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The Biopositive Effects of Diagnostic Doses of X-rays on Growth of *Phaseolus vulgaris* Plant : A Possibility of New Physical Fertilizers

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Abstract : The aim of this study was to evaluate the bio-effects of low doses of diagnostic X-rays on growth rate of *Phaseolus vulgaris* (Pinto) plants.

Before cultivation, *Phaseolus vulgaris* (Pinto) seeds were soaked in tap water for 2 days followed by another 2 days of covering under a wet cloth. Fifteen days after starting cultivation, newly grown plants were irradiated with X-rays. Plants were exposed to a single dose of X-ray (80 kVp, 80 mAs) for 6 days. On day 29, plants were pulled out from the soil. Length of plant stem, length of root, number of leaves and plant weight were measured.

The stem length in irradiated and sham-irradiated plants was 296.51±13.57 and 223.96±15.02 mm, respectively. This difference was statistically significant ($P<0.001$). Although the number of leaves in irradiated plants was higher than that of sham-irradiated plants (7.05±0.18 and 6.74±0.19, respectively), the difference was not statistically significant. The stem diameter in irradiated and sham-irradiated plants were 3.52±0.12 and 3.35±0.09 mm, respectively, but the difference again was not statistically significant ($P<0.001$). Plant weight in irradiated samples was less than that of non-irradiated plants, ageing being statistically non-significant.

The overall results indicate that diagnostic doses of X-rays can accelerate the growth of plants. The growth enhancement ratio for stem length was 1.33 that is a challenging figure. However, current data seem to be insufficient and further studies are needed to confirm these findings as well as to find out the possible hormetic mechanisms.

Key Words : Ionizing Radiation, Diagnostic Radiology, Plant Growth, and Fertilizers

Introduction

Over the past decades some pioneer scientists reported that low-dose ionizing radiation is not only a harmless agent but often has a beneficial or hormetic effect (Fig. 1). That is, low-level ionizing radiation may be essential trace energy for life, analogous to essential trace elements. Despite the fact that high doses of ionizing radiation are detrimental, substantial data from both humans and experimental animals show that biologic functions are stimulated by low dose radiation (Luckey, 1980). In the early 1940s C. Southam and his coworker J. Erlish found that despite the fact that high concentrations of Oak bark extract inhibited fungi growth, low doses of this agent stimulated fungi growth. They modified Starling's word "hormone" to "hormesis" to describe stimulation induced by low doses

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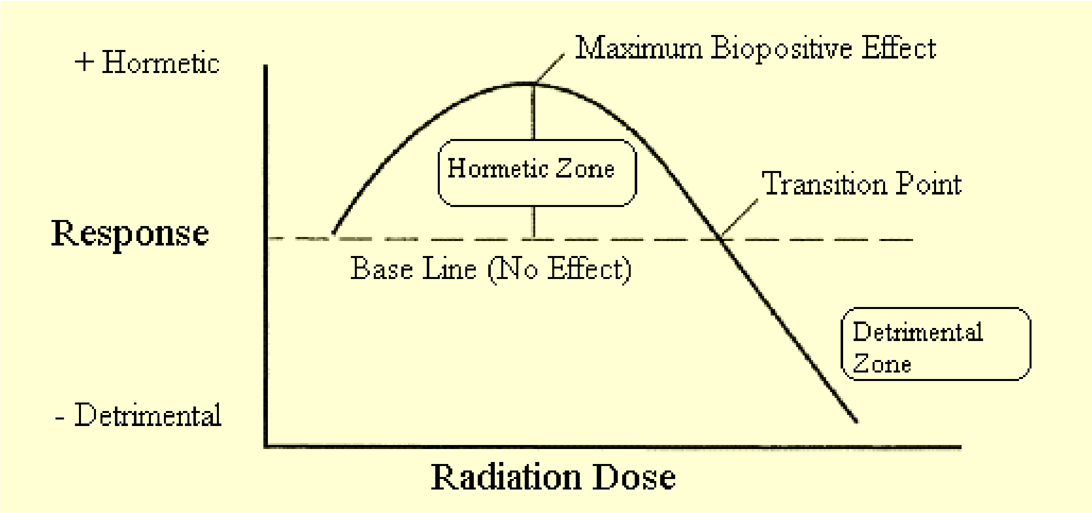

Fig. 1 : General Concept of hormesis. Chemical or physical toxins often have a detrimental effect on the health of living organisms at high doses while low doses show a biopositive effect.

of agents which are harmful or even lethal at high doses. They published their findings in 1943 (Bruce, 1987). Generally, hormesis is any stimulatory or beneficial effect, induced by low doses of an agent that can not be predicted by the extrapolation of detrimental or lethal effects induced by high doses of the same agent (Mortazavi *et al*., 1999, Mortazavi, 2005).

During the 1950's, Luckey indicated that low dose dietary antibiotics caused a growth surge in livestock. Later he found that hormesis could be induced effectively by low doses of ionizing radiation. In 1980 the first complete report on radiation hormesis was published (Luckey, 1980). In this report he reviewed numerous articles regarding radiation hormesis. Since the first reports, 3000 papers have been published on the benefits of low doses of ionizing radiation (for a review see Luckey, 1980, also Luckey, 1982, 1991, 1994, 1997). The concept of radiation hormesis is usually applied to physiological benefits from low LET radiation in the range of 1-50 cGy total absorbed doses (Macklis and Bresford, 1991).

Since the early experiments of Luckey, many studies on the hormetic effects of low doses of ionizing radiation in a wide variety of living organisms including plants have been performed, but due to poor experimental reproducibility and poor dose estimation, the previous studies seem to be unreliable (Yoshida *et al*., 2000). There are published reports on the induction of biopositive effects in plants after irradiation with low dose radiation. In spite of this, as far as we know, there is no any published report on the induction of biopositive effects by diagnostic doses of X-rays in plants.

Afifi *et al*. (2003) investigated the influence of ionizing radiation on the growth of apple mycoflora and the aflatoxins production. They interestingly found that doses of 0.5 kGy stimulated the production of aflatoxins by all fungi tested (*Aspergillus flavus, A. fumigatuis, A. niger, Penicillium* *expansum*) while 1.0-2.0 kGy reduced the aflatoxins production. Recently, Fan *et al*. (2004) irradiated alfalfa seeds with gamma rays to various doses from 0 to 4 kGy. Plants were sprouted and allowed to grow for up to 8 days at 23^{o} C. It was observed that sprouts grown from irradiated seeds had greater antioxidant capacity and ascorbic acid content on a fresh weight basis than those grown from non-irradiated seeds. At the same time, Al-Salhi *et al*. (2004) studied the effects of gamma-irradiation on the biophysical and morphological properties of corn plants. According to the results obtained in their study, Corn grains exposed to 1.5 and 2.5 krad showed highly significant changes in all growth parameters. On the other hand, in 2004, Zaka *et al.* in France studied the effects of short-term gamma radiation on pea plants. In this study, plants were investigated by exposing 5-day-old seedlings with doses ranging from 0 to 60 Gy. They studied plant growth and development over two generations after irradiation. Their results showed that doses higher than 6 Gy significantly inhibited the first generation plants growth and productivity.

Phaseolus vulgaris- Pinto Plant is a warm-season, annual legume. Dry pinto seed is a short-season crop. Dry pinto seed is adapted to a wide variety of soils. Most varieties maturing in a range from 85 to 110 days from emergence to harvest ripe. Dry pinto must be cultivated in well-drained soil. The main purpose of this study was to evaluate if common X-ray exposures by conventional X-ray radiography machines, are capable of producing hormetic effects in cultivated *Phaseolus vulgaris*- Pinto Plant.

Materials and Methods

Plant Cultivation : New *Phaseolus vulgaris* (Pinto) seeds yielded in 2004 were purchased. Before cultivation, the seeds were soaked in tap water for 2 days followed by another 2 days of covering under a wet cloth. Standard soil was used for cultivation. Temperature was adjusted to the optimum growing temperature that is 65 to 75 ºF. Four hundred newly cultivated seeds were randomly divided into two groups of 200 plants each (Fig. 2). In this experiment, two seeds were cultivated in each dish (100 dishes for irradiation group and 100 for sham-irradiation group).

Irradiation : Fifteen days after starting cultivation, newly grown plants were irradiated with X-rays. Plants were exposed to a single dose of X-ray (80 kVp, 80 mAs) for 6 days. On day 29, plants were pulled out from the soil. Length of plant stem, length of root, number of leaves and plant weight were measured.

Data Acquisition and Analysis : Length of plant stem, leaf length, stem diameter, length of root, number of leaves and plant weight were measured. The means of the measured parameters in X-ray irradiated plants were compared to those of sham irradiated plants using Student's *t-test*.

Results :

The growth parameters measured in irradiated and sham-irradiated plants are summarized in Table 1. The stem length in irradiated and sham-irradiated plants was 296.51±13.57 and 223.96±15.02 mm, respectively. This difference was statistically significant ($P<0.001$). The leaf length in irradiated plants showed a non-significant increase in irradiated plants compared to

Fig. 2 : Seeds were randomly divided into two groups; X-ray irradiated and sham-irradiated plants (100 dishes for each).

sham-irradiated plants (60.89±1.59 and 60.45±1.68, respectively). Although the number of leaves in irradiated plants was higher than that of sham-irradiated plants (7.05±0.18 and 6.74±0.19, respectively), the difference was not statistically significant. The stem diameter in irradiated and sham-irradiated plants were 3.52±0.12 and 3.35±0.09 mm, respectively, but the difference again was not statistically significant (P<0.001). We also observed a non-significant difference between the means of root length in irradiated and sham-irradiated plants (12.84±0.40 and 13.15±0.43 mm, respectively). Plant weight in irradiated samples was lesser than that of non-irradiated plants but it was not statistically significant (4.60±0.16 g and 4.68±0.14, respectively).

Discussion

The findings of this study indicate that diagnostic doses of X-rays can accelerate the growth of plants indicating a possible hormetic effect. The growth enhancement ratio for stem length was 1.33 that is a challenging figure. Our data are consistent with earlier studies that showed a hormetic effect in plants irradiated with radiation doses other than the diagnostic doses of X-rays. Especially, our findings are in keeping with recent studies conducted by Fan *et al*. (2004) and Al-Salhi *et al*. (2004). Fan and his colleagues observed that sprouts grown from irradiated alfalfa seeds had greater antioxidant capacity and ascorbic acid content than those grown from non-irradiated seeds. Results obtained in Al-Salhi *et al.* study showed that corn grains exposed to 1.5 and 2.5 krad demonstrated highly significant changes in all growth parameters. However, our findings are inconsistent with those obtained by Zaka *et al.* (2004) who could not show any hormetic effect in irradiated pea plants. In animals, it is usually believed that the induction of hormetic effects is due to three possible mechanisms:

1. DNA repair (Molecular level) : According to this theory, low doses of ionizing radiation induce the production of special proteins that are involved in DNA repair processes (Ikushima *et al.*, 1996). Studies using two dimensional gel electrophoresis indicated new proteins in cells irradiated with low doses of radiation. Also, it was further shown that

Table 1 : Factors showing plant growth measured in irradiated and sham-irradiated plants

Factor	Irradiated	Control	Growth	P-Value
Stem length (mm)	296.51±13.57	223.96±15.02	**Positive**	**P < 0.001**
Leaf Length (mm)	60.89±1.59	60.45±1.68	Positive	NS
No. of Leaves	7.05±0.18	6.74±0.19	Positive	NS
Stem diameter (mm)	3.52±0.12	3.35±0.09	Positive	NS
Root length (mm)	12.84±0.40	13.15±0.43	Negative	NS
Plant weight (g)	4.60±0.16	4.68±0.14	Negative	NS

- Statistically significant differences are shown in bold.

cycloheximide; a protein synthesis inhibitor blocks this hormetic effect. The function and importance of these radiation induced proteins is still unknown. Also it was found that inhibitors of poly ADP-ribose polymerase, an enzyme implicated in DNA strand break rejoining could prevent the induction of adaptive response.

2. Free radical detoxification (Molecular level) : In 1987 Feinendengen and his co-workers indicated that low doses of ionizing radiation cause a temporary inhibition in DNA synthesis (the maximum inhibition at 5 hours after irradiation). This temporary inhibition of DNA synthesis would provide a longer time for irradiated cells to recover (Feinendengen *et al.*, 1987). This inhibition also may induce the production of free radical scavengers, so irradiated cells would be more resistant to any further exposures.

3. Stimulation of immune system (Cellular level) : Despite the fact that high doses of ionizing radiation are immunosuppressive, many studies have indicated that low doses radiation may stimulate the function of the immune system. In 1909, Russ first showed that mice treated with low-level radiation were more resistant against bacterial disease (Russ, 1909). Later in 1982, Luckey published a large collection of references supporting immunostimulatory effects of low doses of ionizing radiation (Luckey, 1982).

Despite the above mentioned suggested mechanisms hormetic effects in plants may need other mechanisms to be explained. However, according to recent studies, at the molecular level, plants and animals are more similar than they are different (Goldbaum, 2005). In this regard, studies on the induced hormetic effects in plants may also contribute toward better understanding how low dose radiation can stimulate the growth, longevity or other biopositive effects in animals.

The overall results indicate that diagnostic doses of X-rays can accelerate the growth of plants. The growth enhancement ratio for stem length was 1.33 that is a challenging figure. These findings confirm the results obtained by some earlier studies that showed a hormetic effect in plants irradiated with radiations or doses other than the diagnostic doses of X-rays. However, current data seem to be insufficient and warrant further studies in order to confirm these findings and to find out the possible hormetic mechanisms.

References :

Afifi A.F., Foaad M.A. and Fawzi E.M. (2003) : Effect of gamma irradiation on elimination of aflatoxins produced by apple mycoflora in apple fruits. *Acta Microbiol Pol.* **52(4),** 379-386.

Al-Salhi M., Ghannam M.M., Al-Ayed M.S., El-Kameesy S.U. and Roshdy S. (2004) : Effect of gamma-irradiation on the biophysical and morphological properties of corn. *Nahrung.* **48(2),** 95-108.

Bruce M. (1987) : Radiation hormesis after 85 years, Health Physics Society Newsletter.

Fan X., Thayer D.W. and Sokorai K.J. (2004) : Changes in growth and antioxidant status of alfalfa sprouts during sprouting as affected by gamma irradiation of seeds. *J Food Prot.* **67(3),** 561-566.

Feinendegen L.E., Muhlensiepen H., Bond V.P. and Sonhaus C. A. (1987) : Intracellular stimulation of biochemical control mechanisms, *Health Physics*, **52,** 663-669.

Goldbaum E. Plants, animals: not so different. The internet. http://www.buffalo.edu/reporter/vol27/vol27n04/f2.html. Accessed on Feb 17, 2005.

Ikushima T., Aritomi H. and Morisita J. (1996) : Radioadaptive response; Efficient repair of radiation induced DNA damage in adapted cells, *Mutation research.* **358,** 193-198.

Luckey T.D. (1980) : Hormesis with Ionizing Radiation, CRC press, Baca Raton.

Luckey T. D. (1982) : Physiological benefits from low levels of ionizing radiation, Health Physics. **43,** 771-789.

Luckey T. D. (1991) : Radiation Hormesis, CRC Press, Boca Raton.

Luckey T. D. (1994) : Low dose radiation reduces cancer deaths, Radiation Protection Management, **11(1),** 73-79.

Luckey T. D. (1997) : A rosseta stone for ionizing radiation; radiation protection management, **14(6),** 58-64.

Macklis R. M. and Bresford B. (1991) : Radiation hormesis, *J Nucl. Med*. **32,** 350-359.

Mortazavi S.M.J. An Introduction to Radiation Hormesis. The internet. http://www.rums.ac.ir/inthorm.html. Accessed on Feb 17, 2005.

Mortazavi S.M.J., Ikushima T., Mozdarani H. and Sharafi A.A. (1999) : Radiation Hormesis and Adaptive Responses Induced by Low Doses of Ionizing Radiation, Journal of Kerman. (1999) : University of Medical Sciences, **6 (1),** 50-60.

Russ V. K. (1909) : Consensus of the effect of X-rays on bacteria, *Hygie*. **56,** 341-344.

Yoshida S., Kurisu Y., Masui H., Murata I., Yamamoto T., Iida T. and Takahashi A. (2000) : Stimulatory Effects of Low Ionizing Radiation on Plant. Proceedings of the 10th International Congress of the International Radiation Protection Association, Hiroshima, Japan.

Zaka R., Chenal C. and Misset M.T. (2004) : Effects of low doses of short-term gamma irradiation on growth and development through two generations of Pisum sativum. *Sci Total Environ.* **320(2-3),** 121-129.