J. of Plant Production, Mansoura Univ., Vol. 14 (9): 429 - 434, 2023

Journal of Plant Production

Journal homepage & Available online at: www.jpp.journals.ekb.eg

Evaluation of Gelling Agent Alternatives during *in Vitro* Multiplication of *Yucca elephantipes* L.

Abass, M. M. M.[1*]; R. S. Thabet[1]; M. F. G. Abdelfattah[1] and Asmaa A. Fahmy[2]

[1] Department. of Ornamental Plant Research Institute, Agriculture Research Center, Giza, Egypt.
[2] Horticulture Department of, Faculty of Agriculture, Zagazig University, Egypt

ABSTRACT

In vitro hardening materials like Agar and Phytagel are increasing in price, therefore cheaper gelling agents were needed. In this study, potato starch (*Solanum tuberosum*) and corn starch (*Zea mays* L.) at different rates (40, 50, 60, and 70 g/l) each alone or combined with agar at 2 or 4 g/l as well as 8 g agar/l as control were tested during *in vitro* multiplication of yucca (*Yucca elephantipes* L.). However, the first experiment, shoot proliferation percentage was 100% with all treatments (potato starch + agar) under study, media status was solid with any combination between potato and agar except that of potato alone was semi solid status. On the other hand, MS medium supplemented with potato alone at 70 g/l produced the most leaves per shoot and shoot length, while MS medium supplemented with agar at 4 g/l + potato at 40 g/l produced the greatest shoots per explant, with significant differences from the other treatments. For the second experiment, shoot proliferation percentage was 100% as well as media status was solid with all treatments (corn starch + agar) under study. MS medium supplemented with corn alone at 70 g/l provided the highest leaf number per shoot and shoot length, while MS medium supplemented with agar at 2 g/l + corn at 40 g/l produced the most shoots per explant with significant differences from the other treatments. Consequently, the present work suggests a low-cost alternative gelling agent that will reduce plant micro propagation production costs.

Keywords: *Yucca elephantipes*, *In vitro*, gelling agent, shoot proliferation.

INTRODUCTION

Yucca (*Yucca elephantipes* L.) plant belongs to Family *Asparagaceae* or *Agavoideae* (Rentsch, 2013). Yucca is a public plant used for indoor ornamentation and some of yucca species had medicinal usage (Cheeke *et al*., 2006 and Patel, 2012). Also, it is an evergreen shrub single-stemmed and medium-sized. Plant leaves are sword-shaped, stiff, striped, spine-tipped, 35-55 cm long, 5 cm wide, long pointed, and mostly in rosettes at the ends of plant trunk (Abdul-Halem *et al*., 2015).

One of the most important factors that affect the chemical and physical characteristics of the culture medium *in vitro*, is the type and concentration of gelling agent. Gelling agents make the medium firm and influence the diffusion characteristics of the medium. Consequently, solidifying agents can significantly impact the morphogenetic response, growth, and development of tissue cultured plant material (Das *et al*., 2015 Mohamed *et al*., 2021). Moreover, they can contribute to the occurrence of hyperhydricity (also known as vitrification) which is a common physiological disorder that causes shoots and leaves to become brittle, with a glassy appearance (Amer and Omar, 2019).

Agar is the most expensive substance in the media which contributes around 70% of costs (Mpatani and Vuai , 2019)

The layout of setback effective micropropagation protocols is a prerequisite in the adoption of the low cost micro propagation technology in growing countries. Agar has stayed the ultimate frequently utilized gelling agent for culture media used for both microbes and plants. The agar properties, which create it a gelling agent of choice, are its high clarity, steadiness, its resistance to metabolism during culture and non-toxic nature (McLachlan, 1985; Henderson and Kinnersley, 1988). Prakash (1993) pointed out that a mixture of potato starch with agar in a ratio of (2:1) reduced the cost of gelling agent by 70-82%. Zimmerman *et al*. (1995) revealed that medium gelled with corn starch at 50 g/l + gelrite at 0.5 g/l was completely as efficacious as medium gelled with agar for apple and raspberry shoot proliferation. Also, Lalitha *et al*. (2014) indicated that corn flour at 22 g/l in combination with 3.5 g/l agar produced significantly higher and healthy micro-shoots of length. In addition, Ullah *et al*. (2015) suggested that the MS medium supplemented with the increasing rate of corn and potato (from 40 to 50 g/l) gelled with agar (2 g/l) progressively improved orchid growth performance.

Gelling agents used to solidify plant growth medium can contain many mineral nutrients reported to affect plant growth (Joshi, 2009). Generally, many studies have shown that the type of gelling agent used can influence tissues growth *in vitro* (Quiala *et al*., 2014; Ramesh and Ramassamy, 2014, 2015; Al-Mayahi, 2015; Raina, 2017; Hegele *et al*., 2021 Sánchez-Gutiérrez 2023), therefore the study on selecting commercial grades of gelling agents is very important. There are limited studies about gelling agent effect routinely used in micropropagation on palm plant growth and development (Al-Mayahi, 2021; Palanyandy *et al*., 2020).

Several alternative gelling agents, such as corn starch, barley starch, white flour, laundry starch, semolina, potato starch, rice powder, sago, sweet potato, maize, rice, wheat,

[*] Corresponding author.
E-mail address: mmmabass1984ag@gmail.com
DOI: 10.21608/jpp.2023.221576.1254

sorghum, cassava and opuntia pads have been proposed, (Ullah *et al*., 2015; Teixeira da Silva, 2015; Amlesom *et al* 2021 Biswas *et al* 2021 Sánchez-Gutiérrez, *et al*., 2023).

The present study was undertaken to look for additional easily and cheap available substitutes of agar to be utilized in tissue culture media to enhance *Yucca elephantipes* callus weight, number of leaves and shoots per cluster as well as shoot length during multiplication stage.

MATERIALS AND METHODS

The present study was carried out at the Plant Tissue Culture Laboratory of Horticulture Department, Faculty of Agriculture, Zagazig University, Egypt from January to September 2020. This study involved two experiments. The first one was to determine the effect of different rates (40, 50, 60 and 70 g/l) of potato starch either alone or combined with 2.0 or 4.0 g/l agar as well as 8.0 g/l agar as a control on shoot multiplication and growth of *Yucca elephantipes* L. during multiplication stage. The second one was devoted to assessing the influence of different rates (40, 50, 60 and 70 g/l) of corn starch either alone or combined with 2.0 or 4.0 g/l agar as well as 8.0 g/l agar as control on shoot multiplication and growth of *Yucca elephantipes* L. during multiplication stage.

Plant material source:

Explants shoot tips about 1.5- 2.0 cm in length of *Yucca elephantipes* were obtained from Plant Tissue Culture Laboratory, Faculty of Environ. Agric. Sci., Arish University, Egypt.

Culturel media and growth conditions during the multiplication stage:

The culture medium was Murashige and Skoog (1962) basal medium. The medium was supplemented with 1 mg/l BA, 30 g/l sucrose. The pH was adjusted to be 5.8 before autoclaving at 120 °C for a duration of 20 min under a pressure of 1.2 kg/cm². Jars (60 × 120 mm) were used as culture vessels and each one was filled with 50 ml medium. The cultures were placed in an air-conditioned incubation room at a temperature of 25 ± 2 °C under 16 h/ day photoperiod which provided by cool white fluorescent lamps (light intensity of 2000 Lux). The explants at this stage were separated and transferred to MS medium supplemented with 1 mg/l benzyladenine (BA) with different rates of gelling agents source (potato or corn).

Recorded data

Growth traits:

After 6 weeks from culturing the following data were recorded: Shoot proliferation (%), media status, callus fresh and dry weights/cluster (g), callus fresh and dry weights/shoot (g), number of leaves/shoot, number of shoots/explant and shoot length (cm).

Chemical constituents:

After 6 weeks from culturing vegetative parts (shoots+ leaves) were used to determine some chemical constituents. Total carbohydrates, total nitrogen, total phosphorus and potassium percentages were estimated as outlined in A.O.A.C. (2012).

Statistical analysis:

The experimental design was completely randomized design. The collected data were subjected to statistical analysis according to Snedecor and Cochran (1980). Mean separation was done using the least significant different test (LSD) at 5% level by using computer program of Statistix version 9 (Analytical software, 2008).

RESULTS AND DISCUSSION

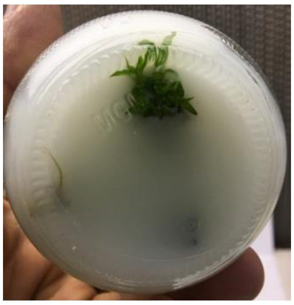
Results:

Effect of potato starch as gelling agents on *Yucca elephantipes* L. shoot growth and multiplication during the multiplication stage:

As shown in Tables 1 and 2 and Plat. 1 supplementation of the medium with potato starch alone without agar addition produces semi-solid media status. While MS media supplemented with agar alone at 8 g/l or all combination treatments between agar and potato starch produced solid medium. Furthermore, all rates of potato as a gelling agent (40, 50, 60 and 70 g/l) alone or in combination with agar (at 2.0 or 4.0 g/l) recorded 100 % shoot proliferation. In addition, using 70 g/l potato starch individually as gelling agent produced the maximum growth values for all *Yucca elephantipes* L. growth parameters i.e., fresh and dry weight of cluster and shoot (g) as well as the number of leaves/shoot and shoot length (cm), which gave, 5.91 and 0.45 (g) as well as 13.05 and 3.47 cm; respectively with increasing percentages estimated by 14.76% and 36.36% as well as 54.44% and 160.90 % for these traits, respectively compared with control mean values (8 g/l agar). Moreover, the combination between 4 g/l agar and 40 g/l potato starch as gelling agent significantly produced the maximum value regarding the number of shoots/explant (16.40) compared to the control and the other combinations under study.

Control (8g/l Agar)

70 g/l of Potato starch



2g/l Agar + 40 g/l Corn starch

Plat. 1. Using different gelling agents on *Yucca elephantipes* L. growth during the multiplication stage

Table 1. Influence of different potato starch (PS) rates alone or combined with 2 or 4 g/l of agar on shoot proliferation percentage, media status and fresh and dry weights of shoot per cluster of *Yucca elephantipes* as compared with 8 g/l agar

Gelling agent (g/l)	Shoot proliferation (%)	Medium status	Weight of cluster (g)		Weight of Shoot (g)	
			Fresh	Dry	Fresh	Dry
8 Agar (control)	100	Solid	5.15 ab	0.33 c	0.534b-f	0.034c-d
40 PS	100	Semi solid	4.70 b	0.42 a	0.338d-f	0.031d-f
50 PS	100	Semi solid	5.67 a	0.41 ab	0.722 bc	0.052b-f
60 PS	100	Semi solid	5.91 a	0.45 a	1.359 a	0.013 a
70 PS	100	Semi solid	2.12 f	0.18 de	0.855 b	0.073 b
2 Agar + 40 PS	100	Solid	2.10 f	0.19 de	0.156 g	0.014 f
2 Agar + 50 PS	100	Solid	2.94 de	0.18 de	0.601b-e	0.037c-f
2 Agar + 60 PS	100	Solid	3.87 c	0.34 bc	0.310e-g	0.027ef
2 Agar + 70 PS	100	Solid	1.26 g	0.14 e	0.537b-f	0.059bc
4 Agar + 40 PS	100	Solid	3.44 cd	0.23 d	0.213fg	0.014 f
4 Agar + 50 PS	100	Solid	1.62 fg	0.14 e	0.658b-d	0.055b-d
4 Agar + 60 PS	100	Solid	2.16 ef	0.17 de	0.450c-g	0.036c-f
4 Agar + 70 PS	100	Solid	1.38 fg	0.12 e	0.340d-g	0.029d-f

Table 2. Influence of different potato starch **(PS) rates alone or combined with 2 or 4 g/l of agar on the number of leaves and number of shoots per cluster and shoot length (cm) of *Yucca elephantipes* L. as compared with control (8 g/l agar)**

Gelling agent (g/l)	No. of shoots/explant	Shoot length (cm)	No. of leaves/shoot
8 Agar (control)	11.70 cd	1.33 c	8.45 de
40 PS	14.80 ab	1.76 bc	8.88 c-e
50 PS	9.20 de	1.84 bc	9.66 b-e
60 PS	5.60 fg	1.63 bc	9.23 b-e
70 PS	3.50 gh	3.47 a	13.05 a
2 Agar + 40 PS	14.20 abc	1.37 c	8.96 c-e
2 Agar + 50 PS	12.70 bc	1.74 bc	9.24 b-e
2 Agar + 60 PS	7.40 ef	2.25 b	10.01 b-d
2 Agar + 70 PS	2.90 h	2.05 b	10.69 b
4 Agar + 40 PS	16.40 a	1.70 bc	8.39 e
4 Agar + 50 PS	4.20 gh	1.72 bc	9.87 b-e
4 Agar + 60 PS	5.40 fgh	2.10 b	10.15 bc
4 Agar + 70 PS	4.20 gh	1.79 bc	9.64 b-e

Effect of potato starch as gelling agents on *Yucca elephantipes* L. chemical constituents during the multiplication stage:

Results recorded in Table 3 indicate that the combination between 4 g/l agar and 60 g/l potato starch as gelling agent significantly produced the maximum values regarding total carbohydrates percentage (21.16%) and total phosphorus percentage (0.713 %) compared to the control and the other combinations, in most cases, under study. Also, the combination between 2 g/l and 40 g/l potato starch as gelling agent significantly produced the maximum value regarding total nitrogen percentage (3.09 %) compared to the control and the other combinations under study. Moreover, the combination between of 4 agar and 40 or 70 g/l potato starch as gelling agents significantly produced the maximum values regarding potassium percentage (2.10 and 2.08 %) compared to the control and the other combinations under study.

Effect of corn starch as gelling agents on *Yucca elephantipes* L. vegetative growth during multiplication stage:

Data presented in Tables 4 and 5 reveals that all rates of corn starch (g/l) as a gelling agent (40, 50, 60 and 70 g/l) alone or in combination with agar (at 2.0 or 4.0 g/l) recorded 100 % shoot proliferation as well as these treatments also attributed to obtaining a solid media status. Furthermore, MS media supplemented with agar alone at 8 g/l or all combination treatment between 2 g/l agar and 40 g/l corn starch gave the highest values in fresh and dry weights of callus /cluster without significant differences between them in the second season. Using corn starch at 70 g/l alone or plus 4 g/l agar as gelling agent gave the highest values concerning the number of leaves/ yucca shoots (12.95 and 12.49 leaves) during the multiplication stage with no significant differences between them. Also, the combination between 4 g/l agar and 40 g/l corn starch as gelling agent significantly recorded the maximum value in the number of shoots/explant (18.90 shoots) compared to the control and the other combinations under study. Similarly, using 60 g/l corn starch plus 2 g/l agar and 70 g/l corn starch alone as gelling agent produced the maximum values in shoot length of yucca elephantipes L., which gave, 2.67 and 2.41 (cm), respectively with increasing percentages estimated by 100.75 % and 81.20% for these traits, respectively compared with control mean value (8 g/l agar).

Table 3. Influence of different potato starch **(PS) rates alone or combined with 2 or 4 g/l of agar on total carbohydrates, total nitrogen, total phosphorus and potassium percentages of *Yucca elephantipes* L. as compared with control (8 g/l agar)**

Gelling Agent (g/l)	Total carbohydrates %	Total nitrogen %	Total phosphorus %	Potassium %
8 Agar (control)	18.34 i	2.62 i	0.675 e	1.80 fg
40 PS	18.39 hi	2.78 f	0.686 d	1.68 i
50 PS	19.06 g	2.98 c	0.710 ab	1.65 i
60 PS	19.36f	2.59 j	0.699 c	1.73 h
70 PS	19.77 e	3.03 b	0.665 f	1.83 f
2 Agar + 40 PS	20.11 d	3.09 a	0.702 c	1.92 d
2 Agar + 50 PS	18.04 j	2.47 k	0.678 e	1.78 g
2 Agar + 60 PS	18.45 h	2.45 k	0.712 ab	1.88 e
2 Agar + 70 PS	18.99 g	2.69 h	0.699 c	1.90 de
4 Agar + 40 PS	20.22c	2.75 g	0.708 b	2.10 a
4 Agar + 50 PS	20.03 d	2.88 e	0.702 c	1.98 c
4 Agar + 60 PS	21.16 a	2.86 e	0.713 a	2.01 b
4 Agar + 70 PS	21.02 b	2.93 d	0.683 d	2.08 a

Effect of corn starch as gelling agents on *Yucca elephantipes* L. chemical constituents during the multiplication stage:

Results listed in Table 6 pointed out that using 70 g/l corn starch individually as gelling agent produced the maximum chemical constituents values for total carbohydrates, total nitrogen, total phosphorus and potassium percentages which gave, 20.73, 2.86, 0.722 and 2.05 (%); respectively. In addition, the combination between 4 g/l agar and 50 g/l corn starch as gelling agent significantly produced the maximum value regard total carbohydrates percentage (20.24) compared to control and the other combinations under study. Also, the combination between 2 g/l or 4 agar and 60 or 40 g/l corn starch as gelling agent significantly produced the maximum values regard total nitrogen percentage (2.83 and 2.83 %) compared to control and the other combinations under study. Moreover, the combination between 2 g/l or 4 agar and 70 or 50 g/l corn starch as gelling agent significantly produced the maximum values regard potassium percentage (2.04 and 2.03 %) compared to control and the other combinations under study.

Table 4. Influence of different corn starch (CS) rates alone or combined with 2 or 4 g/l of agar on shoot proliferation percentage, media status and fresh and dry weights of shoot per cluster of *Yucca elephantipes* L. as compared with 8 g/l agar

Gelling agent (g/l)	Shoot proliferation (%)	Media status	Weight / cluster (g)		Weight of Shoot (g)	
			Fresh	Dry	Fresh	Dry
8 Agar (control)	100	Solid	5.15 a	0.33 a	0.534ab	0.034bc
40 CS	100	Solid	2.11 c-f	0.17 b-d	0.214cd	0.018c
50 CS	100	Solid	2.21 c-e	0.18 bc	0.199cd	0.016c
60 CS	100	Solid	1.90 c-g	0.19 b	0.188d	0.019bc
70 CS	100	Solid	1.88 d-g	0.17 b-d	0.626a	0.056a
2 Agar + 40 CS	100	Solid	3.35 b	0.28 a	0.202cd	0.017c
2 Agar + 50 CS	100	Solid	2.38 cd	0.19 b	0.279cd	0.022bc
2 Agar + 60 CS	100	Solid	1.28 g	0.12 cd	0.422a-c	0.039ab
2 Agar + 70 CS	100	Solid	1.37 fg	0.11 d	0.347b-d	0.028bc
4 Agar + 40 CS	100	Solid	1.52 e-g	0.15 b-d	0.191d	0.019c
4 Agar + 50 CS	100	Solid	2.68 bc	0.21 b	0.405a-d	0.033c
4 Agar + 60 CS	100	Solid	2.57 b-d	0.19 b	0.209cd	0.015bc
4 Agar + 70 CS	100	Solid	1.90 c-g	0.15 b-d	0.257cd	0.021bc

Table 5. Influence of different corn starch (CS) rates alone or combined with 2 or 4 g/l of agar on the number of leaves and number of shoots per cluster and shoot length (cm) of *Yucca elephantipes* L. as compared with control (8 g/l agar)

Gelling agent (g/l)	No. of shoots/explant	Shoot length (cm)	No. of leaves/shoot
8 Agar (control)	11.70 bc	1.33 d	8.45 ef
40 CS	11.20 bc	1.42 d	9.20 d-f
50 CS	11.80 bc	1.63 cd	10.45 b-d
60 CS	10.80 bc	1.44 d	10.48 b-d
70 CS	5.00 e	2.41 ab	12.95 a
2 Agar + 40 CS	18.90 a	1.55 cd	9.40 c-f
2 Agar + 50 CS	10.80 bc	1.55 cd	11.36 a-c
2 Agar + 60 CS	5.60 de	2.67 a	11.63 ab
2 Agar + 70 CS	6.00 de	1.98 bc	11.12 a-d
4 Agar + 40 CS	10.30 bc	1.57 cd	8.30 f
4 Agar + 50 CS	8.10 cde	1.57 cd	9.54 c-f
4 Agar + 60 CS	13.00 b	1.41 d	10.39 b-e
4 Agar + 70 CS	9.00 cd	1.72 cd	12.49 a

Table 6. Influence of different corn starch (CS) rates alone or combined with 2 or 4 g/l of agar on total carbohydrates, total nitrogen, total phosphorus and potassium percentages of *Yucca elephantipes* L. as compared with control (8 g/l agar)

Gelling agent (g/l)	Total carbohydrates %	Total nitrogen %	Total phosphorus %	Potassium %
8 Agar (control)	18.34 g	2.62 d	0.675 e	1.80 f
40 CS	18.33 g	2.34 g	0.608 i	1.78 f
50 CS	18.32 g	2.49 e	0.568 j	1.78 f
60 CS	19.21 ef	2.74 b	0.624 h	1.81 ef
70 CS	20.73 a	2.86 a	0.722 a	2.05 a
2 Agar + 40 CS	19.71 b-e	2.50 e	0.647 g	1.95 bc
2 Agar + 50 CS	18.80 fg	2.65 cd	0.678 e	1.93 c
2 Agar + 60 CS	19.88 bc	2.83 a	0.676 e	1.84 e
2 Agar + 70 CS	19.05 f	2.38 f	0.668 f	2.04 a
4 Agar + 40 CS	19.84 b-d	2.83 a	0.678 e	1.98 b
4 Agar + 50 CS	20.24 ab	2.73 b	0.678 e	2.03 a
4 Agar + 60 CS	19.61 c-e	2.66 c	0.704 b	1.89 d
4 Agar + 70 CS	19.33 d-f	2.71 b	0.689 c	1.69 g

Discussion:

The present work studied the influence of of potato starch as gelling agents on *Yucca elephantipes* L. shoot growth and multiplication during multiplication stage (Tables 1 and 2). Likewise, Mohamed *et al*. (2010) on *Solanum tuberosum*, pointed out that the combination of agar and potato starch could offer a firm support for plant tissues and could be successfully utilized for micropropagation. Also, these results are in harmony with those reported by Ullah *et al*. (2015) on *Dendrobium sonia*. They demonstrated that Murashige and Skoog (MS) was the best medium when supplemented with agar at 2 g/l plus potato at 50 or 60 g/l as gelling agent treatment. Moreover Amlesom *et al*. (2021) on potato micropropagation found that MS with Potato LG gave the highest plant height values as compared to the control (Agar). In addition, Mohamed *et al*. (2021) found that when gelrite was used at a concentration of 2.0 or 2.5 g/L or phytagel was used at a concentration of 2.5 or 3.0 g/L, the best callus formation was seen. Also, it was seen that rice Sakha104 cultivar was better at making calluses than Giza178, no matter what kind of hardening agent was used or how much of it was used.

A lot of researchers have tried to find other gelling agents that could replace agar from tissue culture in part or in whole. Few workers have used starches (rice, corn, barley, wheat, etc. starches), gums (locust bean gum, guar gum, gum kartia, etc.), isabgol, etc., and said that by utilizing these gelling agents, the cost of the product from tissue culture operations can be cut while growth or yield can be improved (Biswas *et al*., 2021).

The decrease on shoots number per explant which obtained from media contained 70 g/l corn starch could be due to the lowering on plant water potential below onset value for cell expansion which is a precondition for growth and shoots formation (Owens and Wozniak, 1991). Furthermore, Toaima *et al*. (2016) cleared that gelrite at the suggested concentrations of 0.2–0.25% was not enough for micropropagation of *G. paniculata*, or this type of gelling agent was not right for this species. When 8 and 10 g/l of agar were used, the amount of hyperhydricity was 13.4% and 8%, respectively. Compared to Agar Type 900, Gelrite® with a concentration of 3.5 g/l was chosen as an effective gelling agent for turning polyembryoids into plantlets because 100% of the polyembryoids survived and 53.3% of the plantlets (multiple shoots with roots) were fully formed (Palanyandy *et al*., 2020).

In the same time, Daud *et al*. (2011) on *Celosia* revealed that the combination of alternative gelling agent (40 g/l corn flour) with agar or gelling agent alone successfully regenerated shoots from stem segment explants. Also, Ullah *et al*. (2013) on orchid, indicated that the MS medium supplemented with the raising rate of corn from 50 to 60 g/l gelled with 2g/l agar progressively enhanced their growth performance than that of 8 g/l agar only. In general, Amlesom *et al*. (2021) pointed out that using corn and potato starches at 50 g/l as solidifying agent instead of agar can be efficient and cost effective for single node potato micropropagation. Also, Ibrahim *et al*. (2022) showed that agar alone and its mixtures with gillan gum at 2–6 gl-1 were the most effective gelling agents throughout the multiplication stage of Paulownia hybrid (*Paulownia elongate* × *Paulownia fortunei*).

CONCLUSION

The present study offers a new possibility of utilizing low cost alternative gelling agents (potato or corn starches) which will decrease the production costs considerably plant micro propagation techniques as presented in multiplication stage of *Yucca elephantipes* L. explants.

REFERENCES

Abdul-Halem AG, Alwash B, Ibrahim KM (2015) Response of *Yucca gloriosa* "Variegata" to tissue culture. J.of Al-Nahrain University Sci, 18 (4): 103-109.

Amlesom WS, Mehari T, Saleh BK (2021) Evaluation of Different Starches as Gelling Agents for Micropropagation of Potato. *The Journal of Agricultural Science*, *13*(5), 144. https://doi.org/ 10.5539/jas.v13n5p144

Amer A, Mohamed G, Pantaleo V, Leonetti P, Hanafy MS (2019) *In vitro* regeneration through organogenesis in Egyptian chickpea. Plant Biosyst. 153,835–842. https://doi.org/10.1080/11263504.2018.1549616.

Analytical Software (2008) Statistix Version 9, Analytical Software, Tallahassee, Florida, USA.

Al-Mayahi AMW, Ali, AH (2021) Effects of different types of gelling agents on *in vitro* organogenesis and some physicochemical properties of date palm buds, Showathy cv. Folia Oecol, 48(1), 110-117. doi: 10.2478/foecol-2021-0012

A.O.A.C. (2012) Official methods of analysis. 19th edition. 2nd revision. Gaithersburg, MD, USA, Association of Official Analytical Chemists.

Palanyandy SR, Gantait S, Sinniah UR (2020) Effects of some gelling agents and their concentrations on conversion of oil palm polyembryoids into plantlets. Journal of Genetic Engineering and Biotechnology, 18 (5): 1- 5. https://doi.org/10.1186/s43141-019-0018-z

Biswas SS, Kalaivanan NS, Pal R, De LC (2021) Substitutes of Agar For Gelling In Tissue Culture Media,Their Benefits And Drawback, A review. International Journal of Recent Scientific Research. Vol. 12, Issue, 02 (D), pp. 41044-41050. DOI: http://dx.doi.org/ 10.24327/ijrsr.2021.1202.5818

Cheeke PR, Piacente S, Oleszek W (2006) Anti-inflammatory and anti-arthritic effects of *Yucca schidigera*: a review. J. Inflamm. 3:6.https://doi.org/ 10.1186/1476-9255-3-6

Das N, Tripathi N, Basu S, Bose C, Maitra S, Khurana S (2015) Progress in the development of gelling agents for improved culturability of microorganisms.Front. Microbial. 6, 698. https://doi.org/10.3389/ fmicb. 2015. 00698.

Daud N, Taha R M, Noor NNM, Alimon H (2011) Potential of alternative gelling agent in media for *in vitro* micro-propagation of Celosia sp. Int. J. of Bot.7 92): 183-188. doi: 10.3923/ijb.2011.183.188.

Hassan HMS, Moubarak M (2020) Micropropagation of yucca plant by using guar and locust bean seed powder as an alternative cheap gelling agent. Sci J. Flowers & Ornamental Plants, 7 (3): 239-246.doi: 10.21608/ sjfop.2020.110364

Hegele S, HegeleM, Wünsche, JN (2021) Low-cost gelling agents for tissue culture propagation of plantain. In IV Int. Symposium on Hort. in Europe-SHE2021 1327 (pp. 341-348).doi: 10.17660/ActaHortic.2021.1327.46

Henderson WE, Kinnersley AM (1988) Corn starch as an alternative gelling agent for plant tissue culture media. Plant Cell Tiss. Organ Cult., 15-17.

Ibrahim SSA, Hassan HMS and Shehata SA (2022) Using explant, media, cytokinine types and gelling agents on paulownia hybrid (*Paulownia elongata* × *Paulownia fortunei*) tissue culture. SINAI Journal of Applied Sciences, 11 (3):441-452. https://doi.org/10.21608/ sinjas.2022.131725.1099

Joshi N (2009) *In vitro g*rowth and shoot multiplication in Nicotiana tabacum L. – influence of gelling agent and carbon source. *Int. J.l* of *Plant Developmental Bio.*, 3 (1): 29–33.

Ibrahim SS, Hassan H, Shehata SA (2022) Using Explant, Media, Cytokinine Types and Gelling Agents On Paulownia Hybrid (Paulownia elongata× Paulownia fortunei) Tissue Culture. *Sinai J of App Sci*, *11*(3), 441-452 DOI: 10.21608/sinjas.2022.131725.1099.

Kaur, A., Kumar, A. (2020). The effect of gelling agent, medium pH and silver nitrate on adventitious shoot regeneration in Solanum tuberosum. *bioRxiv*, 2020-01. https://doi.org/10.1101/2020.01.03.894063

Lalitha N, Devi LM, Banerjee R, Chattopadhyay S, Saha AK, Bindroo BB (2014) Effect of plant derived gelling agents as agar substitute in micropropagation of mulberry (*Morus indica* L. cv. S-1635). Int J of Advanced Res, 2 (2): 683-690.

McLachlan JM (1985) Industrial resources and their utilization. Plant Soil, 89: 137–157.

Mohamed G M, Amer A M, Osman NH, Sedikc MZ, Hussein MH (2021) Effects of different gelling agents on the different stages of rice regeneration in two rice cultivars. *Saudi Journal of Biological Sciences*, *28*(10), 5738-5744. https://doi.org/10.1016/j.sjbs.2021.06.003

Mohamed MAH , Alsadon AA, Al Mohaidib MS (2010) Corn and potato starch as an agar alternative for *Solanum tuberosum* micropropagation. Afr J of Biotechnology, 9 (1): 12-16.

Mohamed GM, Amer AM, Osman NH, Sedikc MZ Hussein MH (2021). Effects of different gelling agents on the different stages of rice regeneration in two rice cultivars. Saudi Journal of Biological Sciences, (28): 5738–5744 https://doi.org/10.1016/j.sjbs.2021.06.003

Murashige T, Skoog F (1962) A revised medium for rapid growth and bioassays with tobacco tissue. Physiol. Plant, 15:473-497.

Mpatani FM, Vuai SAH (2019) Performance of Low-Cost Agar from Gracilaria salicornia on Tissue Culture of Pleurotus HK-37. *The Sci World J, 2019* https://doi.org/10.1155/2019/2565692

Owens LD, Wozniak CA (1991) Measurement and effects of gel matric potential and expressibility on production of morphogenic callus by cultured sugar beet leaf discs. Plant Cell Tissue Organ Cult., 26: 127- 133.

Palanyandy SR, Gantait S, Sinniah UR (2020) Effects of some gelling agents and their concentrations on conversion of oil palm polyembryoids into plantlets. *Journal of Genetic Engineering and Biotechnology*, 18: art. no. 5. https://doi.org/10.1186/s43141-019-0018-z.

Patel S (2012) Yucca: a medicinally significant genus with manifold therapeutic attributes. Nat

Prod Bioprospect 2:231–234. https://doi.org/10.1007/s13659-012-0090-4

Prakash S (1993) Production of ginger and turmeric through tissue culture methods and investigations into making tissue culture propagation less expensive. Ph.D. Thesis. Bangalore Univ. Bangalor.

Teixeira da Silva JA (2015) Response of *Syngonium podophyllum* L. 'White Butterfly' shoot cultures to alternative media additives and gelling agents, and flow cytometric analysis of regenerants. *Nusantara Bioscience*, *7*(1), 26-32. https://doi.org/10.13057/nusbiosci/n070105

Toaima N, Bosila H, El-Ateeq AA (2016) *In vitro* growth regulators, gelling agents and sucrose levels affect micropropagation of Gypsophila paniculata L. *Middle East J. Agric. Res*, *5*, 313-323

Quiala E, Jimenez-Tello MV, Barbon R, Chavez M, De Feria M, La, OM, Perez M (2014) Influence of 6-Benzyladenine and gelling agent on the reduction of hyperhydricity in Tectona grandis L. *Revista Colombiana de Biotecnología*,16 (1): 129–136. https://doi.org/10.15446/rev.colomb.biote.v16n1.44279

Ramesh Y, Ramassamy V (2014) The effect of gelling agents on *in vitro* multiplication of Banana var. poovan. International Journal of Advanced Biological Research, 4 (3): 308–311.

Ramesh Y, Ramassamy V (2015) Influence of gelling agents in micropropagation of banana var. Grand Naine. *International Journal of Current* Research *in Biosciences and Plant Biology*, 2 (5): 174–178.

Raina RJ (2017) Gelling agents for plant tissue culture media: a comparative study. International Journal Innovative Research in Science and Engineering, 3 (1): 324–331

Rentsch JD (2013) Ecology and evolution of southeastern United States yucca species. Ph. D. Thesis, the Graduate Faculty of the University of Georgia, Athens, Georgia.

Sánchez-Gutiérrez AE, Soto-Zarazúa GM, Toledano-Ayala M, García-Trejo JF (2023) A Potential Alternative for Agar in *in vitro* Culture Media Based on Hydrocolloids Present in Nopal: General Structure and Mechanical Properties. In Recent Research and Advances in Soilless Culture. IntechOpen. DOI: 10.5772/intechopen.101745

Sánchez-Gutiérrez A E, Soto-Zarazúa GM, España-Sánchez BL, Rodríguez-González S, Zamora-Castro S (2023) Development of Agar Substitute Formulated with Mucilage and Pectin from Opuntia Local Waste Matter for Cattleya sp. Orchids *In vitro* Culture Media. *Processes*, *11*(3), 717. https://doi.org/10.3390/pr11030717

Snedecor WG, Cochran GW (1980) Statistical Methods.7th Ed., the Iowa State Univ. Press, Ames, Iowa, USA.

Ullah MA, Uddin M I, Puteh AB, Haque MS , Islam MS (2015) Alternative gelling agents for *in vitro* propagation of orchid (*Dendrobium sonia*). The Journal of Animal & Plant Sciences, 25(3): 792-797.

Ullah MA, Uddin MI, Haque MS , Rajib RR, Haque MA (2013) Corn and potato starch as cost effective alternative gelling agents for *In vitro* propagation of orchid. Intl. J. Bio. Res., 15 (6): 4-11.

Wilson V, Tenkouano A (2020) Cassava starch as alternative low-cost gelling agent for *in vitro* micropropagation of three Musa genotypes. Asian J. Plant Sci. Res., 10(1):8-14

Zimmerman RH (1995) Use of starch-gelled medium for tissue culture of some fruit crops. Plant Cell Tiss.Org. Cult. 43: 207–213.

تقييم بدائل عامل التصلب للإكثار الدقيق داخل الأنابيب لنبات اليوكا

محمد محسن محمد عباس[1]، رامز صابر ثابت عيسى[1] ، محمد فوزي جابر عبد الفتاح[1] و أسماء أحمد فهمي[2]

[1] قسم بحوث نباتات الزينة، معهد بحوث البساتين، مركز البحوث الزراعية، الدقي، الجيزة، مصر

[2] قسم البساتين، كلية الزراعة، جامعة الزقازيق

الملخص

نظرا للزيادة الحادة في أسعار مواد التصلب مثل الأجار والفيتاجيل لزراعة الأنسجة النباتية ، كان من الضروري البحث عن البديل لعامل التصلب. اختبر عاملين من عوامل التصلب وهما نشا البطاطس ونشا الذرة بمعدلات مختلفة (40 و 50 و 60 و 70 جم / لتر) منفردة أو بالتداخل مع الآجار بمعدل 2 أو 4 جم / لتر بجانب معاملة الكنترول 8 جم أجار/لتر لمرحلة التضاعف لنبات اليوكا. في التجربة الأولى، كانت نسبة تكوين الأفرخ 100٪ مع جميع المعاملات بين نشا بطاطس + أجار، وكانت حالة الوسط صلبة مع أي توليفة بين البطاطس والأجار باستثناء البطاطس منفردة كانت حالة البيئة شبه متصلبة. تم الحصول على أكبر عدد من الأوراق/فرخ وطول الفرخ في بيئة موراشيج وسكوج المضاف إليها 70 جم/ لتر دقيق البطاطس منفرداً، بينما تم الحصول على أكبر عدد من الأفرخ/ منفصل نباتي في بيئة موراشيج وسكوج المضاف إليها الأجار بمعدل 4 جم/لتر + 40 جم/لتر نشا البطاطس مع وجود اختلافات معنوية مع المعاملات الأخرى. بالنسبة للتجربة الثانية، كانت نسبة تكوين الأفرخ 100٪ وكذلك حالة الوسط صلبة مع جميع المعاملات (نشا الذرة + أجار) قيد الدراسة. تم الحصول على أعلى عدد أوراق/فرخ وطول الفرخ في بيئة موراشيج وسكوج + 70 جم/لتر نشا الذرة بمفرده، بينما تحقق أعلى عدد من الأفرخ/منفصل نباتي في بيئة موراشيج وسكوج + أجار بمعدل 2 جم/لتر + 40 جم/ لتر نشا الذرة مع وجود اختلافات معنوية مع المعاملات الأخرى ، في معظم الحالات. تقدم الدراسة الحالية إمكانية جديدة لاستخدام عامل التصلب البديل منخفض التكلفة والذي سيقلل من تكاليف الإنتاج إلى حد كبير في تقنيات الإكثار الدقيق.