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REVISIÓN

In vitro propagation of turmeric (*Curcuma longa L.*) through direct somatic embryogenesis with reference to types of explants and plant growth regulators: A review

Propagación in vitro de cúrcuma (Curcuma longa L.) mediante embriogénesis somática directa con referencia a tipos de explantes y reguladores de crecimiento vegetal: una revisión

Propagação in vitro da cúrcuma (Curcuma longa L.) por meio da embriogênese somática direta com referência a tipos de explantes e regulardores do crescimento das plantas: uma revisão

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Abstract

Turmeric (*Curcuma longa L.*) is an important medicinal plant belonging to the Zingiberaceae family. Although it is a sterile triploid plant, it exhibits vigorous herbaceous growth mostly due to its underground rhizomes which are used for various purposes because of their physical and chemical properties. Turmeric is propagated vegetatively through the rhizomes; however, multiplication rate is low. On the other hand, *in vitro* propagation techniques are being successfully used for mass propagation of Zingiber species directly or indirectly through organogenesis or somatic embryogenesis. Direct plant regeneration is the desired method to regenerate genetically stable plants. In direct somatic embryogenesis, somatic cells directly differentiate into somatic embryos having bipolar structures. Since studies on direct somatic embryogenesis of turmeric are very limited, this review summarizes the current understanding of different aspects of somatic embryogenesis of turmeric and main regulating factors affecting it.

Keywords: Culture medium, turmeric, growth regulators, direct embryogenesis, explants

Resumen

La cúrcuma (*Curcuma longa L.*) es una importante planta medicinal perteneciente a la familia Zingiberaceae. Es una planta triploide estéril, pero es una planta herbácea vital principalmente debido a sus rizomas subterráneos que se utilizan para diversos fines debido a sus propiedades físicas y químicas. La cúrcuma se propaga vegetativamente a través de los rizomas, sin embargo, la tasa de multiplicación es baja. Por otro lado, las técnicas de propagación *in vitro* se están utilizando con éxito para la propagación masiva de especies de Zingiber directa o indirectamente a través de organogénesis o embriogénesis somática. La regeneración directa de plantas es el método deseado para regenerar plantas genéticamente estables. En la embriogénesis somática directa, las células somáticas se diferencian directamente en embriones somáticos que tienen estructuras bipolares. Los estudios sobre la embriogénesis somática directa de la cúrcuma son muy limitados, por lo tanto, esta revisión resume la comprensión actual de los diferentes aspectos de la embriogénesis somática de la cúrcuma y los principales factores reguladores que la afectan.

Palabras clave: medio de cultivo, cúrcuma, reguladores del crecimiento, embriogénesis directa, explantes.

Resumo

A cúrcuma (*Curcuma longa L.*) é uma importante planta medicinal pertencente à família Zingiberaceae. É uma planta triploide estéril, mas é uma planta herbácea vital principalmente devido aos seus rizomas subterrâneos que são utilizados para diversos fins devido às suas propriedades físicas e químicas. A cúrcuma se propaga vegetativamente através dos rizomas, porém a taxa de multiplicação é baixa. Por outro lado, as técnicas de propagação *in vitro* estão sendo usadas com sucesso para a propagação massiva de espécies de Zingiber direta ou indiretamente através da organogênese ou embriogênese somática. A regeneração direta de plantas é o método desejado para regenerar plantas geneticamente estáveis. Na embriogênese somática direta, as células somáticas se diferenciam diretamente em embriões somáticos que possuem estruturas bipolares. Os estudos sobre a embriogênese somática direta da cúrcuma são muito limitados, portanto, esta revisão resume a compreensão atual dos diferentes aspectos da embriogênese somática da cúrcuma e os principais fatores reguladores que a afetam.

Palavras-chave: meio de cultura, cúrcuma, reguladores de crescimento, embriogênese direta, explantes

Introduction

Turmeric (*Curcuma longa L.*), belonging to the family Zingiberaceae, is a perennial rhizomatous herb native to Asia and India. Turmeric is the most economically valuable member of the genus Curcuma, and there are about 133 species of turmeric worldwide (Prasad and Aggarwal, 2011). India is the biggest producer, exporter and consumer of turmeric (Shrishail et al., 2013). The rhizomes represent the economically important part of the cultivated turmeric plants by the presence of the colouring curcumin, essential oils and oleoresin, besides other nutritive constituents (Filho et al., 2000). Curcumin is responsible for the yellow coloration of rhizomes and biological activity of turmeric.

Turmeric is used in curry powder, chicken bouillon, sauces, gravies, dry seasonings, baking mixes, processed cheese, pickles, relishes, breading, soups, beverages and confections (Sasikumar, 2001). It is used as a colouring material in pharmacy, confectionery and other food industries. In India, turmeric is traditionally used for medicinal, socio-religious, culinary purposes and also as a cosmetic and dye (Shah, 1997). It is largely used as a household disinfectant in India, and also in religious ceremonies (Kandiannan et al., 2008). In South Asian countries, it is a part of religious rituals and it has been used since ancient times as a spice, food preservative, coloring agent, cosmetic, starch and ornamentals (Sirirugsa, 1999) as well as in traditional medicine (Ravindran, 2007).

Turmeric is used as an herbal medicine for arthritis, chronic anterior uveitis, conjunctivitis, skin cancer, smallpox, chicken pox, wound healing, urinary tract infections, and liver ailments (Chauhan et al., 2020). It is also used for cholesterol lowering, platelet aggregation, cardiovascular diseases, HIV replication and multiple sclerosis, dyspepsia and Alzheimer's disease (Aggarwal et al., 2007; Arli and Çelik, 2020). In Sri Lankan traditional Ayurvedic medicine, turmeric is also widely used (Subash, 2003) and turmeric powder has been used to control seed-borne diseases in rice (Gangopadhyay et al., 1998).

Cultivated in tropical and subtropical regions, turmeric requires a warm and humid climate at a temperature range of 20-35°C with an annual rainfall of 1500 mm or above, under both rainfed and irrigated conditions. Though it can be grown on different types of soils ranging from light black loam, red soils to clayey loams, rich loamy soils, it thrives best in well-drained sandy or clay loam soils with a pH range of 4.5-7.5 with good organic status (Shirgurkar et al., 2001). Turmeric can be cultivated organically as an intercrop along with other crops.

In Sri Lanka, turmeric is grown in wet and intermediate zones as a monocrop and as an intercrop under coconut, with major cultivated areas in Kurunegala, Gampaha, Kalutara, Kandy and Matale districts (Department of Export Agriculture, 2019). Although there are many local varieties of turmeric in Sri Lanka, not all of them are individually identified. Also, most of these local varieties are mixed with imported varieties such as Gunter, Puna and Madurasi Majal (Kandiannan et al., 2008). Sri Lanka has produced 11,103 metric tons of raw turmeric in Maha season to an extent of 922 ha (Department of Census and Statistics, 2019). Further, there is a good local and export market for turmeric grown in Sri Lanka as it has higher curcumin content, especially in organically cultivated turmeric (Department of Export Agriculture, 2012). It is one of the commercially cultivated minor export spice crops (Withanage et al., 2015), therefore good planting materials are required for large-scale field planting.

In vitro propagation of turmeric

Conventional plant propagation methods reduce the yield and the quality of the product and also cause important economic losses (Chitra, 2019); therefore, alternative techniques require rapid clonal propagation of turmeric plants. The aseptic culture of cells, tissues and organs under *in vitro* conditions is an important technique for commercial scale propagation of medicinal plants which are difficult to propagate through conventional methods. In turmeric, most tissue culture studies include plant regeneration through shoot multiplication (Balachandran et al., 1990; Panda et al., 2007; Kuanar et al., 2009; Singh et al., 2010), callus culture (Salvi et al., 2001; 2002; Roopadarshini, 2010), and direct regeneration of shoots (Salvi et al., 2000). Also, Ghosh et al. (2013) and Gomathy et al. (2014) cultured rhizomatous bud explants on MS medium containing different concentrations of cytokinins and auxins.

Pandey (2015) studied callus induction and *in vitro* multiplication of turmeric using rhizome buds and shoot tips of *Curcuma longa* cultured in MS medium supplemented with various combinations of growth regulators. A number of protocols for *in vitro* propagation of *C. longa* have been published by several researchers (Babu et al., 1997; Meenakshi et al., 2001; Shirgurkar et al., 2001; Sunitibala et al., 2001; Prathanturarug et al., 2003; Tyagi et al., 2007; Rahman et al., 2004; Panda et al., 2007; Kuanar et al., 2009; Singh et al., 2010). There are three different *in vitro* plant regeneration methods: axillary shoot proliferation, organogenesis, and somatic embryogenesis (Altman and Loberant, 1998). Meanwhile, Trejgell et al. (1998) stated that there are two ways of plant regeneration, without (direct) or with the involvement of the callus (indirect).

Table 1: Studies on *in vitro* micropropagation of turmeric (*Curcuma longa*) plants.

Explants	Best culture media for *in vitro* explants response	*In vitro* morphogenesis	References cited
Rhizome buds	MS + 1 mg/l Kin + 1 mg/l BAP	Multiple shoots	Keshavachandaran and Khader (1989)
Rhizome buds	MS + 3 mg/l BA	Multiple shoots	Balachandran et al. (1990)
Sprouted shoots	MS + 5 mg/l BA	Shootlets	Nayak (2000)
Emerging buds	MS + 1 mg/l BAP + 0,25 mg/l Kin	Multiple shoots	Ali et al. (2004)
Shoot tips	MS + 4 mg/l BAP + 1,5 mg/l NAA	Multiple shoots	Bharalee et al. (2005)
Rhizome buds	MS + 2 mg/l BAP + 2 mg/l Kin MS + 0,5 mg/l IAA	Shootlets Rooted shoots	Hazare et al. (2005)
Rhizome buds (1cm long)	WPM + 4 mg/l BAP + 1 mg/l NAA	Multiple shoots	Nasirujjaman et al. (2005)
Sprouted rhizome buds (1cm long)	MS + 3 mg/l BAP MS + 4 mg/l BAP MS+ 5 mg/l BAP	Multiple shoots Plantlets	Naz et al. (2009)
Sprouted rhizome buds (1.5-2.0 cm long)	MS + 2 mg/l BAP +0,5 mg/l NAA 1/2 MS + 2 mg/l NAA	Shootlets Rooted shoots	Behera et al. (2010)
Vertically excised two halves of aerial stem (0.5 cm long)	MS + 2 mg/l BAP	Direct somatic embryogenesis	Upendri and Seran, (2020a)

MS: Murashige and Skoog (1962) basal medium, WPM: Woody plant basal medium

Somatic embryogenesis

It is a process whereby somatic embryos are formed from somatic cells. *In vitro* somatic embryogenesis is classified into indirect and direct, based on the presence or absence of a phase of callus development respectively (Rout et al., 2006). In direct somatic embryogenesis, the somatic embryo develops directly from pre-embryogenic determined cells without the production of an intervening callus. It is a generally rare process as compared with indirect somatic embryogenesis. In indirect somatic embryogenesis, somatic cells are dedifferentiated into an unorganized mass of dividing cells, termed callus, which develops somatic embryos (Mathew and Philip, 2003). Somatic embryos are bipolar and similar to typical zygotic embryos. Somatic embryogenesis differs from other forms of plant regeneration such as organogenesis in that within a single step it produces a vascular system, functional meristem and a root/shoot axis (Bassuner et al., 2007).

Somatic embryos are used in embryological studies and mass plant propagation. They can be preserved for a longer period. Turmeric somatic embryos are potentially easier to handle than

rhizomes, as they are relatively small and uniform in size. Moreover, somatic embryos are used for the production of artificial or synthetic seeds (Chawla, 2002). It is one of the most desired pathways in plant regeneration through *in vitro* culture technique because it avoids the time-consuming and costly manipulation of individual explants, which is a problem with organogenesis (Folta and Dhingra, 2006; Carneros et al., 2009). It is used to obtain recombinant plants and useful in regeneration of genetically stable plants (Yang et al., 2009). The use of somatic embryos allows flexibility in scheduling production and transport without loss of viability and also maintains the clonal properties of the regenerated plantlets (Ghosh and Sen, 1994; Cervelli and Senaratna, 1995). Therefore, somatic embryos have potential applications for large-scale production of turmeric plants.

Zuraida et al. (2014) showed that a culture medium supplemented with 5 mg/l BAP and 2 mg/l 2,4-D enhanced callus induction after 70 days of culture, and that somatic embryogenesis was most successful with MS medium supplemented with 6.0 g/l agar, 5 mg/l BAP and 0.2 mg/l 2,4-D, obtaining 53% of calli differentiation into green somatic embryos and consequently a large number of plantlets. However, since callus formation causes somatic variation and the regenerated plants are not genetically uniform, *in vitro* screening is necessary. He and Gang (2014) noted indirect somatic embryogenesis in young inflorescences of *C. longa*. Therefore, this method was used to obtain embryogenic suspensions which were used in the genetic transformation procedure. The use of Gamborg medium containing 5 mg/l and 30 mg/l of NAA and BAP resulted in the formation of friable calli which afterward gave way to embryogenic cell suspensions. Procedures for somatic embryogenesis of other Curcuma species, such as *C. amada* and *C. caesia* (Raju et al., 2013) have also been reported. In previous studies, only indirect somatic embryogenesis has been verified in *C. longa* by using nodal segments (Jala, 2013) and young inflorescences (He and Gang, 2014).

Direct somatic embryogenesis

Plant regeneration through somatic embryogenesis from single cells has been established in many medicinal plant species. In this process, somatic embryos directly formed from cultured explants subsequently develop as complete plants (Tripathi and Tripathi, 2003). Direct somatic embryogenesis is a more desirable method to obtain regenerated plants which are similar to the parent plants (Mizukami et al. 2008). Direct somatic embryogenesis is usually rare when compared to indirect somatic embryogenesis. Unfortunately, in most species it is more difficult to obtain plants through direct embryogenesis (Attree and Fowke, 1993).

The first study on direct somatic embryogenesis and plant regeneration protocol for turmeric was done using leaf base explants as initial explants for initiation of direct somatic embryos and 90% of the explants formed primary embryos, 80% of which produced entire plants (Raju et al., 2015). Globular and elongated embryo stages are the key stages in the identification of somatic embryos (Godbole et al., 2002). Also, Bandyopadhyay and Hamill (2000) observed that surface cells of globular embryos were smooth as reported in other plants, and regularly arranged. The presence of protoderm and procambial strands in the developing embryos are additional key features used as indicators of somatic embryo formation (Figueroa et al., 2002; Sharma and Millam, 2004; Jalil et al., 2008). In Zingiberaceae, embryogenesis success is highly dependent on the interaction among genotype, types of explant, explant source and the culture medium containing plant growth regulators. Hence, each Zingiberaceae cultivar requires suitable explants and culture medium, particularly growth regulators for plant propagation.

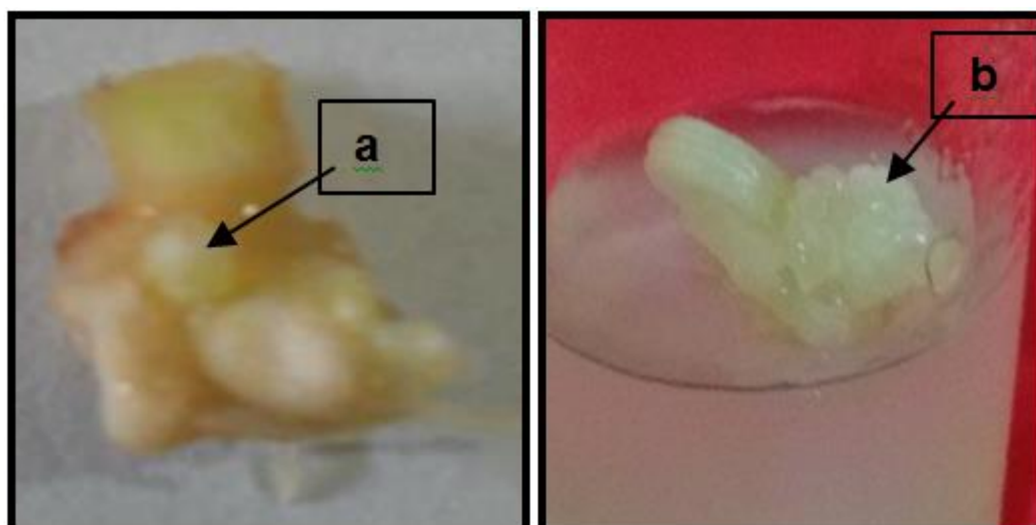

Figure 1: Direct somatic embryogenesis of *Curcuma longa* (a: Somatic embryos induced from exterior part of aerial stem base: b: Somatic embryos induced from interior part of aerial stem base)

Explant selection for somatic embryogenesis

Explant selection is one of the important factors in the induction of somatic embryogenesis. Genotype, explant source, and age of explants all affect somatic embryogenesis (Choudhury et al., 2008). Various explants like rhizome buds, rhizome pieces, axillary buds, shoot tips, leaves, meristem, inflorescences, and aerial pseudostems are used in the culture of Zingibers. The most common explant used for regeneration of Zingibers is the rhizome buds or the aerial shoots. Rhizome buds of turmeric have been utilized as explant for plant regeneration by Shirgurkar et al. (2001), Sunitibala et al. (2001), Salvi et al. (2002), Raihana et al. (2011), Chougule et al. (2011), and Gomathy et al. (2014). Upendri and Seran (2020a) stated that *in vitro* shoot buds and somatic embryoids were formed from cultured rhizome buds and aerial stem explants of turmeric respectively. Further, ginger, a closely related crop in the Zingiberaceae family, has also been successfully micropropagated by inducing growth of axillary and adventitious shoots using rhizome buds (Nadgauda et al., 1980; Sathyagowri and Seran, 2013), shoot tips (Ibrahim et al. 2015; El-Nabarawy et al., 2015; Ali et al., 2016), meristems (Rostiana and Syahid, 2008), aerial stems (Lincy et al., 2004; Lincy and Sasikumar, 2010) and decapitated crown sections (Pandey et al., 1997), and stem discs and axillary buds (Nogouchi and Yamakawa, 1988a, 1988b) *in vitro*. Many researchers have worked on somatic embryogenesis in ginger using different explants. Kackar et al. (1993) also induced somatic embryogenesis *in vitro* in ginger leaf explants. Induction of somatic embryos from ovary explants of ginger and its regeneration were described by Babu et al. (1996). Guo and Zhang (2005) maintained somatic embryogenic cultures of four ginger cultivars from shoot tip explants. A protocol for ginger regeneration via somatic embryogenesis from vegetative bud-derived callus cultures was reported by Suma and Keshavachandran (2005). Sathyagowri and Seran (2011) used 1-cm-long explants vertically excised into two halves to induce somatic embryogenic response of aerial ginger stems. Usually, juvenile plant parts are responsive to somatic embryogenesis or regeneration (Schulze, 2007). Previous reports are in agreement with somatic embryo proliferation in saffron at 25±2 °C and 16/8 h light and dark cycle (Vatankhah et al., 2010).

Figure 2: Turmeric plant (a: aerial stem; b: underground rhizome; c: pseudostem; d: rhizomes)

The size of explants affects tissue response under *in vitro* conditions. Smaller explants are more difficult to handle, an observation further supported by Smith (2000), who stated that larger explants probably contain more nutrient reserves and endogenous plant growth regulators to sustain the culture. Since plants have different growth regulators throughout their structure, the location of the explant source will determine different endogenous levels of plant growth regulators. Internal differences in endogenous growth regulators in the tissue can result in different *in vitro* responses (Smith, 2000). Zuraida (2013) indicated that survival rate was remarkably high (45%) when initial explant length was 3.0-4.0 cm, with explants 2.0 cm long exhibiting the second highest survival rate (31-35%). Further, he reported that the lowest survival pattern was observed in explants 0.5 cm long, as more explants turned brown and died. Thus, browning of the explants is associated with a lower survival percentage.

During culture establishment, explant browning is often a serious problem especially in the case of woody plants, and it could reduce the survival rate of cultured explants. Contamination of explants is related to various factors such as species, age, and health status of plants; type, source, and size of explants; aseptic handling and prevailing weather conditions. Contamination and low multiplication rates are the limiting factors in regenerating healthy and uniform clones of *in vitro* plantlets (Boullani et al., 2013). Webster et al. (2003) reported that the use of field-grown plants as a direct source of explants for the establishment of aseptic *in vitro* cultures is generally considered a major obstacle, especially in explants close to ground level. Commercial bleach (5.25% sodium hypochlorite as active ingredient), ethanol and mercury chloride ($HgCl_2$) have been reported as very effective sterilizing agents for establishing aseptic buds of many Zingiberaceae species (Neeta et al., 2002; Chan and Thong, 2004; Yusuf et al., 2007). Sterilization of living materials should eliminate bacterial or fungal contaminants.

Plant growth regulators for somatic embryogenesis

Somatic embryogenesis is controlled by the type and concentration of plant growth regulators and other substances. The agents used to induce *in vitro* somatic embryogenesis are highly variable, ranging from various plant growth regulators to stress treatments (Feher et al., 2003). According to the literature survey, most species require plant growth regulators for induction of somatic embryogenesis, and auxins as well as cytokinins are significant factors for determining the embryogenic response.

MS medium has been widely used as a basal culture medium, and BA or BAP supplementation has been found to have a significant effect on shoot proliferation in *in vitro* clonal propagation of Curcuma (Chan and Thong, 2004; Ali et al., 2004; Tule et al., 2005). Lincy et al. (2009) reported indirect and direct somatic embryogenesis from the plants in the aerial stem explants of ginger. Somatic embryos were induced in a medium containing BAP and the mature, club-shaped somatic embryos germinated in the presence of BAP and NAA at different concentrations (Lincy et al., 2009). MS medium containing 1.32 µM BAP accelerated somatic embryo formation after preincubation of leaf base explants with 4.49 µM 2,4-D for 20 days under continuous dark condition (Raju et al., 2015). In general, high cytokinin and low auxin levels were used to produce somatic embryos, and 2 mg/l BAP either without (Kamshananthi and Seran, 2012) or with optimum concentration of NAA (0.2-1.0 mg/l) (Seran et al., 2006; Seran, et al., 2007; Shanika and Seran, 2021) was suitable for somatic embryogenesis from cotyledon explants of different plant species. Somatic embryogenesis was reported in various explants of other species in response to BAP (Pacheco et al., 2006; Collado et al., 2010; San-José et al., 2010). BAP requirements for somatic embryogenesis may vary according to explant source (Bhaskaran and Smith, 1990).

The induction of direct somatic embryogenesis and organogenesis in the presence of BAP alone or in combination with auxin has been observed in intact bean seedlings (Malik and Saxena, 1992) and in leaf explants from *Helianthus smithii* (Laparra et al., 1997). Also, Sathyagowri and Seran (2011) reported that the degree of embryonic response rose as BAP concentration was increased from 1.0 mg/l to 2.0 mg/l, while BAP with NAA or IAA resulted in the explant producing shoot buds and roots via organogenesis in ginger. Stanly and Keng (2007) reported that a higher concentration of plant growth regulators was not suitable for *in vitro* culture of Zingiberaceae species. Guo and Zhang (2005) showed that BAP was also effective for regeneration of somatic embryos as well as for induction of somatic embryos. Kackar et al. (1993) also reported that the inclusion of BAP (0.5–1.5 mg/l) in the regeneration medium accelerated the germination of somatic embryos in ginger. The addition of GA3 in the culture media promoted germination of somatic embryos of Curcuma species (Prakash et al., 2001). Mature somatic embryos germinated readily, and maximum plantlet development was achieved in a 1/2-strength MS medium containing GA3 under dark conditions (Raju et al., 2015). Also, plantlets were shown to develop healthy root systems upon transfer to MS media containing only BAP (Nayak, 2000). Sand and compost at a 1:1 (v/v) ratio could be used as a potting medium rather than sand alone for ex vitro plant propagation of turmeric (Upendri and Seran, 2020b).

Figure 3: *In vitro* plantlets

Need for *in vitro* plant regeneration

Turmeric is propagated vegetatively through underground rhizomes; however, a large number of plant materials are unavailable for commercial purposes. Further, turmeric flowering is rare, and there are some diploids and tetraploids which either do not or rarely produce fertile seeds due to incompatibility and high pollen sterility (Joseph et al., 1999). Since turmeric rhizomes have a dormancy period and they sprout only during monsoon, only five or six plants can be obtained from one rhizome in a year (Sunitibala et al., 2001). Balachandran et al. (1990) stated that the rhizome multiplication rate is very low with yields ranging from 15 to 20 tons per hectare. There is a very high demand for planting materials as a large quantity of rhizomes is required for commercial cultivation due to the slow rate of propagation by the conventional method. Shirgurkar et al. (2001) stated that at harvest time, farmers can store 10-20% of the turmeric yield for subsequent cultivation. On the other hand, storage of a large quantity of turmeric rhizomes for planting is expensive and labour intensive, and proper storage facilities are required to preserve the planting materials.

The presence of certain soil-borne diseases in the rhizomes of turmeric negatively affects the quality and quantity of rhizome yield. It is one of the main factors determining the productivity of turmeric. The diseases are transmitted from one generation to the next; hence, use of rhizomes for planting poses high risks, especially for commercial growers. Among the diseases, rhizome rot, caused by Pythium spp. and leaf spot, caused by spp. of Taphrina and Collectrichum are the most serious (Dohroo, 2007). Kuanar et al. (2009) reported that *Pseudomonas solanacearum* (bacterial wilt disease), *Fusarium oxysporum* spp. Zingiberi (fusarium yellows disease) and Pythium spp. (soft/root rot) transmitted through rhizomes may result in about 60% of crop losses to growers. Moreover, the rhizome is infected by nematodes (*Exigua ornithogalli* and *Trichobaris trinodata*), and the larvae of insects which develop inside the rhizome subsequently affect proper plant growth (Zapata et al., 2003; Faisal and Alatar, 2019) causing severe shortage of healthy planting materials. Moreover, Shirgurkar et al. (2001) also mentioned that the improvement of rhizome crops through conventional breeding is difficult. Healthy plantlets could be regenerated under *in vitro* conditions for commercial plantings through somatic embryogenesis.

Conclusion

Turmeric is an important plant used for various purposes. Conventional turmeric propagation presents many problems; therefore *in vitro* regeneration is very significant in Zingiberaceae plants. According to this review, *in vitro* regenerated plants are superior to conventionally propagated plants regarding propagation, productivity and disease resistance. They can be easily hardened and transferred to the field with a high rate of survival. For *in vitro* culture of these plants, a suitable protocol should be identified from the earlier reports. Direct somatic embryogenesis is the best way to propagate turmeric plants that are genetically uniform because of their unicellular origin. Moreover, induction of somatic embryos is also sensitive to genotype, explant source, composition of culture medium, and culture environment conditions.

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