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Effect of irradiation of wheat grains with fast neutrons on the grain yield and other characteristics of the plants

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HIGHLIGHTS

- Irradiation of wheat grains with neutrons (low fluence $\sim 10^2$ n/cm^2) considerably increases the grain yield throughout three subsequent generations.
- The irradiation also remarkably increases total chlorophyll concentration in the higher plant, which results in higher concentrations of sugars and crude protein.
- The irradiation also increases salt tolerance of the plants.

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ABSTRACT

The effects of fast neutrons from a ^{252}Cf source in the fluence range 10^5–10^8 n/cm^2 on the Egyptian wheat cultivar (Sakha 92) were studied. The experiment was conducted for three successive seasons (2008/2009, 2009/2010, and 2010/2011) to study the effect of the irradiation on the plant growth, grain yield, and physiological changes of three generations of plants produced by irradiated moisturized grains. A low fast-neutron fluence 2×10^6 n/cm^2 increased the yield throughout the three mutagenic generations considerably. It also increased concentrations of the total chlorophyll, sugars, and crude protein. These changes improve the quantity and quality of the grain. Also, a study of the effect of salinity of the irrigation water on the characteristics of the third-generation grains produced by neutron-irradiated grains was performed. With increasing concentration of sodium chloride in the irrigation water in the range 0.5–1.5%, concentrations of osmoprotectants, namely, reducing sugars and proline amino acids, increased. The concentration of Na^+ in the grains increased in parallel with the salinity of the irrigation water regardless of irradiation, while the concentrations of Ca^{2+} and K^+ decreased.

1. Introduction

Application of ionizing radiation in agriculture has produced new varieties of plants with improved properties. Hashem (2011) reported that irradiation of potato with fast neutrons increased important salinity tolerance of potato calluses. Olamide et al. (2012) observed an increase in the number of fruits per plant and in the number of seeds per fruit after *Capsicum Annum* and *Capsicum frutescent* were irradiated with fast neutrons. Hanafy et al. (2003) improved the rice yield by irradiating the rice seeds with fast neutron. Similar results of irradiation were observed also by Wu et al. (2005), Baloch et al. (2006), and Jia et al. (2006). Gamma and fast neutron irradiations also produced valuable mutants of tomato and chickpea plants (Nombela et al., 2003; Kharkwall, 2001, 2003). A certain dose of γ-radiation was found to increase plant resistance to unfavorable conditions, such as drought, water stress, cold stress and, in some cases, insects and diseases (Hussein and Atia, 2009). New, better wheat cultivars were obtained after irradiation of grains with fast neutrons (Duggal et al., 2000; Koebner and Hadfield, 2001; Al-Maaroof et al., 2003). Gorgidze (1980) explained how mutation of some species of cultivated wheat could take different paths after irradiation with gamma rays and fast neutrons. There may be (i) an increase in the chromosome number through autopolyploidization resulting in a polyploidy series of wheat; (ii) depolyploidization resulting in the reversion of the tetraploid species to the diploid form; (iii) aneuploidy manifesting itself in morphological changes in the diploid species; and (iv) gene mutation.

The wheat cultivar Sakha 92 was chosen for this study because it is a major food crop not only in Egypt, but also globally. Unfortunately, the demand for it far exceeds the supply. Hence, the main objective of this work was to find a dose of fast neutrons

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that would increase the grain yield. Another goal was to study the effect of various neutron fluencies (10^5–10^8 n/cm^2) on morphological and physiological parameters of the wheat plants through three successive generations (M1, M2 and M3) as well as on the salt tolerance of the plants. Increasing the yield of wheat is undoubtedly of utmost importance.

2. Materials and methods

The analytical work was carried out in the Physiology Laboratory of the Botany Department of the Faculty of Science of Zagazig University, Egypt. The experimental work in the field was conducted in the period from 2008 to 2011.

2.1. Plant material

Wheat grains (*Triticum asetivum* L.) (Sakha, 92) were obtained from the Agriculture Research Center in Cairo, Egypt. The grains were divided into five groups (A, B, C, D, and E), each of which contained 100 grains. All the groups were soaked for 24 h. Group A was kept unirradiated (control), while the other groups were irradiated with various neutron fluencies.

2.2. Irradiation

The irradiations were performed at the Biophysics Department of the Faculty of Science of Cairo University, Egypt. A ^{252}Cf point source (E=2.8 MeV) manufactured by Radiochemical Center (Amersham, UK) was used. Mature soaked wheat grains of Groups B, C, D, and E were irradiated with 4×10^5, 2.0×10^6, 1.0×10^7, and 2×10^8 n/cm^2 of the fission neutrons, respectively. After the irradiation, the grains were planted to produce generations M1, M2, and M3.

2.3. Plantation of wheat grains

The wheat grains were planted in the wheat green house at the Faculty of Agriculture of Zagazig University and regularly watered until complete germination. Grains of the 2008–2009 harvest (M1) were used to produce the second generation grains M2 in the 2009–2010 harvest, and the latter were used to produce the third generation M3 in the harvest of 2010–2011.

Grains irradiated with the fluence that provided the best results (highest yield and good results of the analyses throughout M3) were selected for deeper evaluation (salinity test and spectroscopy of crude protein).

2.4. Morphological and physiological measurements

Extraction and quantification of the photosynthetic pigment (chlorophyll $a+b$) from seedlings were performed as described by Metzner et al. (1965). For each of the three harvests, morphological parameters of the plants were evaluated, which included plant height, number of leaves per plant, root length, weight of dry plant, number of spikes per plant, number of grains per spike, weight of 100 grains, and change in the grain yield. Extraction and analysis of carbohydrates were conducted using the methods proposed by Naguib (1963, 1964). Directly reducing sugars were quantified according to Nelson (1944).

Concentrations of Na^+, K^+, Ca^{2+}, Mg^{2+},Fe^{3+} and Zn^{2+} were determined by the method of Jackson (1967). The total nitrogen was quantified using the automated Macro–Kjeldahl method (Tecator Digestion System, FOSS, Hillerød, Denmark) according to Haynes (1980). Crude protein concentrations were determined by multiplying the total nitrogen concentrations by a factor of 6.25 (A.O.A.C, 1970). The proline content was estimated according to Betes et al. (1973).

2.5. Salinity effect studies

M3 wheat grains (unirradiated and irradiated with 2.0×10^6 n/cm^2) were put on moist cotton pieces in plastic dishes to reach the germination state. Three-day-old seeds were irrigated with a half-strength nutrient solution (Hogland+water, 1:1). One week later, the seeds were transplanted into pots and placed into the green house.

In the winter of 2009–2010, pots 50 cm in diameter were filled with a mixture of clay and fine sand (2:1). The irrigation regimen described by El-Sherbieny et al. (1986) was used, where pots were irrigated until saturation and then completely drained. The modified Hogland solution suggested by Johnson et al. (1957) was employed.

Salinity treatments started one week after the transplantation, when 0.5%, 1.0%, and 1.5% NaCl (5×10^3, 1.0×10^4, and 1.5×10^4 ppm, respectively) was added to the nutrient solutions used for the irrigation. All pots were washed with water after 5 irrigations. Salt concentrations were maintained throughout the life cycle of the plants by irrigation of the pots with the appropriate solutions three times a week.

Random samples of plants were collected at the vegetative stage for chlorophyll quantitation, and, at harvest, grains were collected for quantification of proline, carbohydrates, Na^+, Ca^{2+}, and K^+. The results of the analyses were processed statistically at the 0.05 probability level as described in the book by Snedecor and Cochran (1972).

3. Results

3.1. Morphological characteristics

Fig. 1 shows the effects of fast neutron irradiation on the grain yield and plant growth characteristics. Plant height, number of leaves per plant, root length, average weight of dry plant, number of spikes per plant, number of grains per spike, weight of 100 grains, and grain yield, all gained from the irradiation with 2.0×10^6 n/cm^2.

3.2. Chemical analysis

3.2.1. Chlorophyll concentration

The concentrations of chlorophyll in generations M1, M2, and M3 of the plants grown from the grains irradiated with 2×10^6 n/cm^2 are higher than of the plants grown from grains irradiated with the other neutron fluences (Fig. 2).

3.2.2. Concentrations of other plant components

Plants from grains irradiated with 2.0×10^6 n/cm^2 had the highest concentrations of monosaccharides, disaccharides, polysaccharides, and total sugar (Fig. 3). As mentioned above, this neutron fluence also provided the highest grain yield. By contrast, the irradiation with the highest neutron fluence (2×10^8 n/cm^2) resulted in the lowest concentrations of monosaccharides, disaccharides, and the total sugar.

Fig. 4 shows that the same neutron fluence 2.0×10^6 n/cm^2 resulted in the highest Na^+, Mg^{2+}, Fe^{3+}, and K^+ concentrations in the M1, M2, and M3 grains. In most cases, the highest fluence 2×10^8 n/cm^2 produced somewhat lower metal concentrations, although the concentration of Ca^{2+} was an exception.

As follows from Fig. 5, the grains irradiated with 2.0×10^6 n/cm^2 also produced grains with the highest levels of crude protein.