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Research Article

MUTAGENIC EFFECT OF ULTRAVIOLET IRRADIATION (UV-B) ON THE QUALITY TRAITS OF WHEAT GRAINS

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Abstract

The hydration step is the key stage in the bread wheat manufacturing process. This study aimed to investigate the mutagenic effect of ultraviolet irradiation on improving grain quality traits in wheat. Wheat grains were subjected to nine doses of UV-B irradiation, and non-irradiated grains served as control. This study was conducted in a randomized complete block design to test the genetic diversity induced in some quality traits in wheat grains. Based on the experimental results pre-soaked irradiated grains stimulated hydration coefficient, seedling emergence, and soluble carbohydrate contents than in non-soakers irradiated. Correlation coefficient obtained between the doses of UV irradiation with hydration coefficient and with seedling emergence was higher in pre-soaked than in non-soakers irradiated grains. Meanwhile, correlation coefficient between the doses and carbohydrate contents was positive in pre-soaked irradiated grains and negative in non-soaked irradiated. The estimates of the phenotypic coefficient of variation were relatively greater than the genotypic coefficient of variation for hydration coefficient and seedling emergence. In contrast, the differences between these estimates were relatively low for carbohydrate contents. This indicates the presence of additive gene action in controlling carbohydrate contents and the selection procedure will be effective for improvement across genetic diversity in the population. This study may be valuable for addressing the genes expressed associated with water hydration before irradiation which exhibited positive results. This will be useful for wheat flour improvement programs. The results highlighted the importance of pre-soaked irradiated grains for induced optimum gene expression to improve grain quality contributing traits.

Keywords: Ultraviolet irradiation, Correlation coefficient, Carbohydrate contents, Genetic variability parameters, Grains viability, Hydration coefficient, heritability, Genetic advance.

INTRODUCTION

Wheat, *Triticumaestivum* L., is one of the most cultivated cereal crops worldwide. The second crop is corn and the third is rice (Shewry 2009). Its total cultivated area is 241.29 million hectares with worldwide production of 734.05 million tons. Egypt was involved in these statistics in 2018. Egypt produced annually 8.8 million tons from the cultivated area reached to 1.32 million hectares. Furthermore, Egypt is among the top wheat importers with about 10.16 million tons annually (FAOSTAT, https://www.fao.org/statistics/en). Approximately seven billion people worldwide depend on bread wheat in their daily foods. Wheat (2n =6x = 42 chromosomes, AABBDD) is the main source of carbohydrates, proteins, and fibers (Ola-Olorun *et al*., 2021). Reports indicated that wheat production has declined in the last few decades with 5.5% due to continued climate changes leading to drought and heat stresses (Daryanto *et al.,* 2016). It is interesting to note that a 70% increase in wheat production and consumption will be required by the year 2060 to suffice human requirements (Ortiz *et al.,* 2008). Mutant populations selected in the early generation are important to advance desirable traits in wheat (Ola-Olorun *et al.,* 2020). EMS treatment was suitable to induce mutations in wheat, as well as, to select ide types with improved drought tolerance, high yield, and high root-to-shoot ratios (Ola-Olorun *et al.,* 2021). Wheat is the main source of carbohydrates for the majority of Egyptian people. Therefore, it is considered the most important strategic cereal crop in Egypt. Many foods were manufactured from wheat flour all over the world. The characteristics of wheat grains play a significant role in deciding the suitability of wheat flour for the industrial production of wheat products (Kumar and Pavuluri 2022).

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In wheat flour gluten protein networks are the main component decides the quality of flour. Gluten is a macromolecule protein network formed by the cross-linking of gluten in and gliadin protein subunits in the presence of water during the kneading process (Ferrero 2017). Wheat nowadays is the second source of carbohydrates after rice. The need for wheat in the food sector in Egypt is increasing year by year (Dwinanda *et al.,* 2020). Wheat grains are usually irradiated by ultraviolet rays for induced mutations because they can be grown. Mutations are very effective in changing a few properties in improving plant varieties. Mutation induction was able to speed up the harvest time of some local cultivars of wheat and rice (Suliansyah 2010). Genetic variability could be created through hybridization or mutations followed by selection (Addisu and Shumet 2015). Improving through hybridization is time-consuming with low genetic variation (Hanafiah *et al.,* 2010). The powerful strategy technique used for the direct produced improved genotypes in wheat is mutation (Dhaliwal *et al.,* 2015). Up to date, 3288 mutant varieties in different crops are generated by mutation breeding. These are registered in the database of the International Atomic Energy Agency (FAO/ IAEA, http://mvgs.iaea.org). Most of these mutant genotypes are generated in cereals, oil crops, and legumes. 237 mutant genotypes were generated in bread wheat. Out of these, 195 mutant varieties were released from physical mutagens, 40 mutant varieties were released from chemical mutagens and two were generated from their combinations (Suprasanna *et al.,* 2015). Physical mutagens such as non-ionizing radiation (e.g. UV) or ionizing radiation (e.g. X, gamma rays, alpha and beta rays) caused DNA double-strand breaks and produced DNA large deletion. Thereby these mutagens caused visual effects on chromosome structure and gene expression. It is expected to be obtained one mutation per 6.2 Mb (Sato *et al.,* 2006). The longer dose of mutagens damages the genotype of plant cells. In contrast, the lower doses may cause bio-stimulation effects (Chen *et al.,* 2005). Stimulation performance is due to synergism between plant photoreceptors and polarized monochromatic laser beams (Koper *et al.,* 1996). Mutations generated allelic versions in gene structure and function causing phenotypic variations (Dhaliwal *et al.,* 2015). Chemical and physical mutagens were used before to produce new genotypes in bread wheat (Rakszegi *et al.,* 2010). Mutant lines displayed wide genetic diversity in morphological and physiological traits (Xiong *et al.,* 2018). During the last 50 years, the ozone layer has been depleted. As a consequence, increasing levels of UV irradiation (wavelengths from 280 to 320 nm referred to as UV-B) reached the earth's atmosphere. The reduction in the stratospheric ozone layer resulted from anthropogenic pollutants such as halogenated hydrocarbons (Molina and Rowlands 1974). The depletion of the ozone layer leads to a significant increase in UV-B reaching the Earth's surface (Blumthaler and Amback 1990). In response to UV-B, there were biochemical and physiological changes in plants (Kramer *et al.,* 1991). Elucidation on UV-B stress would help in understanding the impact of partial ozone depletion on plant adaptation to changing environments. Huovinen *et al.* (2006) expected that increased UV-B radiation will affect on phenological phases of wheat with different magnitudes. Hidema *et al.* (2005) found that increased UV-B radiation modified grain protein concentration in rice.

Carbohydrates are biomolecules consisting of carbon (C), hydrogen (H), and oxygen (O) atoms taken from the empirical formula Cm (H_2O)n. The H has covalent bonds with C as CH_2O, but not with O. Carbohydrates include sugars, starch, and cellulose. The sugars are divided into four main groups: monosaccharides, disaccharides, oligosaccharides, and polysaccharides. The smallest molecular weightis monosaccharides and disaccharides. Polysaccharides serve as energy stores like starch and glycogen and as structural components such as cellulose in plants and chitin in fungi and arthropods. The important component in the genetic molecule RNA is 5-carbon monosaccharide ribose. The related deoxyribose is an important component of DNA molecules. Saccharides and their derivatives play major roles in the fertilization immune system and preventing pathogenesis, blood clotting and development (Maton *et al.,* 1993). Carbohydrates are a central component of nutrition. The polysaccharide carbohydrate abundant in cereals is starch (such as wheat, maize, and rice), as well as, potatoes. The main component in plant cell walls is the polysaccharide cellulose. It is not digestible by humans (USDA 2015). Monosaccharides included glucose, fructose, and glyceraldehydes. It can be linked together to develop polysaccharides. Many carbohydrates contain one or more modified monosaccharide units, as deoxyribose a component of DNA. This is a modified version of ribose (Matthews *et al.,* 1999). Carbohydrates are classified into three principal groups, namely sugars, oligosaccharides, and polysaccharides according to their degree of polymerization (Pichon *et al.,* 2006). The joined monosaccharides are called disaccharides as sucrose and lactose. Oligosaccharides are polymers of saccharide composed of three to ten monosaccharide units connected with glycosidic linkages as in disaccharides. They are usually linked with lipids or amino acids. They have roles in cell recognition and cell adhesion (Avenas 2012). Carbohydrate in foods yields 3.87 kilocalories of energy per gram of simple sugars and 3.57 to 4.12 kilocalories per gram of complex carbohydrates in most other foods (Pichon *et al.,* 2006). Starches were break down easily into glucose by many organisms. Most cannot metabolize cellulose or other polysaccharides as chitin. These carbohydrate types can be metabolized by some bacteria (Pichon *et al.,* 2006). Wheat is a prime source of energy and staple food worldwide for the human population. It is a major component of most diets associated with human food. Wheat products are mainly consumed as a carbohydrate source. Cereal grains contribute 50% of the world's dietary calories. Wheat contributes less than 20% of these total calories (Pomeranz 1988). There are three major steps in the pre-cooking process i.e. soaking, steaming, and drying. They have a great influence on the final quality of the end product. Since water is absorbed at different rates by wheat grains, it is interesting to characterize the absorption process of the material. Soaking is an important tool that facilitates the grains for further processing. Hence there was a need to study the hydration characteristics of irradiated wheat grains with UV-B, for the development of value-added products from wheat sterilized with UV-B before storage in silos, to protect the grains from infections with microorganisms (Becker 1960). Therefore, this study aimed to evaluate the mutagenic effects of UV-B irradiation on wheat grains contributing quality traits, and their genetic variability parameters associated.

MATERIALS AND METHODS

Genetic material

Grains of wheat (*Triticumaestivum* L.) variety Sakha 171 were kindly obtained from the Wheat Research Section, Field Crops Research Institute, Egyptian Agriculture Research Center, Egypt. This cultivar was selected based on it is used as a commercial cultivar.

Reagents

Anthrone reagent

It was prepared by dissolving 200 mg anthrone in 100 ml concentrated H_2SO4.

Ultraviolet radiation source

A source of ultraviolet irradiation (UV-B) is a UV lamp in the laminar air cabinet located in the Laboratory of Microbial Genetics, Faculty of Agriculture, Mansoura University, Egypt. This is an artificial source of UV irradiation according to Ehrenberg (1961). This lamp with a characteristic emission in the range of 300 nm (125 W, Phillips, The Netherlands). Therefore, it was classified as UV-B. The distance between wheat grains and the UV lamp was 25 cm. One minute of exposure time to UV-irradiation was equal to 188.2 joules/m^2 according to Kondrateva *et al.* (2020). The joules are defined as the amount of energy extracted when a force of one newton is applied over a displacement of one meter which is equivalent to one watt of power radiated for one second (Kondrateva *et al.,* 2020).

UV-B irradiation test

The UV-B treatments tested were characterized by two different groups of soaked and non-soaker grains exposed to UV irradiation, conventionally referred to as UV-B-T_1 and UV-B-T_2. Tests were performed on two main groups of wheat grains. Each group contains eleven Petri dishes each of them containing one hundred wheat grains. The wheat grains in the first group (T_1) were weighed and then soaked in tap water for

12 hours. The soaked grains were transferred from the water to filter paper to dry for five hours and then weighed. Soaked grains were treated separately with UV-B rays at the eleven doses; 0, 10, 20, 30, 40, 50, 60, 70, 80, 90, and 100 minutes. The second group (T_2) of wheat grains were separately treated directly as dried grains with UV-B rays. A preliminary experiment of UV-B irradiation sensitivity test was conducted using Petri dishes each containing 100 dry grains subjected to ten doses, 0, 10, 20, 30, 40, 50, 60, 70, 80, and 90 minutes. The grains were then weighted followed by soaking in tap water for 12 hours. The soaked grains were transferred from the water into filter paper to dry for five hours and then weighed. The wheat grains in both groups were re-soaked enough for 24 hours in tap water to be subjected to a seedling emergence test according to Rakszegi *et al.* (2010). The treatments were arranged with three replicates. The wheat grains were weighted in dry and soaked phases with an electronic balance. Average grain weight was estimated using three random subsets (100 grains each) per Petri dish according to Calderini *et al.* (2008).

Seedling emergence

One hundred wheat grains were steeped in tap water for 12 h at laboratory temperature and then completely drained of steep water using sieves. The drained grains were then spread on a moistened cotton and allowed to germinate at laboratory temperature for seven days at 25 ± 3. The effect of UV-B treatments on seedling emergence was expressed as the reduction or stimulation in germination of irradiated grains about the germination of unirradiated control according to Romano *et al.* (2024) as follows:

Viability = Treatment / Control

Standard glucose solution

The stock standard was prepared by weighing 500 mg of glucose and transferring it carefully into 500 ml distilled water located in a one-liter conical flask and then sterer the solution carefully. Take 00, 5, 10, 15, 20, 25, 30, 40, 45, 50, 55, and 60 ml from the standard solution separately to each of 13 volumetric flasks 250 ml. Then complete the volume in each flask to 100 ml with distilled water. Add 4 ml of anthrone reagent to each flask. Well mixing carefully the contents in each flask. Cover each flask well with aluminum foil. A boiling water bath was used for heating the contents in all flasks for eight minutes. Cool rapidly all the flasks to room temperature. Read the optical density of each solution spectrophotometrically at 630 nm. Simultaneously, the blank was prepared with four ml of anthrone reagent and one ml of distilled water. Construct the calibration curve using a regression line by plotting glucose concentrations on the X-axis and the absorbance at 630 nm on the Y-axis. Calculate the sugar concentration in the sample from the calibration curve using the regression formula as follows:

$$x = \frac{Y - a}{b}$$

Where, x = unknown concentration, Y = absorbance of unknown sample, a = y-axis intercept, b = slope value which can be obtained from the standard curve equation.

The concentration of unknown carbohydrate samples can be calculated from the standard curve equation. The result should be expressed as mg/ml, depending on the initial sample volume and dilution factor according to Sadasivam and Manickam (1996).

Assessment of total carbohydrates

Carbohydrates are dehydrated with concentrated H_2SO4 to develop furfural. The origin of furfural is the dehydration of monosaccharides. Furfural condenses with anthrone to develop a green colour complex which can be measured spectrophotometrically at 630 nm. Anthrone reacts with polysaccharides, disaccharides, and monosaccharides. The reaction of furfural with anthrone reagent produced a blue-green color complex. The sources of color are carbohydrates. Wheat grains were carefully cleaned and then five grams of each irradiated grains. Weight 100 mg of wheat flour and then place it into a boiling tube. Add five ml of 2.5 N-HCl to each sample in a boiling tube. Keeping the samples in a boiling water bath for three hours to hydrolyze the samples. Cooling the samples at room temperature. Add Sodium carbonate to neutralize the solution until the effervescence. Adjust the solution volume to 100 ml and centrifuge. Add 0.5 ml distilled water to 0.5 ml of the supernatant collected to reach the volume of one ml in all the tubes including the sample tube to be analyzed. Cool on ice the contents in all the tubes before adding anthrone. Then, add four ml of ice-cold anthrone reagent. Heat all the tubes for eight minutes in a boiling water bath. Cool the tubes rapidly on ice and then read the color ranging from green to dark spectrophotometrically at 630 nm. Glucose concentrations were calculated from the glucose standard curve that was prepared before. The amount of carbohydrates present in 100 mg of wheat flour was calculated according to Hedge and Hofreiter (1962) using the following formula as follows:

Amount of carbohydrates in 100 mg samples =
Glucose concentration obtained from the standard curve x Volume of test sample (0.1g)

Hydration coefficient

For each irradiation treatment, 300 healthy grains were selected at random. The grains were carefully cleaned and freed from dirt, stones, chips, and other extraneous grains or dirt. From each treatment of irradiated grains, 100 grains were counted randomly in triplicated and their dry weight was recorded. The grains were stored in a clean airtight beaker and then soaked in tap water at a ratio of one grain part to four parts of water for 24 hours. The hydration coefficient was calculated according to Abdelgani *et al.* (1999) as follows:

$$\text{Hydration coefficient} = \frac{\text{Weight of soaked grains}}{\text{Initial weight}}$$

Statistical analyses

Each irradiation sample was analyzed in triplicates. The significance of differences between treatments was determined by the analysis of variance (ANOVA) followed by LSD according to Steel and Torie (1960). Regression analyses were used to assess the degree of association between variables. The Figures of regression analysis were then averaged according to Calderini *et al.* (2008). Genotypic and phenotypic variance were assessed according to Johnson *et al.* (1955). Phenotypic and genotypic coefficient of variation were assessment according to Burton and Devane (1953). Heritability in a broad sense was estimated according to Hanson *et al.* (1956).

RESULTS AND DISCUSSION

Hydration coefficient

As shown from the results presented in Table 1, there were significant differences in hydration coefficient between soaked irradiated grains. In contrast, UV treatments had insignificant differences in hydration coefficient between non-soakers irradiated wheat grains. This indicated that the hydration coefficient was significantly affected by soaker grains before irradiation treatment. This is in line with the findings of Elsheikh and Elzidany (1997), who found that the hydration coefficient of the faba bean was significantly influenced by organic, chemical, and biological fertilization. The factors that make wheat grains hard to cook cause serious problems. A low hydration coefficient as shown in non-soaker grains before UV-irradiation indicates that the grains are not capable of absorbing water very efficiently.

particles during processing. The high hardness of wheat grains may affect flour quality and bread bread-making industry by increasing water absorption. This leads to viscosity changes in bread dough since damaged starch can retain more water four times than native starch (Kweon *et al.,* 2011). Therefore, hardness grains contained the highest damaged starch and absorbed more water capacity than non-soaker grains before irradiation which contained high native starch. The highly damaged starch in soaked grains was produced by alpha-amylase activity in hydration grains. Thus, the flours containing highly damaged starch are not only technologically important due to retained water content, but also have speed of water absorption as the phenomena involved in cookie production (Kweon *et al.,* 2011). In this respect, Huovinen *et al.* (2006) expected that increases in UV-B irradiation doses will influence on phenological phases of wheat with different magnitudes.

Table 1. Hydration coefficient of irradiated wheat grains

Doses of UV (minutes)		Non-soakers grains before UV irradiation		Soaked grains before UV irradiation		Mean HC	Hydration yield
		HC	HY	HC	HY		
0		0.98	1.00	0.78	1.00	0.88	1.00
10		0.78	0.80	0.94	1.21	0.86	0.98
20		0.86	0.88	0.99	1.27	0.93	1.06
30		0.92	0.94	0.94	1.21	0.93	1.06
40		0.87	0.89	0.91	1.17	0.89	1.01
50		0.95	0.97	0.96	1.23	0.95	1.08
60		0.90	0.92	0.91	1.17	0.90	1.02
70		0.91	0.93	0.97	1.24	0.94	1.07
80		0.85	0.87	0.94	1.21	0.90	1.02
90		0.93	0.95	0.97	1.24	0.95	1.08
Mean of irradiated		0.89	0.91	0.95	1.22	0.92	1.04
F-Test		NS	NS	NS	*	NS	NS
LSD	0.05	0.211	0.215	0.137	0.141	0.135	0.148
	0.01	0.290	0.294	0.187	0.192	0.185	0.203

HC: Hydration coefficient. HY: Hydration yield.
NS, *: Not significant and significance at 0.05 probability level, respectively.

Therefore, the hydration coefficient is a very valuable quality factor for wheat night consumers. The results reflected that soaked wheat grains before exposure to UV irradiation led to an increased hydration coefficient. Thus, all UV treatments significantly increased the hydration coefficient over the control in soaked irradiated grains. So, water absorption is a key value for functional characteristics of wheat flour especially that used for the production of bread wheat and cookies. Water plays a major role in the interaction between dough ingredients and contributes to the structure of dough (Blanco *et al.,* 2019). Therefore, soaked irradiated grains increased water absorption which is a suitable maker for the production of cookies. In contrast, wheat flours have a low water absorption as seen in non-soaker grains which are suitable for the production of short-dough cookies (Blanco *et al.,* 2019). As a result, the technological properties during the production of wheat products and changes in quality parameters of grains as increased hydration coefficient can lead to increased acceptability by consumers. Thereby increasing the consumption of whole grains. Soaked irradiated grains present the highest damage of starch when exposed to UV irradiation (Angelidis *et al.,* 2016). Some studies have shown that wheat genotypes influence the damaged starch content leading to grain hardness (Duyvejonck *et al.,* 2012). This finding agrees with Yu *et al.* (2015), who found that wheat grains have higher grain hardness and contain elevated values of damaged starch under the same milling conditions. Higher-hardness wheat grains contain the highest damaged starch require more energy to break down and increase flour

Thus, spring wheat cultivars would exposed to high levels of UV-B irradiation since biomass was accumulated at a high rate during the reproductive stages between November and January (Ugarte *et al.,* 2007). So, grain quality is an important objective in the systems of wheat production. In the last few decades, Hidema *et al.* (2005) found that increased UV-B irradiation modified the concentration of grain protein in rice. In addition, Calderini *et al.* (2008) found that wet gluten values in wheat grains tended to be greater under increased the doses of UV-B radiation. Meanwhile, Kakani *et al.* (2003) found that UV-B irradiation has little influence on crop phenology. Li *et al.* (2000) observed that seven out of 20 wheat genotypes did not appear to significantly decrease in biomass yield when UV-B exposure was increased by 5 KJ/m^2. However, Alexieva *et al.* (2001) hypothesized that grain quality traits associated with proteins may be influenced by UV-B radiation during the grain-filling period. This hypothesis was supported later by Hidema *et al.* (2005), who obtained higher protein concentrations in rice grains in response to increased doses of UV-B irradiation. In this study, the quality of wheat grains was assessed by evaluating the hydration coefficient of grains, which is commonly accepted as the main trait associated with the bread dough quality of this crop. Soaked grains before UV irradiation leading irradiation likely increased the damaged starch in wheat grains, because of alpha-amylase activity. As a consequence hydration coefficient was increased in soaked irradiated grains than in non-soakers. Therefore, damaged starch was associated with soaked irradiated grains leading to be increased hydration coefficient. It has been well documented that the hydration step in bread bread-making industry is the critical factor in many bread-manufacturing products from cereals (Boucheham *et al.,* 2019). The solid-water interactions in hydration process lead to produce particles binding to form an agglomerated dough (Hebrard *et al.,* 2003). The components that contribute to the interactions with water are starch, proteins, and ash. These are characterized in wheat because of higher contents in starch (61.43% - 87.97%) than in legumes (32.86% - 39.09%) (Koehler and Wieser 2013). The wheat flour capacity of water indicates how bioactive compounds interact with water. The pentosans can absorb liquid water 15 times their weight as a very hydrophilic components. Meanwhile, gluten absorbs water 2.15 times its weight. Although, the native starch absorbed water 0.44 times its weight. The damaged starch absorbed water two times its weight (Feillet 2000). The hydrophilic capacity of gluten protein would be expected to have a major effect on the rheological properties of bread dough (Sidhu *et al.,* 1980). For instance, Graveland *et al.* (1979) observed that carbohydrates are covalently bound with gluten protein.

Regression analysis of the hydration coefficient

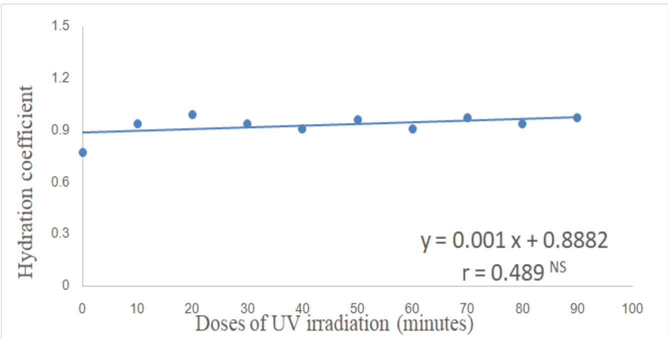

Figure 1. Regression analyses of hydration coefficient in pre-soaked irradiated wheat grains

As shown from the results diagrammatic in Figure 1, the regression coefficient of 0.001 means that the hydration coefficient of soaked wheat grains before UV irradiation increases by 0.001 with each additional minute of exposure to UV. The time of exposure to UV was chosen to express the units of independent variable in this model. Regression analysis can be used in this study for predicting the outcomes in a dependent variable named hydration coefficient based on the relationship between hydration coefficient and UV doses. This analysis denotes the estimated increase in hydration coefficient for every unit increase in the UV exposure time. The correlation coefficient between the hydration coefficient and UV doses was positive (r = 0.489). The coefficient of determination (r^2) in this relationship was equal to 0.239. This means that 23.9% of the variance in hydration coefficient is due to UV irradiation. The remaining 76.1% might be explained by other factors that were not taken into account in the analysis, such as wheat genotype, hydration time, water temperature, quantity of water, etc. These results agree with Schneider *et al.* (2010), who reported that if the coefficient of determination between human height and weight is 0.785, then 78.5% of the variance in human weight is due to height. The remaining 21.5% is due to other factors that were not taken into account in the analysis, such as eating, exercise, sex, or age.

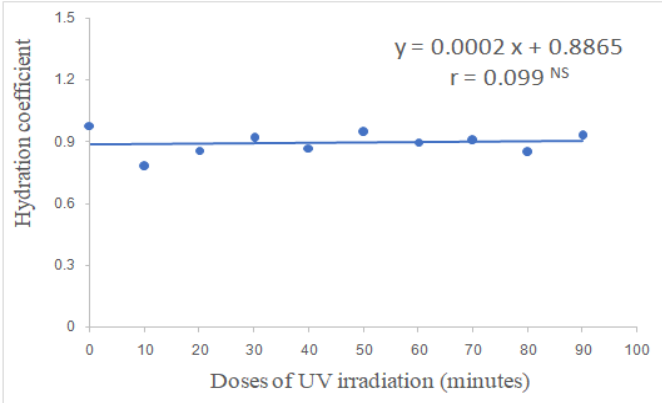

Figure 2. Regression analyses of hydration coefficient in non-soakers irradiated wheat grains

As shown from the results presented in Figure 2, the regression coefficient of 0.0002 means that the hydration coefficient of non-soaked grains irradiated with UV increases by 0.0002 with each additional minute of UV exposure time. However, hydration coefficient and irradiation time vary in such a way that movements in irradiation time are accompanied by movements in hydration coefficient, then these variables are correlated. The prediction in hydration coefficient based on correlation analysis will be more reliable and near to reality. The correlation between both variables is positive because these variables vary in the same direction. It means that if irradiation time increase then the hydration coefficient is also increasing by 0.0002. The correlation between both variables was equal to 0.099. Then the coefficient of determination (r^2) = 0.01. This means that a change in exposure time to UV irradiation doesn't influence well on hydration coefficient of non-soakers irradiated grains. The coefficient of termination is the ratio between the explained variance to the total variance (Moore *et al.,* 2013). The regression coefficient increased five times in irradiated soaked grains than in irradiated non-soaker grains. This indicated that hydration was increased in soaked irradiated grains than in non-soakers irradiated grains. This leading correlation coefficient was increased more than five times in soaked irradiated gains than in non-socked irradiated. This is in line with the findings of Guardianelli *et al.* (2019), who established that during hydration the main storage biopolymers as carbohydrates, lipids, and proteins are hydrolyzed to lower molecular weight compounds, due to the activity of hydrolytic enzymes, which are inactive in non-soaked grains. In addition, Ballaré *et al.* (2011) observed the positive effects of UV-B irradiation as stimulation of secondary metabolism. The magnitude of hydration correlates well with soaked irradiated than in non-soaked irradiated grains, which is of interest in this study. This reflected the stimulating effect of UV irradiation on the hydration coefficient of wheat grains. It was experimentally established that the highest values of regression and correlation were obtained in soaked grains irradiated with UV. This agrees with Semenov *et al.* (2020), who achieved an improvement in seed vigor and germination in wheat indices under applying 400 – 500 Jm^{-2} doses of UV-C. The same authors also found a significant effect of UV irradiation on the development of root systems and aboveground parts in wheat grains treated with 250 Jm^{-2} and 500 Jm^{-2} UV-C doses. The effectiveness of UV-B on soaked grains could be attributed to the physiological and biochemical states of soaked grains before irradiation. This contributed to an increase in hydration coefficient as compared to non-soakers before irradiation.

The obtained results prove the expediency of using soaked grains before UV irradiation to increase mutations, seed vigor, and germination, where sometimes there is a threat of losing some of the breeding material with low seed vigor and germination. Water plays a complex role in wheat grains since it affects the nature of bread dough, modifying its rheological behavior. So, hydration is one of the most significant functional characteristics of wheat flour (Ren *et al.,* 2008). Based on the obtained results, it has been established that, although UV-B radiation is a useful technique in plant breeding programs, it is extremely important to increase the hydration coefficient in wheat grains and bread dough to obtain high-quality grains and flour from seeds. Further, UV exposure to grains accelerates the synthesis of functional agents by triggering phenolic metabolism in plant cells (Dubrov 1963).

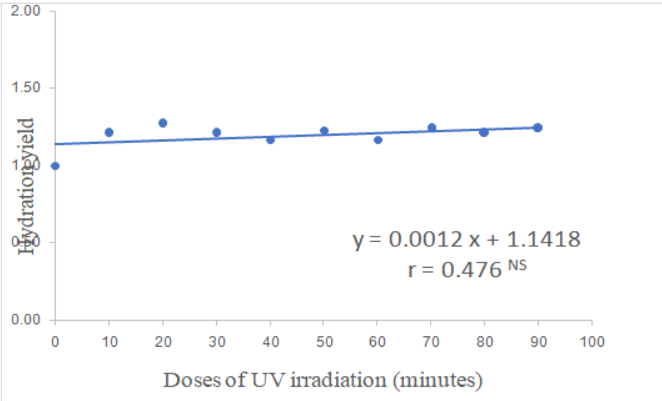

Figure 3. Regression analyses of hydration yield by pre-soaked irradiated wheat grains

According to Figure 3, the regression coefficient of 0.0012 means that hydration yield about unirradiated grains increases by 0.0012 in irradiated soaked grains with each additional minute of UV-B exposure time. The independent variable in this Figure is UV-B doses considered as input, driver, or factor that has an impact on a dependent variable which is called an outcome. Regression analysis can be useful for predicting the outcomes of hydration yield based on the relationship between dependent and independent variables. This analysis allows geneticists to investigate the relationship between UV doses and hydration yield in soaked irradiated wheat grains. These variables are usually labeled as dependent or independent. The direction of the strength relationship indicates the nature of the relationship among variables. This may be positive if the increase in one variable as UV doses results in an increase in the other as hydration yield. The relationship may be negative if an increase in the independent variable results in a decrease in the dependent variable. Therefore, regression analysis is a powerful technique with many implications in genetic research. It enables geneticists to describe, predict, and estimate the relationship between the interrelated variables of any phenomena studied in genetics (Montgomery 2012). The results appeared correlation coefficient (r) = 0.476. This is a medium correlation. Squaring this number results in the coefficient of determination (r^2) = 0.2266. This means that approximately 22.66% of the variance in the response variable, hydration yield, can be explained by the UV-B doses. The rest variance of 77.34% remains unexplained and is attributed to other factors that are not present in the regression formula as wheat genotype, water temperature, chemical composition of wheat grains, etc. These results are in harmony with Moore *et al.* (2013), who considered correlations above 0.7 to be strong because if r = 0.7, then r^2 = 0.49. This indicated that approximately 50% of the variance in the dependent variable can be explained by the independent variable.

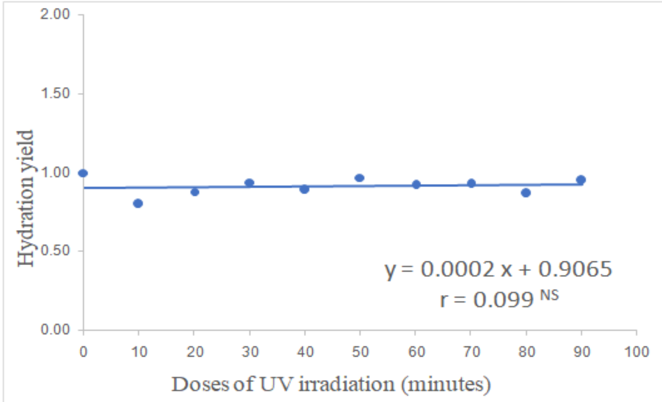

Figure 4. Regression analyses of hydration yield by non-soakers irradiated wheat grains

Regarding Figure 4, the regression coefficient of 0.0002 means that hydration yield in non-soakers irradiated grains increases by 0.0002 over unirradiated grains with each additional minute of UV-B exposure time. The correlation coefficient between hydration yield and UV doses was equal to 0.099. This is a fairly small correlation. Squaring this number results coefficient of determination which is denoted by r^2 = 0.0098. This means that 0.98% of the variance in hydration yield can be explained by the independent variable UV-B exposure time. This indicated that UV-B doses do not influence well on hydration yield of non-soakers irradiated wheat grains. Therefore, the correlation coefficient helps to identify the changes in hydration yield that are explained or attributed to the independent variable doses of UV-B irradiation. This agrees with the work of Schneider *et al.* (2010), who established that the determination coefficient (r^2) for assocciation between human height and weight is 0.785. This means that 78.5% of the variance in human weight is due to height. So, correlation analysis is a statistical method used to estimate the relationship between two or more variables. This technique was used to assess the degree and direction of association between UV-B doses and hydration yield in wheat grains. This relationship between these two variables was positive because both variables vary in the same direction. Irradiation dose orientation used in this study is conducted to obtain an effective hydration yield due to generating genetic diversity.

The effective hydration yield was obtained from soaker-irradiated grains because of the high frequency of mutations with little physical damage by UV irradiation. The effect of UV on non-soakers irradiated grains did not improve hydration as obtained by soakers irradiated grains. This agrees with Esnault *et al.* (2010), who established that ionizing radiation exposure activates biological systems with several chemical and physical reactions. This results in the formation of ionized water molecules and free radicals (Esnault *et al.,* 2010). The free radicals can damage cell organelles or modify the chemical components of plant cells. These lead to an effect on chemical and biological processes that may be vital to the viability of the organism (Marcu *et al.,* 2013). Thus, soaked grains before exposure to UV-B irradiation are a very valuable factor in producing high genetic variability.

Table 2. Seedling emergence from 100-irradiated wheat grains

Doses of UV (minutes)		Non-soakers grains before UV irradiation		Soaked grains before UV irradiation		Mean of seedling emergence	Mean of viability ratio
		Number of seedling emergence	Viability ratio	Number of seedling emergence	Viability ratio		
0		88.00	1.00	84.67	1.00	86.33	1.00
10		89.33	1.02	80.00	0.94	84.67	0.98
20		88.67	1.01	84.67	1.00	86.67	1.01
30		88.00	1.00	88.00	1.04	88.00	1.02
40		93.33	1.06	90.00	1.06	91.67	1.06
50		89.33	1.02	83.33	0.98	86.33	1.00
60		92.00	1.05	86.67	1.02	89.33	1.04
70		91.33	1.04	88.67	1.05	90.00	1.04
80		91.33	1.04	88.67	1.05	90.00	1.04
90		90.67	1.03	87.33	1.03	89.00	1.03
F-Test		NS	NS	NS	NS	*	*
LSD	0.05	5.61	0.060	6.59	0.079	3.710	0.041
	0.01	7.68	0.083	9.03	0.109	5.082	0.056

NS, *: Not significant and significance at 0.05 probability level, respectively.

It is expected that the induction of mutations to increase genetic diversity can cause very few chromosomal aberrations, physical damage, and sterility. At the same time, mutations can produce the desired genotypes (Datta 2011). Guided by the results of this study, hydration yield was optimum in soaked irradiated grains than in non-soakers irradiated. Mutations induced by UV radiation in the hydration coefficient of wheat gains may occur in chromosomal genomes or extra chromosomal genomes in the cytoplasm as mitochondria and chloroplast. This needs to be done through genomic analysis to determine the type of mutations that may occur. Mutations induced by UV irradiation in soaked grains may cause chromosomal deletion or DNA band deficiency. This deficiency may lead to an influence on the function of hydration (Ramezani and More 2014). The genetic change in the hydration of wheat grains may be permanent and passed down to the next generation. If this happens, then these genotypes in new individual plants have differed in their characteristics from their parent (Cober *et al.,* 2010).

Seedling emergence in irradiated grains

The results tabularized in Table 2 revealed that there were significant differences concerning seedling emergence between UV doses among non-soakers and soaker grains before UV irradiation. This indicates the presence of adequate diversity which can be exploited through selection to improve the viability of wheat grains. Meanwhile, the mean of seedling emergence and viability ratio showed significant differences in this trait between UV doses. The dose of 40 minutes of exposure time showed a significant increase over the control in seedling emergence and viability ratio. In this study, UV doses may increase or decrease the viability ratio of germination with different magnitudes for each dose. This follows the convention that UV doses are an important modifier agent that impacts wheat germination (Olaerts *et al.,* 2016). The germination promotes amylolytic activity leading starch to degradation with lower flour viscosity (Ding *et al.,* 2018). Starch degradation occurred with a longer germination time as reported by Stern *et al.* (2021). The same authors observed that there was a positive relationship between germination time and free sugar content in wheat flour. This suggests that breads manufactured from longer germinated wheat flour may be sweeter. The results obtained herein agree with Charoenthaikij *et al.* (2010), who established that no statistically significant differences were obtained in the sensory perception of bread manufactured with and without germinated flour. Despite the damaging effect of UV-B photons that limit photosynthesis and plant growth.

The damaging effect of UV-B has been considered rare (Choudhary and Agrawal 2014). The positive effects of UV-B include stimulation of secondary metabolism, phyto-reagent production, and many others (Ballaré *et al.,* 2011). Numerous studies include the positive effect of UV-C on seed health, germination, and seedling growth of different crops. It also influences physiological and biochemical processes in grains based on radiation dose (Sadeghianfar *et al.,* 2019). The results obtained herein are in line with the findings of Semenov *et al.* (2020), who observed an improvement of about 8% in seed vigor after wheat grains treated with 250 Jm^{-2} of UV-C radiation. Increasing the dose to 500 Jm-2 decreased the seed vigor to 15 - 31% if compared with the control. The magnitude of seed vigor was correlated well with germination (Semenov *et al.,* 2020). Ultraviolet irradiation in the range of 230 - 400 nm was considered non-ionizing which possesses green technology impact on several macromolecules such as proteins, DNA, RNA, etc. In this respect, Kumar *et al.* (2021) found that UV rays from the wavelength of 254 nm significantly affect the functionality of wheat flour by inducing gluten cross-linking networking patterns. In addition, Falconi and Mendizabal (2018)investigated that seedlings grown from UV-C irradiated seeds achieved enhanced concentrations of peroxidase and catalase as a defense enzymes if compared with that grown from unirradiated seeds.

Regression analysis of seedling emergence

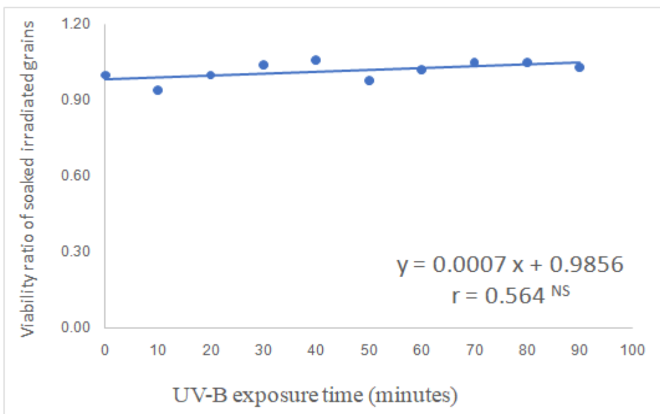

Figure 5. Regression line represents the relationship between UV-dosesand viability ratio of pre-soaked irradiated wheat grains

According to the results diagrammatic in Figure 5, the regression coefficient of 0.007 means that the viability ratio increases by 0.007 with each additional minute of UV-B irradiated soaked grains. This regression line represents the relationship between UV doses and the viability ratio of soaked irradiated wheat grains. Both variables vary in the same direction because the movements in UV doses are accompanied by movements in viability ratio. Therefore, this correlation is positive. It means that if irradiation doses are increasing, then the viability ratio is also increasing. The correlation coefficient between both variables was equal = 0.564. Then the coefficient of determination (r^2) was equal to 0.3181. Thus, 31.81% of the variance in viability ratio was attributed to UV irradiation. The rest variance of 68.19% remains unexplained and is attributed to other factors that are not present in the regression formula as variety genotype, growth conditions, water hydration chemical structure of grains, physiological state, epidermis thickness of the grains coat, and grains composition, etc. Ultraviolet irradiation is abiotic stress which significantly influences the viability of wheat grains. Generally, it is interesting to note that the influence of any stress leads to induce formation of reactive oxygen species (ROS) in plant cells (Badridze *et al.,* 2015). ROS-induced activation in various genes in Arabidopsis (Mittler *et al.,* 2004). However, UV irradiation is one of the environmental stress factors attacking plants during the whole life cycle. It is known that high doses of UV irradiation negatively influence plant growth and development. The low doses of UV-B may increase the stress adaptive reactions in plants, which leads to the activation of enzymatic and non-enzymatic defense mechanisms (Rai *et al.,* 2011). Irradiation of seeds with a full spectrum of UV influenced the plant metabolism that developed from the irradiated seeds. This leads to activating stress adaptive mechanisms through stimulation of the synthesis of antioxidants in plant cells. This depended on plant genotype, radiation intensity, and the type of antioxidant developed (Kacharava *et al.,* 2013). Pre-sowing treatment of wheat grains with UV irradiation supplies the grains with extra energy. This stress leads to produce free radicals that change cell membrane permeability. The free radicals initiate diverse metabolic responses as biosynthesis of antioxidants that generate adaptive mechanisms against unfavorable environments (Dubrov 1977). It is established that the concentration of ROS serves as a signal for switching on some genes and signal mechanism systems, leading to controlling responses to stress factors (Foyer and Noctor2003). Evidently as seen in this study UV-irradiated pre-soaked grains may stimulate to some degree the type of signaling systems in grains which serves as a signal for switching on some genes related to the viability of wheat grains. Besides, this stimulation effect was so strong, that it influenced the plan's vital processes through a long period after emergence.

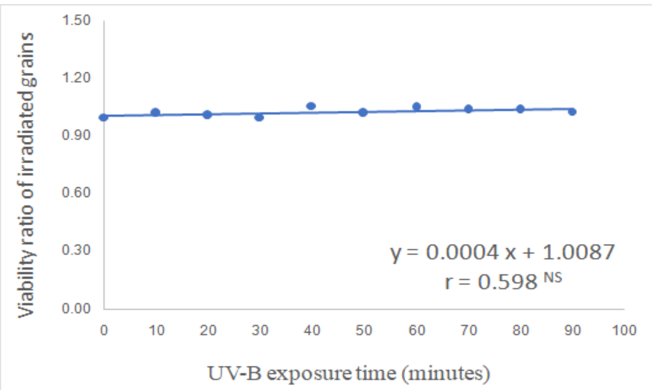

Figure 6. Regression line represents the relationship between UV-dosesand viability ratio of non-soakers irradiated wheat grains

Regarding the results diagramming in Figure 6, the regression coefficient of 0.0004 means that the viability ratio of non-soakers irradiated grains increases by 0.0004 with each additional minute of UV irradiation inside this Figure. Mathematically it is possible to assess the viability of irradiated grains whose doses are outside the range used in this study. Such an extrapolation is generally not useful. The regression coefficient indicates the changes in viability ratio that correspond to a change in UV doses. Therefore, regression analysis is a powerful procedure with many applications in genetics research. The correlation coefficient between the viability ratio of non-soakers irradiated grains and the doses of UV-irradiation was equal to 0.598. Then, the coefficient of determination resulting from squaring this number was equal to 0.3576. This indicated that 35.76% of the variance in viability ratio was attributed to UV irradiation. The rest variance of 64.24% is attributed to other factors such as wheat genotype, hydration coefficient, temperature, grains composition, etc. Thus, the correlation coefficient helps to identify the percentage of viability ratio that can be attributed to UV irradiation. The results obtained in this study are in line with the finding of Calderini *et al.* (2008), who established that wheat biomass was unaffected by UV-B irradiation due to adaptive biochemical responses involved in acclimation which protect the embryos inside the grains from the harmful effect of irradiation. During the stress of UV irradiation, some enzymes as catalase accumulate in peroxisomes and then destroy hydrogen peroxide. Under this stress, the number of peroxisomes increases in the cell. This leads to the detoxification of hydrogen peroxide then diffused in peroxisome from the other parts of the cell (Mittler 2002). Peroxidase is another enzyme produced during stress conditions of UV irradiation. Its main function is to protect the cell or organism from the harmful effects of hydrogen peroxide. Peroxidase plays an important role in plant metabolism (Passardi *et al.,* 2005). Peroxidases neutralize the hydrogen peroxide generated during UV-B stress to take an active part in the adaptation of plant organs as wheat grains used in this study to avoid unfavorable environments. In addition, cell wall peroxidases play an important role in the formation of ROS, which induces a protective part against abiotic UV-B stresses.

It activated stress-defensive mechanisms (Mika *et al.,* 2004). Under the stress of UV-irradiation catalase-synthesizing genes take place in the cell to increase activity in irradiated grains (Scandalios *et al.,* 1997). Therefore, Badridze *et al.* (2015) suppose that UV-irradiated grains of wheat caused the intensification of respiration in mitochondria and increased the content of ROS in plant cells. Thus, the same authors established that activation of catalase during UV stress for neutralizing these stressors would be a logical result. Activation of peroxidase under abiotic stress is the result of essential shifts in the metabolism of the respiration system. This aiming adaptation of plant cells and retention of cell homeostasis (Tucic *et al.,* 2007). Accordingly, irradiated wheat grains changed the metabolic processes to varied directions, since the increase in peroxidase activity takes place to avoid the sensitivity to UV irradiation, which may lead to an increased viability ratio as seen in this study. It must be mentioned that despite the important role in ROS detoxication the enzymatic antioxidative mechanism was not able to provide full protection in plant cells (Blokhina *et al.,* 2003). The results obtained in this study confirm the data obtained by Tertyshnaya *et al.* (2018), who examined the impact of UV-B radiation on seed germination of spring and winter wheat. The authors established that wheat germination was increased by 1-3% as a result of grains irradiated with UV. Moreover, Semenov *et al.* (2020) found a significant increase in wheat germination by 38% at the optimal radiation dose of 250 Jm-2. The same authors established that there is no direct dependence between the exposure time and seed germination. According to the results of Semenov *et al.* (2020), who revealed that cultivar properties play a significant role in genotype reaction with irradiation stress in determining seed vigor. Thus, the same authors found that 28% of the total variations in seed vigor of wheat are explained by the genotype properties, and 67% was attributed to the doses of irradiation. For germination indices, Semenov *et al.* (2020) found that 46% of the total variation in germination was stipulated by genotype characteristics, and 47% was attributed to irradiation doses. The results of UV-B irradiation showed a stimulating effect on the improvement of grain viability.

Glucose standard curve

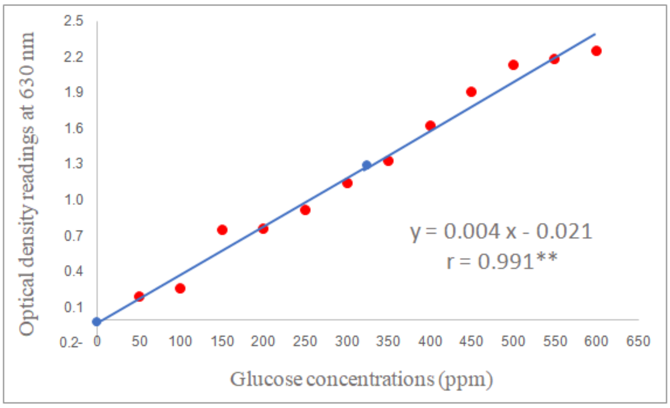

Figure 7.Glucose standard curve represents the relationship between glucose concentrations and their optical density readings at 630 nm

According to the results of the glucose standard curve (Figure 7), the regression coefficient of 0.004 means that the optical density readings increase by 0.004 with each additional ppm of glucose. The constant (a) was equal to - 0.021 in the regression equation. Linear regression was used to estimate the glucose concentration of any optical density reading that lies within the observed data. So, the regression coefficient exhibited a change in the dependent variable that corresponds with a change in the independent variable. These data describe the relationship between the optical density influenced by glucose concentrations. The variable to be explained is the response variable or the dependent variable as optical density readings. The variable that explains the response variable was called the independent variable or predictor variable as glucose concentrations. Measurement of association between variables reflected the initial impression about the extent of statistical dependence between both variables. The linear regression line in the glucose standard curve was used to estimate the values of glucose concentrations from the corresponding optical density reading observed as the outcomes of glucose concentrations. The correlation coefficient between glucose concentrations and their outcomes of optical density reading was equal to 0.991. Then, the coefficient of determination (r^2) obtained from squaring this number was 0.98208. This means that 98.21% of the variance in optical density readings can be attributed to glucose concentrations. The rest variance of 1.79% remains unexplained and is attributed to other factors as the experimental errors that were not included in the regression equation. The correlation obtained herein is strong because its value was above 0.7. This is in agreement with Moore *et al.* (2013), who stated that correlations above 0.7 are considered strong because if $r = 0.7$, then $r^2 = 0.49$. This means that about 50% of the variance in the dependent variable can be explained by the independent variable, and the rest variance was not included in the regression equation.

Carbohydrates in irradiated grains

The results tabularized in Table 3 indicate the contents of carbohydrates in soakers and non-soakers irradiated grains. The results showed significant differences between irradiation treatments for the contents of carbohydrates among non-soakers and soaked irradiated grains. This indicated the presence of considerable variability among irradiation doses for carbohydrate contents in wheat grains. Most of the irradiation doses 10, 20, 40, 60, 70, 80, and 90 minutes showed a significant decrease in carbohydrate contents of non-soaked irradiated grains. In contrast, most doses of UV irradiation achieved a significant increase over the control in pre-soaked irradiated grains. The doses revealed a significant increase in carbohydrates as 10, 40, 50, 70, 80, and 90 minutes of exposure time to UV irradiation. These results indicated that UV-irradiated non-soaker grains appeared to be suppressing the formation of soluble carbohydrates. Thus, the sharp decrease in soluble carbohydrates under UV-B stress was shown in non-soaker grains. Meanwhile, the high content of soluble carbohydrates in pre-soaked irradiated grains was induced by most doses of UV-B stress. This indicated that hydrated grains before UV irradiation stimulated the formation of soluble carbohydrates. This leads to an increase in the fermentation power of yeast cells. The soluble carbohydrate development is related to the metabolic ways of ROS. These are formed as the results of UV stress which regulates gene expression relating to increased soluble carbohydrate contents as seen before in by Hidema *et al.* (2005) concerning the storage protein in rice grains which increased due to elevated UV-B irradiation. The formation of ROS leads to the activation of different genes (Mittler *et al.,* 2004). Thus, ROS plays an important role in the neutralization of hydrogen peroxide formed during UV stress to take an active part in the adaptation of plant cells to unfavorable environmental conditions such as UV stress. The results obtained herein are in line with the finding of Badridze *et al.* (2015), who demonstrated that abiotic stress increased the level of glucose and sucrose as well. Additionally, Van den Ende and Valluru (2009) suppose that sucrose may protect cell membranes against cold and drought stressors. Pre-soaked UV irradiation produced free radicals during this stress leading to changes in cell membrane permeability. This exhibited hydration coefficient in soaked irradiated grains presumably initiates diverse metabolic responses including the biosynthesis of antioxidants. These from its side must be reflected formation of adaptive mechanisms against UV stress (Dubrov 1977). The accumulation of soluble carbohydrates in pre-soaked irradiated grains as shown herein in response to UV stress was shown in different plant parts in several works as Finkelstein and Gibson (2001), Nayer and Reza (2008), and Prado *et al.* (2000). From the points of view, it is established that the metabolism of soluble carbohydrates generated under the stress of UV-irradiation is a dynamic tool comprises reactions of catabolism, as well as, synthesis simultaneously (Hilal *et al.,* 2004).

Table 3. Proximate composition of carbohydrates in UV irradiated grains of wheat

Doses of UV (minutes)		Non-soakers grains before UV irradiation		Soaked grains before UV irradiation	
		Carbohydrates content	Yield ratio	Carbohydrates content	Yield ratio
0		36.26	1.00	34.31	1.00
10		34.92	0.96	36.30	1.06
20		30.46	0.84	33.72	0.98
30		37.20	1.03	34.04	0.99
40		33.99	0.94	36.64	1.07
50		34.14	0.94	35.61	1.04
60		29.99	0.83	32.67	0.95
70		26.37	0.73	35.55	1.04
80		26.86	0.74	36.38	1.06
90		32.61	0.90	35.59	1.04
F-Test		**	**	**	**
LSD	0.05	0.970	0.027	0.568	0.017
	0.01	1.329	0.036	0.779	0.023

** : Significance at 0.01 probability level.

Significant decrease of carbohydrates in non-soakers irradiated grains if compared with the control may be due to the intensification of their respiration (Hilal *et al.,* 2004). This decrease in dry irradiated grains may be attributed to an inactive form of alpha-amylase which cannot break down carbohydrate complexes into more absorbable sugars as reported before by Hung *et al.* (2011). Barsing and Malz (2000) suggested that the significant effect of UV-B-exposed maize leaves was a decrease in glucose, whereas the sucrose content was not decreased. In contrast, Garrard *et al.* (1977) decided that UV-B-sensitive phanerogams were characterized by the reduced values of sucrose. The same authors used carbohydrates as a marker for tolerance to enhanced UV-B. They decided that maize leaves were tolerant to enhanced doses of UV-B irradiation if carbohydrates were used as a marker.

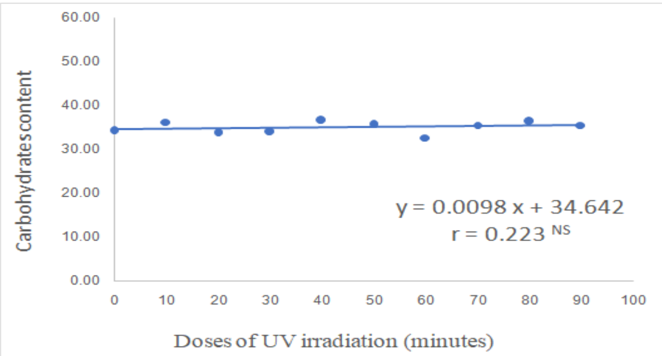

Figure 8. Regression line of carbohydrates composition in pre-soaked irradiated wheat grains

According to the results diagrammatic in Figure 8, the regression coefficient of pre-soaked irradiated grains of 0.0098 means that soluble carbohydrate contents increase by 0.0098 with each additional dose of UV-B irradiation inside this model. Therefore, this linear regression can be used to estimate the changes in carbohydrate contents for any dose of UV irradiation that lies within the observed doses in this model. Mathematically it is possible to estimate carbohydrate contents for UV-B doses outside the range of doses used in this study. This extrapolation is generally not useful. The correlation coefficient obtained in this model between the doses of UV-B irradiation and corresponding values of carbohydrates was equal to 0.223. Squaring this number produced the coefficient of determination (r2) = 0.0497. This means that about 4.97% of the variance in carbohydrate contents is attributed to UV-B doses. The rest variance of 95.03% remains unexplained because they are not included in the regression equation.

This may be due to other factors such as alpha-amylase activity, hydration coefficient, cell membrane permeability, grains composition, epidermis thickness, physiological state, etc.

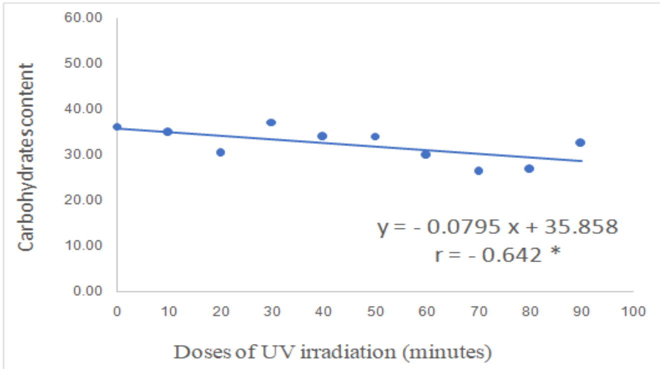

Figure 9. Regression line of carbohydrates composition in non-soakers irradiated wheat grains

Therefore, represents the percentage of carbohydrate contents that can be explained by the doses of UV irradiation. Regarding the results presented in Figure 9, the regression and correlation coefficients between UV-B doses and carbohydrate contents in non-soakers irradiated grains were negative. This means that if irradiation doses increase the carbohydrate content decreases. Then the correlation and regression are said to be negative. This indicated that both variables vary in opposite directions. The regression coefficient of - 0.0795 means that carbohydrate content in dry-irradiated grains decreases by - 0.0795 with each additional unit of UV-B irradiation. So, the abiotic stress of UV-B decreased the level of glucose and sucrose as well. This reflects the metabolism of soluble carbohydrates under stress conditions of dry irradiated grains as a dynamic process stimulated catabolism and synthesis simultaneously. This agrees with the work of Van den Ende and Valluru (2009), who established that the cells under the effect of different stresses such as UV-B, drought, and cold may metabolize carbohydrates to protect their membranes. Therefore, the accumulation of soluble carbohydrates in response to these stressors was decreased, which may be due to the intensification of their respiration (Hilal *et al.,* 2004). Under stress conditions of UV-B irradiation of non-soaker grains, soluble carbohydrates may take part in metabolism via intensive synthesis of phenolic compounds to protect cell membranes against these stresses. During UV-B stress of non-soaker grains before irradiation, the main storage of biopolymers, namely carbohydrates are hydrolyzed by hydrolytic enzymes to lower molecular weight compounds, which are inactive in non-irradiated grains (Guardianelli *et al.,* 2019). Therefore, the decrease in carbohydrates in non-soaker grains before irradiation may be attributed to the activity increase in alpha-amylase. This enzyme breaks down the complex carbohydrates into lower molecular weight molecules as more absorbable sugars (Hung *et al.,* 2011). In addition, carbohydrate contents were lower in non-soakers irradiated grains than in soaked irradiated. This could be due to the utilization of carbohydrates in non-soakers for biochemical activities of protecting the cell membrane against UV irradiation (Wang *et al.,* 1997). The correlation coefficient between UV doses and carbohydrate contents in non-soakers irradiated grains was equal to - 0.642. This is a significant negative correlation because both variables vary in opposite directions. If one variable increases as UV doses, the other variable as carbohydrate contents decreases. Then, this correlation is said to be a negative correlation. Squaring correlation coefficient induced the coefficient of determination (r^2). This is a measure of how well the regression model describes the obtained results. The coefficient of determination for this relationship between doses and carbohydrate contents is 0.4122. This means that 41.22% of the variance in carbohydrate contents is due to UV-B irradiation. The remaining variance of 58.78% is due to other factors that are not included in the regression equation as wheat genotype, carbohydrates accumulated, enzyme activity, moisture content, etc. This is in line with the findings of Schneider *et al.* (2010).

Homogeneity of quality traits in irradiated grains

The degree of homogeneity was assessed based on the coefficient of variability (CV) as shown in Table 4. It was used to determine the magnitude of variation within every irradiation dose. The hydration coefficient in non-soaker grains before UV irradiation showed coefficient of variance values higher than the check treatment in the control at the doses of 10, 30, 40, 50, and 60 minutes of UV irradiation. These doses produced high heterogeneity in the hydration coefficient, since they gave coefficient of variance (CV) values higher than that of the check treatment. The other doses of UV irradiations including 20, 70, 80, and 90 minutes of exposure time showed CV values lower than the check treatment. This indicated high homogeneity in the hydration coefficient of non-soakers irradiated grains. Thus, these doses exhibited higher uniformity in hydration coefficient than other doses. Pre-soaked irradiated grains showed the same trend of homonymity in hydration coefficient at all doses of UV irradiation, except for the dose of 20 minutes achieved high homogeneity in this trait. Concerning seedling emergence, the non-soaker grains before irradiation recorded the highest heterogeneity at the doses of 10, 50, and 60 minutes of exposure time to UV irradiation. This is because the coefficient of variance values higher than that in the check control, indicating that they were not uniform in grain viability. This reflected that these doses produced substantial variation and could be utilized in different breeding programs. The other doses of UV-irradiation 20, 30, 40, 70, 80, and 90 minutes of UV exposure time showed CV values lower or close to the check. This indicated that the viability of wheat grains was highly homogeneous at these doses. Thus, the viability of non-soaker grains before irradiation was highly uniform at these doses. Moreover, the seedling emergence of pre-soaked irradiated grains showed CV values higher than the check at most doses of UV irradiation at 10, 20, 40, 60, 70, 80, and 90 minutes of exposure time.

Table 4. Homogeneity of some quality traits in UV irradiated wheat grains

Doses of UV irradiation (minutes)	Hydration coefficient		Seedling emergence		Carbohydrates content	
	Non-soakers	Soaked	Non-soakers	Soaked	Non-soakers	Soaked
0	0.123	0.155	0.038	0.013	0.003	0.002
10	0.137	0.023	0.046	0.070	0.002	0.001
20	0.087	0.165	0.034	0.027	0.042	0.005
30	0.158	0.028	0.038	0.000	0.010	0.013
40	0.135	0.072	0.026	0.040	0.010	0.022
50	0.194	0.017	0.046	0.013	0.008	0.001
60	0.253	0.013	0.044	0.035	0.027	0.007
70	0.056	0.017	0.026	0.035	0.026	0.003
80	0.118	0.103	0.013	0.058	0.008	0.003
90	0.122	0.008	0.013	0.075	0.004	0.007

This indicated high heterogeneity in grain viability at these doses. Therefore, these doses exhibited high variations in the viability of pre-socked irradiated grains, since they showed higher values of CV than that of the check. Additionally, the other doses at 30 and 50 minutes recorded CV values lower or close to the check, indicating high homogeneity in this trait. Thus, the doses of 30 and 50 minutes exhibited higher uniformity in grain viability from the pre-soaked irradiation group than other doses of UV irradiation. Regarding carbohydrate contents, the third qualitative trait studied in this investigation, non-soakers irradiated grains recorded CV values higher than the check at all doses of UV-irradiation, except for the dose of 10 minutes. This could be considered the highest heterogeneous in carbohydrate contents at these doses, where they gave the highest variations within their grains. This indicated that carbohydrate contents at the doses of 20, 30, 40, 50, 60, 70, 80, and 90 minutes of UV exposure time were considered more chemically varied than at the dose of 10 minutes of exposure time. The later dose of UV showed a CV value lower than the check, indicating that carbohydrate content at this dose was more chemically uniform than at other doses. This indicated high homogeneity in carbohydrates containing grains at the dose of 10 minutes irradiation time.

On the other hand, pre-soaked irradiated grains recorded CV values higher than that in the check at the doses of 20, 30, 40, 70, 80, and 90 minutes of exposure time. This indicated the highest heterogeneity in carbohydrate contents in pre-soaked irradiated grains at these doses, where they recorded the highest variations within their contents. This indicated high heterogeneity in carbohydrate contents since they exhibited high variations at these doses. The doses of 10 and 50 minutes exhibited high homogeneity in carbohydrate contents of pre-soaked irradiated grains since they recorded CV values lower than the check. This indicated that carbohydrate contents at these doses were more phenotypically uniform than at the other doses. The results obtained herein are in line with the findings of Ahmed *et al.* (2017), who developed new lines of tomatoes through a selection of plants in a generation that is homogeneous enough for all the studied traits, which exhibited low CV values. Furthermore, El-Morsy *et al.* (2021) established that the values of CV were lower than the check indicating high homogeneity in this trait. If the CV values in any trait are higher than that in the check, this indicates high heterogeneity. This reflected that the genotypes were more phenotypically variant than other genotypes. The results obtained in this study are confirmed with those found by Mona and Mahmoud (2019), who selected new lines of tomato from the F2 generation which exhibited lower CV values than that in the check cultivar. Furthermore, Kumamaru *et al.* (1988)

selected rice mutants carrying changes in glutelin content and improved grain storage protein. Additionally, Hidema *et al.* (2005) reported that elevated UV-B irradiation influenced the accumulation of storage protein and the expression of genes related to protein biosynthesis. Regarding these effects, elevated UV-B irradiation regulates gene expression of defense genes in an up or down direction and the photosynthetic proteins (Takeuchi *et al.,* 2002). The results obtained in this study are in harmony with Hidema *et al.* (2005), who found that not only elevated UV-B irradiation was markedly influenced by rice grain size but also by grain storage protein.

Coefficient of variation in grain quality traits

The phenotypic and genotypic variability parameters play a significant role in wheat breeding programs through hybridization followed by selection (Aye *et al.,* 2018). Genetic variability parameters are one of the important considerations in wheat improvement technique which needs to be studied in detail. Variability was measured by the assessment of phenotypic (σ^2p) and genotypic (σ^2g) variance, heritability (H), phenotypic (PCV) and genotypic (GCV) coefficient of variation, expected genetic advance (EGA) and genetic advance as percent of the mean (GAM). Genetic parameters helped the wheat breeders in selection to improve the desired quality traits. Furthermore, environmental factors play an important role in gene expression reflected in the phenotype. The observed phenotypic variability includes heritable genotypic and non-heritable environmental variation. Therefore, variability can be assessed from biometric markers such as GCV, genetic advance, and heritability in a broad sense. As stated in the results tabularized in Table 5, the PCV was greater than GCV for hydration coefficient and seedling emergence among non-soakers and soakers irradiated grains. This indicated that environmental factors played an important role in the expression of these traits. According to Shivasubramanian and Menon (1973), the PCV and GCV were ranked as follows, low (0-10 %), medium (10-20 %), and high (> 20%). Likewise, GCV obtained for hydration coefficient and seedling emergence were considered low because they are ranged between 0.884 - 7.03. This indicated that the apparent variation is not only due to genotypes but also mainly influenced by environmental factors (Bandila *et al.,* 2011).

Table 5. Genetic variability parameters of some quality traits in wheat grains

Genetic parameters	Hydration coefficient		Seedling emergence		Carbohydrates	
	Non - Soakers	Soakers	Non - Soakers	Soakers	Non - Soakers	Soakers
σ^2G	0.0007	0.0017	3.44	4.266	0.010	0.001
σ^2E	0.0075	0.006	10.21	14.77	0.0002	0.0001
σ^2P	0.008	0.008	13.65	19.036	0.011	0.001
PCV %	3.03	2.87	12.30	14.86	3.51	1.20
GCV %	0.884	1.35	6.175	7.03	3.35	1.17
ECV %	2.90	2.54	10.639	13.08	0.003	0.001
H %	8.48	22.077	25.20	22.41	90.91	93.95
EGA	0.02	0.04	1.92	2.01	0.19	0.07
GAM %	1.76	4.25	2.12	2.33	21.04	7.20

σ^2G, genotypic variance; σ^2E, environmental variance;σ^2P, phenotypic variance; PCV (%), phenotypic coefficient of variation in percent; GCV (%), genotypic coefficient of variation in percent; ECV (%), environmental coefficient of variation in percent; H (%), heritability in broad sense in percent; EGA, expected genetic advance; GAM (%), genetic advance as percent of mean at 5% selection intensity.

Thus, these traits offered less scope of selection because they are mainly influenced by environmental factors (Panse 1957). These results agrees with the Chand *et al.* (2008), who established that days needed to maturity in barley appeared to have considerable variability leading to little opportunity for improvement through selection. The magnitude of PCV was found to be slightly higher than their respective GCV for hydration coefficient and seedling emergence which might have resulted from the influence of the environmental factors on the development of these traits. This is in agreement with Abebe *et al.* (2017), who reported that a considerable difference between the phenotypic and genotypic coefficient of variations means greater effects of environmental factors on the phenotypic expression. Thus, selection based on the phenotype would be ineffective in bringing considerable genetic improvement. So, genetic diversity is a prerequisite to beselect superior genotypes in wheat. Human selection over thousands of years led to the loss the potentially important allelic variation. Programs aimed to increase genetic variations through crosses and mutagenesis within wheat crops can dramatically improve the efficiency of breeding techniques. Based on the classification by Shivasubramanian and Menon (1973), the PCV of seedling emergence was considered medium because it ranged between 12.30 % (non-soakers grains) – 14.86 % (soakers grains). Among hydration coefficient and seedling emergence, the ECVs were greater than the corresponding values of GCV over non-soakers and soaker grains. This indicated the greater share of environmental variance in the total variability of hydration coefficient and seedling emergence. Thus, these traits were governed by non-addition gene action. Besides, selection may be ineffective for the genetic improvement of these traits. These results agree with the work of Hailu *et al.* (2016), who observed low values in GCV and PCV (< 10%) for days to heading and maturity in all environments cultivated barley (*Hordeumvulgare* L.). The phenotypic variance was greater than the genotypic variance for hydration coefficient and seedling emergence among non-soakers and soaker grains. This indicated that the environmental factors had a greater share in total variability concerning the expression of these traits. Therefore, direct selection for these traits may be ineffective since it is exhibit a greater share of environmental factors. So, grain quality traits improvement in wheat is one of the most important objectives that is directly related to bread making industry, as well as, flour quality. Thus, the present study was carried out to analyze genetic variability parameters of qualitative traits in wheat grains to understand the extent to which the variations observed are due to genetic or environmental factors.

The concept of heritability assessment in this study explains whether variations obtained in grain quality parameters arose as a result of differences in genetic makeup or due to environmental conditions. Based on Singh (2001), heritability was categorized into five categories: low < 40%, medium 40-59 %, moderately high heritability 60-70%, and very high 80% or more. According to this classification, the heritability obtained in this study for hydration coefficient and seedling emergence among soakers and non-soaked grains was low (< 40%). This because the obtained heritability in these traits was ranged between 8.48 - 25.20%. The lower heritability obtained leading selection may be considerably difficult due to the masking effect of the environment. Thus, heritability is a good index and reliable indicator for gene transmission from parents to their offspring (Falconer 1989). Heritability estimated would help plant breeders select the elite genotypes from diverse genetic populations. The estimates of heritability are very important if expressed in terms of genetic advance. It is interesting to note that a trait that has a high heritability value doesn't need to also exhibit high genetic advance (Johnson *et al.,* 1955). On the other hand, GCV is not enough measure to understand the heritable variation present in the population but should be taken together with heritability estimates. Besides, heritability values expressed in terms of genetic advancement are more advantageous than heritability estimates alone in predicting the results of selecting the superior genotypes (Johnson *et al.,* 1955). Thus, assessment of heritability and genetic advancement in combination are more advantageous for selection than heritability alone. Based on Johnson *et al.* (1955), the genetic advance as a percent of the mean (GAM) was classified as high (> 20%), medium (10-20%) and low (0-10%). Accordingly, low GAM was obtained in hydration coefficient and seedling emergence because their values ranged between 1.76 and 4.25%. Therefore, these traits were controlled by non-additive gene effects and phenotypic selection would be ineffective for their improvements. This indicates the higher influence of environmental factors in their expression and the prevalence of non-additive gene action in their inheritance. These results are in line with the findings of Tigabu *et al.* (2021), who observed low heritability coupled with low genetic advance for biomass yield and oil content in ten sesame varieties which may be due to non-additive gene action. The results also agreed with Ejara *et al.* (2018), who investigated low heritability coupled with low genetic advance as a percent of the mean for seeds number per pod in common bean and grain yield/ha which is due to the influence of environmental factors that limit the scope of genetic improvement by selection. Additionally, Dursun (2007) also found low broad-sense heritability estimates for grain yield /ha in common beans. The phenotypic and genotypic variances cannot be used for directly comparing the magnitude of variability. However, the coefficients of variations among phenotypic and genotypic levels are used to compare the investigated variability. In this study, the GCV values were lower than PCV without a greater difference in carbohydrate contents. This indicated that the genetic factors had an important role in the expression of carbohydrate contents in wheat grains. The PCV and GCV of carbohydrate contents were low values because they ranged between 1.17 - 3.51 %, based on Shivasubramanian and Menon (1973), who ranked the low values that ranged from 0 to 10%. Thus, there is a scope for selection based on carbohydrate content.

Then, the diverse genotypes obtained from UV-B irradiation among soaked and non-soaker grains can provide materials for a sound breeding program. The results agreed with Sudhakar *et al.* (2007), who investigated low phenotypic and genotypic coefficient of variation for the traits days to fifty percent flowering, days to maturity, and oil content in sesame. So, the genotype with a wide range of genetic variation for carbohydrate grain contents and extensive genetic variation is recorded as a potential genotype. Therefore, high genetic variability in the wheat population plays an important role in wheat breeding programs, as well as, in developing new genotypes to sustain the level of high carbohydrate productivity. A high variation induced in carbohydrate contents of wheat grains is novel recombinant. Thus, mutagenesis could be used as a successful method to develop new genotypes in wheat with high-grain carbohydrate contents. The results indicated that carbohydrates in wheat grains are governed by additive gene action. Hence, direct selection based on this trait would be effective for the improvement of carbohydrates in wheat grains. No great difference was obtained between PCV and GCV estimates indicating a scope of selection for improvement carbohydrates in wheat grains. This is because one of the important goals in wheat breeding programs is to improve varieties with high grain carbohydrate content. The lower difference obtained between PCV and GCV estimates indicated that the observed phenotypic variation in carbohydrate grain contents was mainly attributed to genetic effect. This is because PCV and GCV estimates are very close to each other for carbohydrate contents. Thus, the little difference obtained between PCV and GCV values indicated that the observed phenotype was majorly attributed to genetic factors. In contrast, the greater differences between PCV and GCV indicated the greater contribution of environmental factors to the observed phenotype (Bandila *et al.,* 2011). Based on Singh (2001), very high heritability (80% or more) values in the broad sense were obtained for carbohydrate contents which ranged between 90.91% (non-soakers) to 93.95% (soakers). Carbohydrate contents in wheat grains exhibited very high estimates in the broad sense indicating the minimum effect of environmental factors on the phenotypic expression of this trait. This leads to the effectiveness of selection in the improvement of carbohydrates in wheat grains (Singh 2001). In contrast, if the trait has low values in heritability estimates, then selection may be considerably difficult due to the masking effect of the environmental factors (Luzi-Kihupi 1998). Therefore, heritability is an important genetic parameter that determines the effectiveness of selection based on the heritability category. So, selection for carbohydrates in wheat grains will be more effective as a major portion of genetic variation available in the population which is exhibiting additive gene effects.

So, the genetic advance is another important key as a genetic parameter for select, the superior genotypes in wheat. The genetic advance obtained for carbohydrate contents ranged between 7.2 (low < 10%) in soaked grains to 21.04 (high > 20%) in non-soakers based on the categorization by Yeshiwas and Negash (2017). The results indicated that very high heritability values were coupled with high or low genetic advance. Thus, carbohydrate contents in wheat grains were controlled by additive gene action and the phenotypic selection based on biochemical analysis of carbohydrates would likely be effective. This indicates the lesser effects of environmental factors in the expression of carbohydrates in wheat grains because of additive gene effects prevalence in their inheritance. Thus, low heritability coupled with low genetic advancement may be attributed to non-additive gene action. Similar, results were reported by Krishnaiah *et al.* (2002) in sesame. The spectrum of mutations induced by ultraviolet rays in carbohydrate contents is more diverse. Using similar investigations, Sato *et al.* (2006) assessed the rate of mutations induced in rice by gamma rays to be one mutation induced per 6190 kb. Meanwhile, Hwang *et al.* (2016) found that the mean number of mutations induced in M2 rice-irradiated genotypes was 1/ 492 per gene, as well, the percentage of mutation sites induced per total sequence was 0.67. The high genetic diversity obtained the this study for carbohydrate contents suggests that the wheat population is suitable for improving this trait through direct selection. Thus, this study highlights the efficacy of employing phenotypic and genotypic screening approaches. This will provide good information on gene expression related to carbohydrates accumulated in wheat grains, as well, as provide markers to facilitate wheat breeding technique, particularly on topics surrounding the breed wheat industry and their technology. In general, high coefficient of variability estimates indicates that there were a scope of selection for improvement carbohydrate contents in wheat grains. In contrast, the low values of estimates as seen in hydration coefficient and seedling emergence indicate that the genotypes investigated need for the creation of variability either by mutation followed by selection (Tiwari *et al.,* 2011). Therefore, heritability alone did not indicate the amount of genetic improvement that resulted from the selection of the individual genotypes. Thus, information about heritability coupled with genetic advancement and GCV are most useful, because they would give a more reliable selection value (Fentie *et al.,* 2014). According to Johnson *et al.* (1955), high heritability coupled with high genetic advance is usually more helpful in predicting selection gain than heritability assessment alone. So, emphasis should be placed on the traits such as carbohydrates in wheat grains that had very high heritability coupled with high genetic advance for formulating reliable selection program indices for genetic improvement and development of high-yielding genotypes with a significant increase of carbohydrates in their grains.

Concluding remarks

In this study, the effect of UV irradiation on some quality traits of non-soakers and pre-soaked irradiated grains were investigated. The results appeared that UV-B irradiation had a positive effect in pre-soaked irradiated grains for improving hydration coefficient, grains viability and carbohydrate contents. Pre-soaked irradiated grains showed more efficient activity in improving quality traits in wheat grains than that in non-soakers irradiated. UV treatment of wheat grains had neither a positive effect as seen in pre-soaked irradiated grains nor a negative effect as seen in non-soakers irradiated. UV treated wheat grains is a promising environmentally friendly and a cheap way for sterilize the grains against the stores insects and pathogens before storage in silos. It is also increased the energy of germination in pre-soaked irradiated grains than in non-soakers irradiated. The process of pre-soaked irradiated grains has the ability to degrade starch into soluble carbohydrates. Therefore, UV sterilize wheat grains before storage in silos followed by washing with water before grinding is recommended before bread making industry to increase the quality of products. So, pre-soaked irradiated grains induced full stimulation rate of enzymatic system leading to increase the quality of grains and wheat flour to be used for produced high quality products. Pre-soaked irradiated grains activate the synthesis of phenolic compounds to protect the cell system against stress with the phenil propanpidway of phenolics formation. The decrease in soluble carbohydrates in non-soakers irradiated grains may caused by the inactive form of alpha-amylase which showed a negative correlation with the doses of UV-irradiation. Increased glucose content in pre-soaked irradiated grains improved flour functionality via less starch containing and less weakening. These leading to longer mixing stability time during bread dough mixing. Therefore, pre-soaked wheat grains before irradiation improved bread quality to be acceptable for consumers. This could limit the need for added sugar to bread dough for improved fermentation and rheological characteristics, to preserve the sensory characteristics of product. Furthermore, the use of UV-treatment for sterilizing wheat grains before storage in silos prolong shelf life and reduce the growth of fungal species and insects which contaminate most bakery products. This needed designed a system of multiple UV lamps in silos that allows the radiation to reach every point of the silos. Radiation attenuation and the effects of exposure on the packaging food material are important to considerations in system design to ensure the system efficiency. Pre-soaked grains before irradiation seems to bring out some adaptive mechanism to reduce the damage effects of UV irradiation, despite wide genetic and physiological differences. So, mutations induced by UV irradiation in pre-soaked wheat grains is a valuable tool for improving morpho-physiological and quality traits in bread wheat. Pre-soaked grains provided an optimum amount of absorbed radiation energy leading to the maximum positive effect. This study is a suitable technology to be used for improving the baking characteristics of wheat flour without using chemicals to suppress fungi growth on wheat grains served in silos. It also provides insight on the effect of UV irradiation on the molecular response expressed in pre-soaking and non-soakers irradiated grains. Further investigation is needed for clearing information'sabout the biochemical activities that may be happen in the cell due to the interaction between UV irradiation and the water treated grains before irradiation.

Conflict of interest statement

The author declares that this manuscript was done in the absence of any commercial or financial relationships that could be conducted as a potential conflict of interest.

Author's contribution statement

All research stages including laboratory works, collection and analysis of data, write and revised the manuscript in its final form were all done by the author.

Ethical approval

This study does not indicate any human or animal testing or feeding on irradiated products.

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