THE UV-B INFLUENCE ON THE REGENERATION CAPACITY OF VARIOUS *ZEA MAYS* L EXPLANTS

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Abstract

Due to the decrease in atmospheric ozone, the influence of the UV-B radiation has become increasingly visible. This situation called for a reevaluation of the scientific efforts, for a better understanding of the UV radiation's effect on the plants and other organisms. The consequences of the high level of the UV-B radiation in the atmosphere are that it has increased the danger of the cytotoxic, mutagenic and carcinogenic effects. Biological systems are generally vulnerable to wavelengths between 280 and 320 nm. The increased exposure to UV-B is concerning for all organisms, but especially for plants, because of their need for light and their inability to move. For a better understanding of the significance of the changes induced by the increase in the UV-B radiation level, the scientific focus needs to shift on the research that includes biochemical, morphological and physical aspects resulted from the UV-B's actions, noticeable in the laboratory; said data needs to be validated in the field and/or in the agricultural lands. Purpose of this paper was the study of the UV-B influence on the regeneration capacity of various Zea mays L. explants. It was shown that the process of organogenesis in maize differs, depending on the explant source, the culture media and the combination of the growth regulators.

Keywords: *Zea mays L.*explants, UV-B radiation, regeneration

INTRODUCTION

The UV radiation is an important stress factor for the plants, which can lead to the alteration of the genetic system and of the cellular membranes' structure, as well as to a number of metabolic processes. The UV-B has a more destructive effect on plants due to the fact that the macromolecules as DNA (deoxyribonucleic acid) or protein have a higher absorption of sunrays at the 280-320 nm (Casati et al, 2004, Casati et al, 2003, Paul et al, 2003).

In order for the plants to develop, one needs to know exactly the optimal humidity conditions and precisely when to use chemicals and irradiation. A number of studies have shown that plants respond to the UV-B's action with a high variability (Băra, 2008, Băra et al, 2006, Basiouni, 1986).

For a better understanding of the significance of the changes induced by the increase in the UVB radiation level, the scientific focus needs to shift on the research that includes biochemical, morphological and physical aspects resulted from the UVB's actions, noticeable in the laboratory; said data needs to be validated in the field and/or in the agricultural lands (Cachiță et al, 2008, Caldwell et al, 1989, Caldwell et al, 1983).

The natural conditions and the changes of it, via the phenomena that occur, definitely determine changes in the composition of the parts of plants and in the way of their development (Bungău, 2014, Bungău et al, 2011, Bungau et al, 2011, Bungău et al, 2003); default, all these external conditions influence the people's lives.

I tackled this topic because the indirect changes caused by the UVB radiation (such as changes in the plant's form, in how nutrients are distributed inside the plant, changes during different stages of development or in the secondary metabolism) may be as important or even outweigh the direct negative effects of the UVB radiation (Pop et al, 2011, Rao, 2001, Barnes et al, 1988, Rao et al, 1996).

These changes can have important ramifications in maintaining the balance of the ecological system, preserving certain plant genotypes and even the social and economic life (Pop et al, 2011, Perry et al, 1999).

MATERIAL AND METHODS

In the vegetal vitro cultures, the proper selection of the biological material is a decisive factor, in terms of the natural qualities that the cells of the phytoinoculs plants will have, in marking their positive reactivity, as the sterilization procedures may profoundly affect the vitality and regenerative capacity of the *in vitro* cultivated cells or tissues (Băra et al, 2003, Sancar, 1994).

The material used to initiate the vitro culture consisted of caulinary, apical explants, 1 mm in size, taken from young shoots of *Zea mays*, ZP471 Helga hybrids, grown in the field.

The material was ground and pressed on the blade of a microtome, yielding a sample that was subjected to 405 nm wavelength UV radiations for 3 hours. During this time, the enzymatic reaction was periodically monitored (Rowland, 1991).

A great induction capacity, as well as the reliability regarding its properties and physiological basis, have led to the conclusion that the elite line of the ZP471 and Helga maize hybrids is, by far, the best to be used in research, considering the suggested protocol and the topic (Rozema, 2002, Rao et al, 1985, Roberts, 1965).

We focused our attention on these, carrying out the various stages of treatment: at regular intervals (before blooming, during and a week after that), the plants were watered and administered nutrients in order to eliminate any type of stress.

For the collection of meristems and their inoculation in the culture medium, in order to initiate appropriate *in vitro* cultures, to test their efficiency, two basic media were used: Murashige-Skoog (1962), abbreviated MS and Lindsmaier-Skoog (LS), but also LB (Sambrock 1989) and MSTop – Agar (0.5% agar) compound fertilizer (Cachiță et al, 2008, Casati et al, 2004, Rozema, 2002).

Table 1.

Bio-stimulating substances used in in vitro maize cultures

Type of hormone [mg/L]	Initiation of in vitroculture	Introduction of callus genesis	Callus regeneration
ANA	0.5	1	0.5
BAP	1	0.5-1.5	-
Sucrose	20	20	-
Thiamin	0.35	1.2	-
Pyridoxine	0.5	5	-

RESULTS AND DISCUSSIONS

The *in vitro* evaluation of the segment culture was carried out over 8 weeks since the incubation and consisted of determining the length of the shoots.

It was shown that the process of organogenesis in maize differs, depending on the explant source, the culture media and the combination of the growth regulators. This fact is reflected in the difference in size between the developed Helga and ZP471 shoots, after the irradiation with different wavelengths.

Results obtained are presented in table 2 and in figure 1, 2 and 3.

Table 2.

The size of the shoots (mm) developed from meristems grown in the MS and LS media and UV-B type irradiation, different wavelengths, in mm, after 20, 40, 60 days of culture

Utilized variety	UVB Λ [nm]	Stalk meristem					
		MS			LS		
		20 zile	40 zile	60 zile	20 zile	40 zile	60 zile
Helga	280	0.9	1.9	3.9	0.85	1.75	3.75
	285	0.98	1.95	3.95	0.88	1.95	3.85
	287	1.1	2.1	4.8	1.12	2.15	4.50
	295	0.98	1.95	4.95	0.98	1.90	4.55
	310	0.7	1.3	2.3	0.65	1.45	2.55
ZP471	280	0.75	1.75	3.75	0.70	1.80	3.65
	285	0.79	1.75	3.75	0.75	1.86	3.70
	287	0.8	1.8	4.2	0.82	1.82	4.35
	295	0.73	1.75	3.75	0.75	1.78	3.85
	310	0.6	1.6	3.6	0.60	1.55	3.55

Table 2 -continuation.							
Utilized variety	UV-B Λ [nm]	Apical meristem					
		MS			LS		
		20 zile	40 zile	60 zile	20 zile	40 zile	60 zile
Helga	280	1.25	1.85	3.55	1.15	1.80	3.65
	285	1.20	1.90	3.60	1.18	1.86	3.60
	287	1.26	1.95	3.85	1.25	1.90	3.65
	295	1.20	1.90	3.70	1.22	1.85	3.70
	310	1.05	1.55	2.30	1.00	1.50	3 .0
ZP471	280	0.9	1.9	3.20	0.98	1.85	3.0
	285	0.95	1.95	3.20	0.98	1.95	3.10
	287	0.10	2.10	3.25	1.10	2.15	3.15
	295	0.10	2.10	3.15	1.00	2.00	3.10
	310	0.7	1.3	2.85	0.65	1.50	2.3

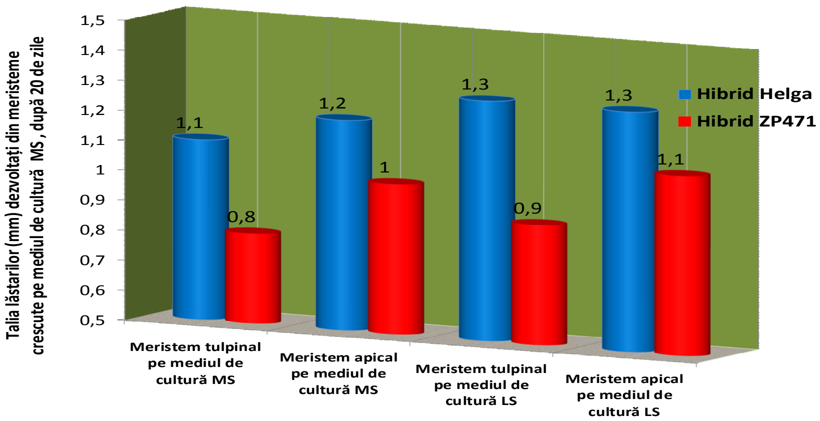

Fig. 1. The size of the shoots (mm) grown on culture media, after 20 days

Therefore, the stalk meristem is recommended for the *in vitro* regenerative experiments, due to its increased tolerance toward UVB. Note the similar reaction of the genotypes to the other wavelenghts, indicating the more prononced effect of the regenerative processes, independently of the explant type or the used culture medium.

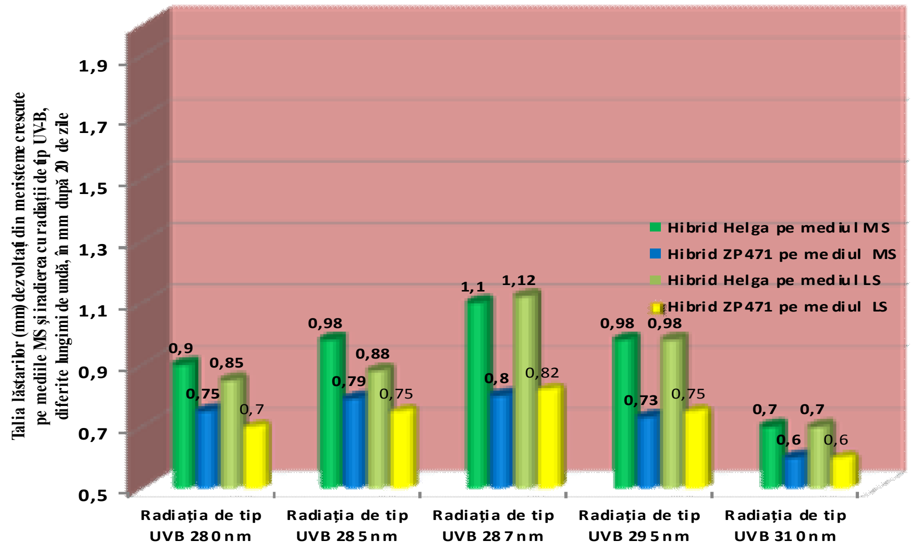

Fig. 2 The size of the shoots (mm) developed from stalk meristems, grown on the LS and MS media, in mm, after 20 days of culture, under different wavelength UV-B influence, for Zea mays L., the Helga and ZP471 hybrids.

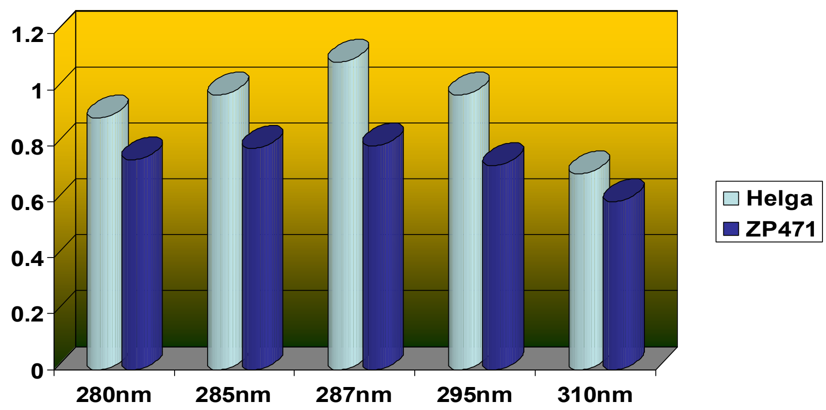

Fig. 3 The size of the shoots (mm) developed from stalk meristems, grown on the LS and MS media, in mm, after 20 days of culture, under different wavelength UV-B influence, for *Zea mays L.*, the Helga and ZP471 hybrids.

CONCLUSIONS

The comparison of the culture media MS and ML, with the same level of growth regulators, in terms of the neoplantlets development, highlights a slight superiority of the LS medium compared to the MS one, with insignificant differences in the observed parameters.

An irradiation of 287 nm wavelength makes it more relevant, reassuring us that the use of the two culture media is suitable for the *in vitro* multiplication of the maize.

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