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Review article

A Mini Review on Tissue Culture Responses of Some auspicious Wheat (*Triticum aestivum* L.) cultivars in different Media, Salts and Growth hormones

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Abstract: Wheat (*Triticum aestivium)* is grown all over the world as a cereal crop. Diverse biotic and abiotic stresses seriously affect the wheat growth, which results in low yield. Plant tissue culture is a technique used for in vitro regeneration of plants. Standardization for callus induction and regeneration is of vital importance to deploy the transformation aim full in wheat but Poor tissue culture performance, which is another obstacle in transformation of wheat genotypes. Therefore, to address this obstacle this review depicts various works of various scientist. They conducted many researches to evaluate response of different wheat genotypes to callus induction and plant regeneration. Therefore, the information generated through this review could be utilized to overcome poor tissue culture performance in wheat.
Keywords: Wheat - Tissue culture - Regeneration - Transformation - Callus.

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INTRODUCTION

Tissue culture is an important tool for last few decades for various crop improvements. Many scientists used novel genes by many techniques to create genetic variability for crop genetic improvement. La Rue (1949) narrated about the very first culture from the origin maize endosperm, cereal callus culture. This is the reason behind *in vitro* culturing of wheat, rye and rice. Mendoza & Kaeppler (2002) described the tissue culture as very fine technique for cereals to genetically engineered. The genetic engineering of these cereals is fully dependent on the techniques of tissue culture. Cilar *et al.* (2006) utilized mature form of the embryo among 5 different species of the *Triticum*, that give rise to two mutually different media of MS like Murashige and Skoog (MS) media that is Naphthaleneacetic acid (NAA) free or 2, 4-D. In the results of callus induction, its percentage difference occurred depicted over two different media. The Yakar of *Triticum aestivium* depicted highest amount of regenerative capacity in both media. Kiziltan cultivar (*Triticum durumthe)* depicted the maximum regeneration capability in 4-D media and MS +2 but comparing the results with cultivar Yilmaz that showed the maximum capacity for regeneration in the media MS+NAA. It suggests that on the induction of callus strong effects of genotypes seen clearly. From these results, it concluded that various wheat species showed different behavior in diverse media.

Redway *et al.* (1990) is his investigation used coconut milk in the media and eight hexaploid fined lines of wheat's immature embryo. The outcome depicted a huge difference in the progression of shoot and primordial in the used wheat lines. Moreover, it was narrated that the induction of the callus is dependent on the genotype. Bouiamrine *et al.* (2012) observed that response of seven different durum wheat genotypes were recorded with the help of various immature and mature embryos for checking regeneration and the callus induction. Here five different characteristics were measured *i.e.* zygotic germination, percentage of induced calli, callogenesis,

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number of plantlets per regenerated callus and callus growth. Genotype influenced the induction of callus and the source of the explants. Mature embryo give comparatively low yield of the callus induction percentage as compared with those embryos that were immature. Therefore, immature embryos produced by in vitro could be select but albino immature plant showed excellent regeneration capacity so selection of albino plant would be fruitful. Sarker *et al.* (2007) reported the use of immature embryos for the regeneration of the plants and embryogenesis at the somatic stages. He moreover described that the presence of parameters like L –asparagine, casein hydrolysate and 2, 4-D, L- Proline, L –asparagine can cause momentous effects on the callus formation and embryogenesis at the somatic stages. Dagustu (2008) used four different segregating lines of wheat and five Iranian cultivars of wheat. Different media including cold, induction medium were used for the placement of the anthers of these cultivars and were subjected to the gamma radiations that resulted in giving differential response i.e. higher in one genotype as compared with others cultivars. Anther culturing is usually inhibited by the use of gamma radiations that caused a difference due to different concentrations of the radiations in the different genotypes. Here the result also showed that the anther culture is dependent on genotype.

Machii *et al.* (1998) examined varying concentrations of the NaCl in medium culture to observe the result of 28 different durum wheat cultivars on salt tolerance via *in vitro* technique. Immature embryo, subjected to experimentation; fresh weight growth of the cells of callus and frequency of induction of the callus was measured. For checking the salt tolerance, different traits like relative necrosis percentage of callus and fresh weight growth was examined. All the cultivars were significant different. The forms of callus derived from "Dipper-6" and "PI40100" showed more salt tolerance compared with *in vitro* conditions of other cultivars. Mahmood *et al.* (2012) used *Triticum durum* cultivar and *Triticum aestivium* cultivar for the induction of callus. The composition of MS media used was 2, 4-D, 1KT, and 3 % sucrose. For the regeneration purpose, a basal salt of MS without any hormones was utilized. Therefore, variety of variations showed by both the cultivars. Regeneration was not initiated by the medium with 2, 4-D. but for some of the cultivars of wheat Naphthaleneacetic acid (NAA) and KT were most effective regarding the formation of different organs. That showed that for regeneration the medium and genotypes are dependent. Explants' frequency of regeneration can be increased with the help of different growth hormones. Hassan *et al.* (2009) used six diverse wheat genotypes and compared the results of callus induction with the process of regeneration in media of MS in varying concentrations of the carbohydrate sorbitol the varying concentrations of the sorbitol, different genotypes thus show different results. Shimada & Makino (1975) noticed the maximum value of frequency of the induction of the callus, which had concentration of sorbitol in increasing amount from 0–20 $g.L^{-1}$. in contrast to that, Szakacs *et al.* (1988) noticed the minimum value of frequency of the induction of the callus. Therefore, from the above results, it has been concluded that with the increased in sorbitol concentration, there were chances of the increase in frequency of the regeneration but time duration is lowered. Guo *et al.* (1982) narrated the same outcomes. Bahieldin *et al.* (2000) reported that in the regeneration process, Dicamba has huge importance. Three spring wheat genotypes had some immature embryos, which were used for checking the response of this dicamba on the rate of calli regeneration and shoot formation. The lower concentration of the plant dicamba give reciprocal effect on the shoot of per callus as the process of regeneration was totally genotype dependent.

RESPONSE OF CALLUS CULTURE IN SALTS

Akhtar *et al.* (2007) used MS media along with different concentrations of chloride salts for wheat genotypes. Then the frequency of the regeneration of these different varieties was observed under sodium chloride and calcium chloride stress that showed considerably differences in diverse wheat genotypes. Among these four, LU-26 genotypes were declared most resistant genotypes. Abdullah *et al.* (2012) showed the salinity effects on both without salt or with salt on callus regeneration capacity. Those genotypes with indole acetic acid (IAA) and kinetin were mixed with the different concentrations of NaCl. By the addition of the salt low generation of shoots number and low rate of callus regeneration was observed. Moreover, that callus which was induced usually in the presence of the salt NaCl gives a rate of regeneration at 48% along with number of shoots of as per callus that were generated of two shoots. This rate is relatively higher as compared to those that have low amount of salt or no amount of saltinity at 32% along with number of shoots of as per callus that were generated of 1.6 shoots. Regeneration from callus induced in the presence of salt can screen the cells within a callus capable of regenerating plantlets. Salinity affects the rate of growth and the growth rate of relatively fresh weight. If the salinity increases, the reduction in the rate of relatively fresh weight occurs. The results that were seen of rate of relatively fresh weight were as after four weeks as: 0.1831 g, 0.1884 g and 0.1894 g for AS-2002,

Punjab- 76 and Chenab-70 respectively. Chen (1986) also reported a reduced rate of relatively fresh weight. Karadimova & Djambova (1993) recorded that with the progressing concentration of NaCl apparently necrosis and brownish color was caused that resulted in the lowering of the growth of the callus. Arzani & Mirodjagh (1999) described the similar experience related to the diverse durum wheat genotypes.

RESPONSE OF CALLUS CULTURE IN GROWTH HORMONES

Farooq *et al*. (2004) showed the results of the responses of three different wheat genotypes *i.e.* Inqilab-91, Bakhtawar-92 and Punjab-96 subjected to different media combinations. Bakhtawar-92 was considered best suitable genotype for the induction of the callus. This had the rate of regeneration frequency as 40 %. This result was for the media with BAP 2.5 $mg.l^{-1}$ and IAA 0.1 $mg.l^{-1}$ as compared to the Inqilab-91 and Punjab-96 that showed the rate of regeneration frequency as 33 % and 25% respectively on the specified medium having BAP 0.5 $mg.l^{-1}$ and IAA 0.1 $mg.l^{-1}$ finally he gave results by concluding that the media having 2, 4-D shows maximum rate of regeneration frequency of callus. Shah *et al.* (2003) reported the utilization of various growth regulators required for the are also used in wheat for the induction of the callus and also in many other cereals like Kinetin, BAP, IAA and 2, 4-D. Bouiamrine *et al.* (2013) placed immature embryo of four cultivars of *durum* wheat media with varying concentrations of the ABA along with 2, 4-D 2 $mg.l^{-1}$ and then the ability of the regeneration was observed under the treatment of absicic acid. Results declared that the increase in ABA concentration inside the medium, cause reduction in callus growth. Hence lowest concentrations of the ABA cause increase in the frequency of the callus regeneration. Soliman *et al*. (2013) used six durum wheat genotypes. This experiment was done to check the PEG response on calli culture necrosis, regeneration and water content of growth. The result helped in the conclusion that the process of regeneration and parameters of callus growth were gradually decreased in the specific media with increased PEG concentration.

Various effects of ascorbic acid were reported by Fazelienasab *et al.* (2004) on five different Iranian bread wheat cultivars. He used varying concentrations in media of MS of abscisic acid (ABA) along with sucrose as source. Huge difference was seen in callus induction among these five diverse cultivars and with the enhancement of ABA concentration reduction in callus induction was observed. However, when the conditions were controlled, optimized results were seen with glowing colors in callus but in the same way higher concentrations of the ABA, the color of the callus is lowered. The conclusion was then made about the interaction of the ABA and the five cultivars that was highly important. Gaspar *et al.* (1996), Mzouri & Amssa (2002) and Carman *et al.* (1987) reported the influence about the callus induction in cereals by cytokinens and auxin along with the morphogenesis. Wheat has mature embryos that were used by Mendoza & Kaeppler (2001). They also used four types of auxins that were Propionic acid, Picloram, Dicamba and 2, 4-D for checking the sugar and auxins effects on the callus induction and the ability to regenerate. They also compared sucrose and maltose effects on the callus induction under all the optimized and sterilized conditions. Propionic acid is the only acid that does not has good effect on the callus induction while all other forms of auxins are considered beneficial. Sugar effect on the callus induction is usually depending on the treatment of auxins. Combination of 2, 4-D and maltose caused enhancement in the regeneration frequency. However, when 2, 4-D was used in combination with that of Dicamba, they lowered the frequency of regeneration. The Picloram use also induced the callus induction and the frequency of the regeneration.

Fahmy *et al.* (2012) used two media in his findings that induced the callus *i.e.* CIM1, CIM2 and five different media for regeneration *i.e.* WRM1, WRM2, WRM3, WRM4 and WRM5 have varying response on the wheat cultivars of various Egyptian alite was also clearly checked. From his experimentations in Green house, he made an opaque conclusion that medium WRM2 showed the greater shoot number per callus that was cultured. Haliloglu (2006) reported in his study that 6-Benzyl Aminopurine, Kinetin and Indole-3-Acetic acid have a remarkable high role in the plantlets regeneration.

Salt stress was applied to the calli of the genotypes of ten different wheat plants. These were further used for the development and regeneration of plant in the MS media with no auxin *e.g.* 2, 4-D. The result obtained via this experiment was in accordance with Varshney *et al*. (1991) and Hussain *et al*. (2009) in which explants of immature embryos of *triticum aestivium* was used. They observed that plants were easily regenerated on that MS media without hormone. Farshadfar *et al*. (2012) utilized wheat immature embryo for checking twenty bread wheat genotypes response *in-vitr o*f callus induction and the process of regeneration under drought stress. They revealed that's the variations was present among genotypes for callus chlorosis percentage, callus water content and relative growth rate of callus usually at drought level. Drought stress caused by Polyethylene glycol

(PEG) was compared at different concentrations and various effects were observed at varying stress levels in *in vitro* tolerance (INTOL), callus relative growth (CRG), callus growth rate (CGR), percentage of callus chlorosis (PCCH) and percentage of callus water content (PCWC), callus relative growth rate (CRGR), and percentage of callus water content (PCWC) and these effects were lowered with the increased drought effect.

CONCLUSION

Wheat (*Triticum aestivum* L.) is consideredas most vital cereal crop andplant breeders took interest to boost up its productivity. Since several years, numerous efforts have been made to boost up its productivity under various conditions through numerous techniques especially tissue culture. In this review, we focused on responses of diverse wheat genotypes under different Media, salts and growth hormones. This comprehensive review showed tremendous work of many scientists in field of tissue culture of wheat to overcome poor tissue culture technique and transformation of wheat cultivars. So there is a need of time explore various techniques in tissue culture to develop genotype having better performance.

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