RELATIONSHIPS BETWEEN WATER ABSORPTION RATE AND ULTRAWEAK LUMINESCENCE FROM WHEAT GRAIN AT THE INITIAL STAGE OF SWELLING

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A b s t r a c t. The comparative studies of water absorption rate (V) and the intensity (I) of ultraweak luminescence (UWL) emitted by samples of wheat grain during the first 15 min of swelling were carried out. The V rate was estimated using the capillary method in which the volume changes of water in the capillary vessel within each 1 min period were measured. The intensity I was measured using high sensitivity apparatus working in the single-photon-counting arrangement. As a result of the experiment several kinetic curves of V rate and the intensity I, as well as the dependencies between V and I obtained in the initial periods of grain imbibition, have been presented. It was found that the V rate and the intensity I depend on the thickness, mechanical damage, initial moisture content and temperature of grain. Comparing the results, it has been found explicitly that the intensity I depends on the water absorption rate V by wheat grain.

K e y w o r d s: water absorption, wheat grain, luminiscence

INTRODUCTION

Wheat grain, like other cereal seeds, is hygroscopic and quickly reacts to the changes of humidity in every environment. Hence the amount of water occurring in grain is always varying in relation to the condition prevailing in the environment to which it is exposed. Under the conditions suitable for storage the moisture of grain is the result of the existence of only bounded water [1]. In such cases, the colloidal-porosity systems of cell walls, protoplasm and stock substances are dehydrated and the grain rapidly absorbs the water at the initial stage of imbibition.

It is well known that water penetration into the grain plays a significant role in numerous processes, not only physiological. For example, the increase of moisture and rise in volume of the swelling grains influence many physical factors of fundamental importance in the technological processes of transporting, cleaning, drying and storing. On the other hand, the entry of water into the wheat grains has also considerable interest because of its importance in the conditioning studies for milling. Hence several authors have looked into the methodology for the evaluation and measurement of the water absorption rate by grain at the initial stage of swelling [1-8,10].

It has been reported in a previous paper [9] that imbibing wheat grains reveals ultraweak luminescence (UWL) detected by a photon-counting device of high sensitivity in the spectral region from 300 nm to 760 nm. In the study it has been postulated that this phenomenon is generated by processes binding the water molecules in the seed coats and/or aleurone layer as well as in its endosperm. The obtained results suggest also that the intensity (I) of UWL depends on the V rate of the water uptake by wheat grain. For further confirmation of this fact comparative studies on relationships between water absorption rate V and intensity *I* of UWL emitted in the first 15 min of the wheat grain swelling were undertaken in this paper. The studies were also conducted to investigate effects of thickness (*d*), pressure (*p*) of mechanical loads damaging the samples, moisture content (m_c), and temperature (*T*) of the grain on both the *V* rate and intensity *I*.

MATERIALS AND METHODS

The subject of the investigation was a parcel of manually harvested grain of winter wheat Liwilla variety grown in 1989. The grain used in the experiment was properly cleaned and dried in the standard condition in the laboratory obtaining equilibrium moisture content of 10.7 % after four weeks of storage. The mean germination capacity of grain was about 93 ± 2 %. Next, the parcel was divided into three fractions of grain thickness d in intervals: 2.0-2.5 mm, 2.5-3.1 mm, and 3.1-4.0 mm, respectively.

The grain of the second fraction was divided into three subsamples. The first two subsamples were placed on the plastic sieves and incubated for two-week periods in the desiccator over a water solution of sulphuric acid. By preparing various concentrations of these solutions, the following moisture contents m_c of grain were obtained: 8.5 and 19.5 %. The third subsample was divided into two groups. The first included the undamaged grain while the second group was divided into four parts, each subjected to the action of quasi-static pressures p of 3, 6, 9 and 12 MPa, respectively. During the measurement of both water absorption rate V and UWL intensity I small working samples of the same mean mass (5.532 ± 0.009 g) were used.

Measurements of the water absorption rate

The study of the V rate by wheat grain was undertaken with the help of the implement presented in Fig. 1. The respective sam-



Fig. 1. The scheme of the implement used for measurements of the water absorption rate by wheat grain. CT - capillary tube, MR - measuring rule, FT - airtight flat tray, FP - filter paper, G - grain, PT - plate tightening the grain layer, AT - adhesive tape, PS - plate scaling the vessel.

ples of grain (G) were placed on an adhesive tape (AT) glued to the down side of the plate (PT) and inserted in the air-tight flat tray (FT). The plate PT was tightening the layer of the grain G to the filter paper (FP) which adhered to the capillary tube (CT) inside the tray FT. Capillary tube CT was 265 mm long, 6.50 mm outside diameter, and 0.462 ± 0.007 mm inside diameter. The tube was calibrated by weighing the mercury contained in a measured length. Rates V of water absorption were calculated from the movement of the meniscus and the diameter of the tube and were expressed as mm³ of water absorbed per minute. Temperatures (T) were controlled at 293 $\pm 1 K$, 303 $\pm 1 K$ or $313 \pm 1 K$ and evaporation was prevented by introducing a water drop into the tube CT near the open end.

Measurements of UWL from wheat grain

Intensity I of UWL was measured using a high-sensitivity single-photon-counting apparatus (Fig. 2) equipped with a 42 mm end-window photomultiplier tube of M12 FQC 52 type (Carl Zeiss Jena, Germany).



Fig. 2. Scheme of the single-photon-counting arrangement for measurements of ultraweak luminescence.

The apparatus detected UWL in the spectral range from 300 to 760 nm with extreme power, equalling less than 10^{-15} W for a wavelength of 450 nm. The geometry of the apparatus is such that the system detects about 55 % of the photons from one side of the flat sample if they are emitted isotropically. In order to minimize noise and background counts the photomultiplier tube was cooled down to 253.0 ±0.5 K. The phototube output was sent to an amplifier and discriminator connected to a pulse counter which registered pulses in one minute periods.

The sample of grain was placed in a single layer in a flat vessel of 6.0 cm in diameter and stored for 1 h in darkness in an optical box. Next, it was inserted under the photomultiplier tube and the intensity I of UWL emitted within 15 min periods after the moment of submerging the grain with 7.5 ml of distilled water was measured. The intensity I was expressed in terms of average counts per min (cpm) and corrected for background counts. The background counts were registered when the sample was absent under the photomultiplier tube. The measurements of UWL from grain were taken at the same temperature as the water absorption rate V. Each result given in the paper is the average of three measurements. The standard errors were less than 0.6 mm³/min and 320 cpm for the measurements of V rate and intensity I, respectively.

RESULTS AND DISCUSSION

The effect of the grain thickness d on both the V rate and intensity I and the relationships between V and I are shown in Fig. 3. As can be seen the rate V (Fig. 3A) and intensity I (Fig. 3B) decrease during swelling time (t) and they had higher values for the samples of grain of smaller thickness d. It is noteworthy that samples of smaller d had taken up the water more quickly than samples of higher d. The most probable explanation is that there V rate depends on total external surface area of all grain in the samples. Assuming that the wheat grain had a spherical shape it was calculated that the total external surface area of grains in the samples of thickness in the range from 2.0 to 2.5 mm and from 2.5 to 3.1 mm was about 1.26 and 1.79 times higher, respectively, than the total external surface area of grains in the samples of thickness in the range from 3.1 to 4.0 mm. The water absorption rate Vand the UWL intensity I from the above mentioned samples was about 1.20 and 1.39 times higher for the V or 1.22 and 1.54 times higher for the I, respectively. The similar increase of V and I suggests that the intensity of UWL from the imbibing grain depends on the water absorption rate. The results of measurements V and I, obtained at the same time t, are compared in Fig. 3C. The data are well estimated by a line plotted as an exponential function of the form: $I=I_0 \exp(KV)$,

where I and K were fitted parameters.

Results presented in Fig. 4A, B, and C show the effects of pressure p on the kinetics of both water absorption rate V and UWL intensity I as well as the relationship between V and I, respectively. The data plotted



Fig. 3. The effect of the grain thickness d on the water absorption rate V(A), intenesity of ultraweak luminescence I(B) and relationships between V and I (C) obtained in the same time t. Measurements were made with undamaged grain of moisture conent $m_c = 10.7 \%$ and temperature T=293 ± 1 K. Δ - 2.0 mm < d < 2.5 mm, o - 2.5 mm < d < 3.2 mm, \bullet - 3.2 mm < d < 4.0 mm.

in Figs 4A and 4B illustrate typical decay of the V and I during swelling time t. It is directly seen that the significantly greater value of V rate and intensity I were obtained for the grain damaged at the higher level of the pressure p. It seems that the higher intensities of UWL from damaged grains are caused by the accelerated process of water absorption as a result of the existence of miscellaneous cracks and scratches in their internal and/or external structures. Hence the intensity I may be representative of the water absorption rate V and it increases together with the increase of mechanical



Fig. 4. The effect of pressure p on the water absorption rate V(A), intensity I of ultraweak luminescence (B) and relationships between V and I (C) obtained at the same time t. Measurements were made with grains of thicksses from 2.5 to 3.1 mm, moisture content $m_c = 10.7 \%$ and temperature $T = 293 \pm 1$ K.

damage to the grain in samples. Comparing the rate V with intensity I, measured for various kinds of samples in the same time t, the relationship presented in Fig. 4.C was obtained. It is directly seen that intensity I clearly increases with the increase of the V rate.

The influence of moisture content m_c of grain on the kinetic of both water absorption rate V and UWL intensity I in the first phase of its imbibition are plotted in Figs 5A and 5B, respectively. As can be seen in Fig. 4A the V rate decreases during swelling time t and depends on the moisture content m_c of the grain. The results presented Next, it declines while the intensity from the grain of $m_c = 18.4$ % constantly increases in the whole period of time t. The difference between kinetic curves of UWL emitted by the grain with lowest moisture content and the remaining ones indicates that intensity I depends on the rate of water penetration into wheat grain. The shape of the kinetic curve obtained for the grain with highest m_c suggests that the UWL may be in part the result of an active transport of water into grain membranes.

A graph expressing relationships between V rate and intensity I for the grain of



Fig. 5. The effect of grain moisture m_c on the water absorption rate V(A), intensity I of ultraweak luminescence (B) and relationships between V and I(C) obtained at the same time t. Measurements were made with undamaged grains of thicknesses from 2.5 mm to 3.1 mm and temperature $293\pm1K$. • $m_c = 18.4\%$, o $-m_c = 10.7\%$, $\Delta - m_c = 8.5\%$.

show that the decay of V rate was much slower for grain of higher moisture content m_c . All the data clearly prove that the V rate of grain with low levels of m_c is higher in comparison to V rate for grain with higher moisture m_c . The effect of m_c on the kinetics of UWL intensity I is shown in Fig. 4B. It is seen that the intensity I quickly increases after the addition of water to investigated samples of grain. The much faster increase and higher value of I occurs for the grain with two lowest levels m_c (8.5 and 10.7 %). various m_c is presented in Fig. 5C. Comparing the results it was concluded that *I* depends on both the *V* rate and m_c which is clearly reflected by two separate curves. It is seen that higher values of intensity *I* response to higher *V* rate were registered at grain $m_c = 8.5 \%$ (upper curve). It proves the supposition that the UWL is connected to the state of water in imbibing grain. As it is known the water penetration into the grain with low level m_c was registered as bounded water (starch-bounded or protein-bounded), [1].

Hence the rate V quickly decreases with the increase of the grain space adsorbed by water. Similar relationships between V and I presented in Figs 3.C and 4.C and by the upper curve in Fig. 5C suggest that UWL emitted in the first phase of imbibing grain of two low level m_c (8.5 and 10.7 %) is the result of sorption processes affecting its structures. The curve presented in the lower part of Fig. 4C reveal another pattern of the relationship between I and V at the grain $m_c = 18.4$ %. Under this condition the grain contain an amount of liquid water hence the emission of UWL were related to the active transport of water through the grain membranes.

The temperature (T) dependence of V rate and intensity I are presented in Figs 6A and 6B, respectively. All the results show typical declines of V and I, which were noted in the earliest part of the work at the grain moisture m_c equal 8.5 or 10.7 %. From these results it is clearly seen that both V and I are increased with the increases of temperature T. The separate curves in Fig. 6C indicate, however, that the processes of water absorption and UWL emitted by wheat grain depend on different manner of temperature T.

CONCLUSIONS

From the data presented in the paper it is evident that the wheat grain quickly took up the water in the first 15 min of its imbibition. The amount of water absorbed by the grain may be successfully estimated using the employment presented in Fig. 1. As it was obtained the V rate of water absorption by grain sharply increases during the first phase of the imbibing process and next declines during swelling time t (Figs 3A, 4A, 5A, and 6A). Comparing the kinetic curves it has been noted, however, that the V rate reached higher values in the samples of smaller thickness d (Fig. 3A) and increased with the growth of pressure p, applied during action of mechanical loads on the sample (Fig. 4A), as well as it increased with the decline of the moisture content m_c (Fig. 5A)

and increase of temperature T (Fig. 6A).

The measurements of intensity *I* of UWL emitted by grain have proved similar influence



Fig. 6. The effect of temperature T on the water absorption rate V(A), intensity 1 of ultraweak luminescence (B), and relationships between V and I (C), obtained at the same time t. Measurements were made with undamaged grains of thicksses from 2.5 to 3.1 mm and moisture content $m_c = 10.7 \%$. • - T=293 K, o - T=303 K, Δ - T=313 K.

of the thickness d (Fig. 3B), pressure p (Fig. 4B), moisture content m_c (Fig. 5B) and temperature T (Fig. 6B), as it was noted for the influence of these parameters on the water absorption rate V. Taking into account these relationships with the shape of kinetic curves expressed dependencies of I on the swelling time t, obtained for the two lower levels of m_c (8.5 and 10.7 %) it seems, however, that the intensity I depends on the V

rate of water absorption by grain.

The results compared in Figs 3C, 4C, 5C, and 6C indicate that the measurements of UWL intensity I at the initial stage of wheat grain swelling may be used to investigate the rapid process of the entry of water into the grain.

REFERENCES

- Aksenov S.J., Askotchenskaya N.A., Golovina Y.A.: The state of water studied in seeds of different qualitative composition and its changes upon the action of temperature (in Russian). Plant Physiol., 24(6), 1251-1260, 1977.
- Campbell J.D., Jones C.R.: The effect of temperature on the rate of penetration of moisture within damped wheat grains. Cereal Chem., 32, 132-139, 1955.
- 3. Campbell J.D., Jones C.R.: The rates of penetra-

tion of moisture to different points in the central cross-sections of the endosperm damped Manitoba wheat grains. Cereal Chem., 34, 110-116, 1957.

- Hinton J.J.C.: Resistance of the testa to entry of water into the wheat kernel. Cereal Chem., 32, 296-306, 1955.
- Jackson G.M., Varriano-Marsten E.: Simple autoradiographic technique for studying diffusion of water into seeds. Plant Physiol., 65, 1229-1230, 1980.
- Jones C.R., Campbell J.D.: Micro-determination of endosperm density as a means of mapping moisture distribution in wheat grains. Cereal Chem., 30, 177-189, 1953.
- Seckinger H.L., Wolf M.J., Dimler R.J.: Micro method for determining moisture distribution in wheat kernels, based on iodine staining. Cereal Chem., 41, 80, 1964.
- Stenvert N.L., Kingswood K.: An autoradiographic demonstration of the penetration of water into wheat during tempering. Cereal Chem., 53, 141-149, 1976.
- Tryka S.: Ultraweak luminescence from mechanically damaged wheat seeds during inhibition. In: Biological Luminescence. (eds): B. Jeżewska-Trzebiatowska, B. Kochel, J. Sławiński, W. Strek. World Scientific Publishing Co., Pte. Ltd., Singapore, 630-646, 1990.
- Tryka S., Koper R.: Application of the capillary method for determination of the water absorption rate by wheat grain at the initial stage of swellig. (in Polish). Rocz. Nauk Roln., 109 (A), 43-51, 1990.