in each other in the regeneration process in *Alstroemeria.* CEC type showed more a high regeration efficiency than that of FEC. But, FEC is supposed to be more useful for genetic modification system due to it's a high proliferation rate and less variation in further regeneration stage.

Based on this regeneration system via vegetative explants, *Agrobacterium*-mediated transgenic *Alstroemeria* plants were produced for the first time (Kim et al. 2007). In this report, Kim et al. (2007) optimized several factors influencing on *Agrobacterium*-mediated transformation efficiency and produced transgenic *Alstroemeria* plants with healthy roots and flowers in the greenhouse. Although there have not been mant research on genetic transformation in *Alstroemeria* species recenly, optimizations of regeneration and transformation system in *Alstroemeria* plants are conducting and plant biotechnology techniques such as genetic transformation can be fused with the conventional breeding system to provide cocan be applied for the development of virus-resistanc plants, mmercially elite line or cultivars. In the near future, these two main methods of *Alstroemeria* breeding program multiple abiotic-stresses tolerant plants, or plants which have a variety of fragrances and high contents of starch in roots. In this review, the general introduction and commercial importance of *Alstroemeria* plants were introduced. Also, its botanical features and breeding history including recent biotechnological application cases were described to inform that this plant species has a commercial importance in the plant industry of Korea. From this review report, further research with *Alstroemeria* plants will be performed and thus provide economic and resource values us in the future.

References

Aker S, Healy E (1990) The phytogeography of the genus *Alstroemeria.* Herbertia 46:76-87

Akutsu M, Sato H (2002) Induction of proembryos in liquid culture increases the efficiency of plant regeneration from *Alstroemeria* calli. Plant Sci 163:475-479

Akutsu M, Ishizaki T, Sato H (2004a) Transformation of the monocotyledonous *Alstroemeria* by *Agrobacterium tumefaciens*. Plant Cell Rep 22:561-568

Akutsu M, Ishizaki T, Sato H (2004b) Transformation of the monocot *Alstroemeria* by *Agrobacterium rhizogenes*. Mol Breed 13:69-78

Bayer E (1987) Die Gattung *Alstroemeria* in Chile. Mitteilungen der Botanischen saatssammlung. Munchen 24:1-362

Bridgen MP, Langhans R, Graig R (1989) Biotechnological breeding for *Alstroemeria*. Herbertia 45:93-95

Buitendijk JH, Ramanna MS, Jacobsen E (1992) Micropropagation ability: towards a selection criterion in *Alstroemeria* breeding. Acta Hort 325:493-498

Buitendijk JH (1998) A cytological characterization of genomes of *Alstroemeria*, the production of interspecific hybrids, and their performance during micropropagation. Ph.D thesis of Wageningen University. Wageningen, The Netherlands. pp. 131

Buxbaum F (1951) Die Grundachse von *Alstroemeria* und die Einheit ihres morphologischem Typus mit dem der echtem Liliaceen. Phytomorphology 1:170-184

Dahlgren RMT, Clifford HT (1982) Monocotyledons. A comparative study. Academic Press. London, New York. pp. 378

Dahlgren RMT, Clifford HT, Yeo PF (1985) The families of monocotyledons. Springer- Verslag Berlin

De Jeu MJ, Sasbrink H, Garriga CF, Piket J (1992) Sexual reproduction biology of *Alstroemeria*. Acta Hort 325:571-575

De Jeu MJ, Lasschuit J, Chevalier F, Visser RGF (1995) Hybrid detection in *Alstroemeria* by use of species repetitive probes. Acta Hort 420:62-64

Gonzalez-Benito E, Alderson PG (1990) Regeneration from *Alstroemeria* callus. Acta Hort 280:135-138

Gonzalez-Benito E, Alderson PG (1992) Callus induction and plant regeneration in *Alstroemeria*. J of Exp Bot 43:205-211

Hang A, Tsuchyia T (1988) Chromosome studies in the genus *Alstroemeria*. II. Chromosome constitutions of eleven additional cultivars. Plant Breed 100:273-279

Hannibal LS (1942) *Alstroemeria* and *Bomarea* from seeds. Herbertia 9:183-187

Healy WE, Wilkins HF (1982) Repose of *Alstroemeria* 'Regina' to temperature treatments prior to flower-inducing temperatures. Sci Hort 17:383-390

Healy WE, Wilkins HF (1986) Relationship between rhizome temperatures and shoot temperatures for flower initiation and cut flower production of *Alstroemeria* 'Regina'. J of Amer Soc Hort Sci 111:94-97

Herbert W (1837) Amaryllidaceae; preceded by an attempt to arrange the monocotyledonous orders and followed by a treatise on cross-bred vegetables, and supplement. London: Ridgway and Sons, pp 88-103

Hutchinson J (1957) The families of flowering plants II. Monocotyledons. Oxford. Clarendon Press

Hutchinson MJ, Tsujita JM, Saxena PK (1994) Callus induction and plant regeneration from mature zygotic embryos of a tetraploid *Alstroemeria* (*A. pelegrina* X *A. psittacina*). Plant Cell Rep 14:184-187

Hutchinson MJ, Senaratna T, Tsujita JM, Saxena PK (1997) Somatic embryogenesis in liquid cultures of a tetraploid *Alstroemeria*. Plant Cell Tiss and Org Cult 47:293-297

Kim JB, Bergervoet JEM, Raemakers CJJM, Jacobsen E, Visser RGF (2005) Isolation of protoplasts, and culture and regeneration into plants in *Alstroemeria*. In Vitro Cell Dev Bio-Plant 41(4):505-510

Kim JB, Raemakers CJJM, Jacobsen E, Visser RGF (2006)

Efficient somatic embryogenesis in *Alstroemeria*. Plant Cell Tiss and Org Cult 47:293-297

Kim JB, Raemakers CJJM, Jacobsen E, Visser RGF (2007) Efficient production of transgenic *Alstroemeria* plants by using *Agrobacterium tumefaciens*. Ann of Appl Bio 151: 401-412

Lin HS (1998) Development of two *in vitro* regeneration systems through leaf explant and callus culture and the application for genetic transformation in *Alstroemeria*. Ph. D thesis, Wageningen University, Wageningen, The Netherlands. pp 119

Lin HS, De Jeu MJ, Jacobsen E (2000a) Development of a plant regeneration system based on friable embryogenic callus in the ornamental *Alstroemeria*. Plant Cell Rep 19:529-534

Lin HS, Van der Toorn C, Raemakers CJJM, Visser RGF, De Jeu MJ, Jacobsen E (2000b) Genetic transformation of *Alstroemeria* using particle bombardment. Mol Breed 6:369-377

Park TH, Han IS, Kim JB (2010) Review on the development of virus resistant plants in *Alstroemeria*. J Plant Biotechnol 37:370-378

Ravenna P (1988) New or noteworthy species of *Alstroemeria*. Phytologia 64:281-288

Sage DO, James L, Neil H (2000) Somatic embryogenesis in *Narcissus pseudonarcissus* cvs. Golden Harvest and St. Keverne. Plant Sci 150:209-216

Tsuchyia T, Hang A (1987) Chromosome studies in genus *Alstroemeria*. Acta Hort 205:281-287

Uphof JCT (1952) A review of the genus *Alstroemeria*. Plant Life 8:37-53

Van Schaik CE, Posthuma A, De Jeu MJ, Jacobsen E (1996) Plant regeneration through somatic embryogenesis from callus induced on immature embryos of *Alstroemeria* spp. L. Plant Cell Rep 15:377-380

Whyte RO (1929) Chromosome studies. I. Relationship of the genera Alstroemeria and Bomarea. The New Phytologist 28:3119-344

Ziv M, Kanterovitz R, Halevy AH (1973) Vegetative propagation of *Alstroemeria in vitro*. Sci Hort 1:271-277