DIGITAL ROENTGENOGRAPHY IS AN INNOVATIVE METHOD OF ASSESSING SEED QUALITY

Evgenia P. Klyuchka[1], Alexander D. Lukyanov[2],
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Abstract: An experimental study, which begins with an evaluation of the quality of seeds by Digital roentgenography, will bring significant savings for the organization, increase the accuracy and reproducibility of research. The choice should be made in the assessment of seed quality. Using the Digital roentgenography, it is possible to determine the tomato seed germination by two approaches. The first one is a percent ratio between the linear seed's dimension (R_{gL} = 45,78 %), and the second one is a percent ratio between the section square seed's dimension (R_{gS} = 49,33 %). Both values have a 10% tolerance.

Key words: Digital roentgenography, seeds, seedlings, assessment of seed quality, tomato

Introduction

Plants and seeds for an experiment are a sophisticated element. The analysis should begin with an assessment of seed quality. A preliminary evaluation will bring significant savings for the organization of scientific research to increase the accuracy and reproducibility of scientific experiments. Standard methods can be morphological, biochemical, luminescent, and others. Conventional methods for assessing the quality of seeds do not cover the entire spectrum of modern requirements. An analysis of the literature showed that radiography is an effective method for determining seed quality (ISTA, 1999).

The advantage of Digital roentgenography (DR) is the use of imaging techniques. The DR preserves the integrity of the seeds in assessing their quality, which allows for further analysis by other methods. The DR enables the use of seeds for new sowing, which is an essential factor in working with small batches of seeds, reveals the extent of injured seeds, the presence of microcracks, and examines the population and damage of seeds by microorganisms. This information is of great importance in industrial practice, in cases where the seeds are stored or shipped to other regions. The DR provides almost the only

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opportunity to detect hidden seed germination (Nikolsky, 2015, Musaev et al., 2016). X-ray diffraction determines defects in the internal structure of seeds and the degree of viability and energy growth. The seeds of vegetables are poorly studied previously by the method of radiography compared with grains. The seeds of vegetable plants have significant differences in biology, morphology, biochemical composition and physiological state of the plant, which form the seeds (Gryaznov, et al., 2017). For these reasons, it requires unique methodological solutions and the organization of analytical processes (Musaev and Bukharov, 2016, Kim et al., 2015).

Thus, DR should be used as a universal imaging method for assessing the impact of various factors on the internal processes of the vegetative development of plants. For example, the influence of water quality on the process of swelling and germination, the effect of light regimes, the impact of various physical fields on the vegetative and root parts of plants (Musaev et al., 2015). The results of the experiments can be used for parametric modeling of an artificial microclimate in closed systems.

The aim of the study is to explore the possibility of using the DR for evaluation of seed quality by identifying the intrinsic attributes of seeds at different stages of development in the process of tomato germination.

Material and methods

The DR was used to identify seeds with a visual assessment of standardized roentgenography of the original seeds and seeds of the next reproduction. The main components of the seeds are high molecular weight organic compounds, consisting mainly of polysaccharides and proteins, which complicates the visualization using X-ray computed tomography. We used correlative radiography with a resolution of about 20 μm in combination with magnetic resonance imaging with a resolution of about 120 μm to create the basis for visualization of germinated seeds. The seeds were grown using LED lighting of a different spectrum. In total, 40 tomato seeds were selected for research. For research, a microfocus radiation source RAP50 was used, having a range of variation of the anode voltage of the X-ray tube 5 – 50 kV, a variety of change of the anode current between 20 – 200 μA.

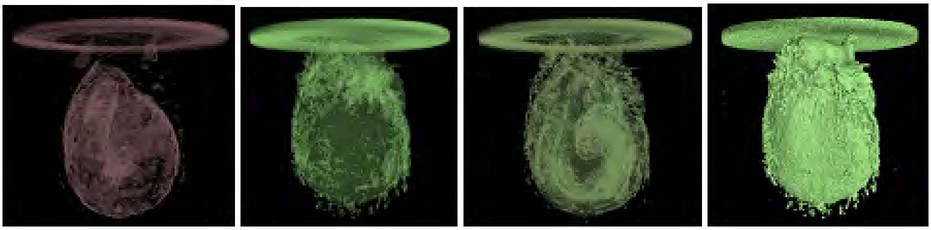
The device Xradia Versa 520 (Carl Zeiss X-ray Microscopy, Inc., Germany) was used in a scientific experiment. The sample was placed in a plastic tube with a small piece of water moistened sponge and sealed with wax. The sample was applied at the lowest possible distance to the X-ray source. X-ray projections of the sample were obtained with a detector based charge-coupled array on the matrix solution 2048×2048 and cooled with Peltier elements to − 59 °C. The photography was done with binning 2; therefore, the resolution of the images, to store projections of up to 1024×1024 pixels. The choice of the filter to X-ray radiation was based on measured values of transmittance by the recommendations of the user's guide Xradia Versa 520. To minimize ring artifacts has been used Dynamic Ring Removal (DRR), which provided a small arbitrary displacement of the sample before taking each next projection. In the process of imaging was done shooting ten backgrounds (reference) image of the air (without a sample between source and detector), which were averaged and applied to each received projection. The remaining parameters are listed in: Optical zoom (lens) 4X; Voltage, 30 kV; Power, 1,8 W; The target current, 58,8 μA; Filter Without a filter; Exposure time, 7 sec; The voxel size, 3.39 μm; The number of projections 1601. The method is standardized ОСТ56-94-88, ИСО6639/4-87. Analysis of seeds was performed by the “Method of Roentgenography in agriculture and crop production” (Musaev et al., 2015).

Results and discussion

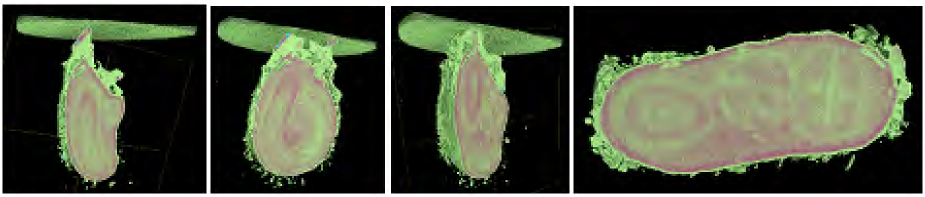
The study presents the regulation of morphogenesis in the growth process of tomato seeds, depending on the conditions of artificial microclimate (Graph. 1.). The physical properties of seeds are characterized by extended sizes and shapes, which are essential indicators of seed quality. The form of the seed indicates the degree of level of viability and energy germination. The type of the seed is also an external indicator of the processes of synthesis, transformation, distribution, and accumulation of organic substances in the embryonic and storage tissues. The specific form of the seed determines the kind and grade of culture and is characterized by the ratio of the linear dimensions: length, width, and thickness. A preliminary conclusion about the quality of the seed is formed by measurement and analysis of linear dimensions of the seeds of some form. The linear dimensions of the seeds are determined with a ruler; the complexity and uncertainty of such a method are apparent.

One of the most significant elements of the integrity of plant organisms is the polarity, that is, the axial or an axial organization of the body in space. Polarity is a specific orientation of activity and morphogenesis of plants in the area. Polarity can be defined as the existence of functionally important asymmetrical structures formed in response to a vectorized external or internal stimulus. Axial (or axial) polarity requires a well-developed longitudinal axis that bears lateral organs: lateral branches, roots, leaves, and flowers. Axial symmetry is the foundation on which to create a specific three-dimensional structure of the organism is orientation in space, the coordination of its functions, and adequate reaction to various stimuli. The axis of polarity appears on the stage of the zygote, forms the embryo and directs the processes of growth and development during the further ontogeny of the plant.

The analysis, which is based on the ratio of the linear dimensions, shape and axial polarity, has revealed the typical defects and anomalies of the internal structure of the seeds (Graph. 2.). For a full analysis of seed quality recording of abnormalities and weaknesses of the internal structure of seeds is not enough, it will be essential to determine the extent of their influence on viability by germination of seeds in the future. This approach addresses methods of digital phenotyping that are not considered here.



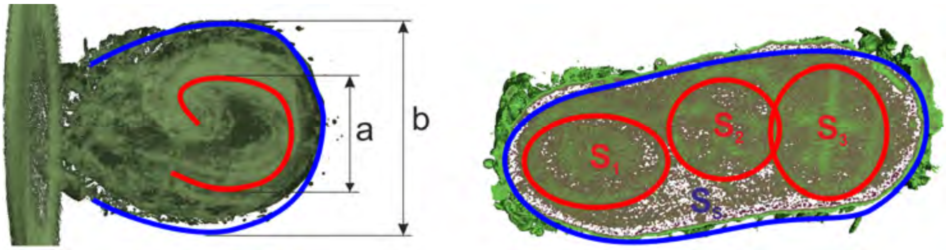
Graf. 1. Spoljašnja struktura semena paradajza
Graph. 1. The external structure of the seed tomato



Graf. 2. Unutrašnja struktura semenki paradajza u fazi bubrenja
(3 dana tretiranja na veštačkom osvetljenju na 2000 Lux i temperaturi od 25 °C)
Graph. 2. The internal structure of tomato seed in the stage of swelling
(3 days of training at artificial illumination at 2000 Lux and temperature of 25 °C)

Traditional roentgenography is based on the collection of information focused on the film, which is both a detector image, the carrier image (memory), and virtual material archiving. The advantage of DR is the possibility of separation processes get the picture into stages, each of which can be individually optimized. These steps are the detection of spatial images; image processing, image recording; the image and the browsing; archiving. The first phase of the research involves creating a Bank of reference images. The difficulty that we are facing towards creating a Bank of reference images, associated with the notion of ontogenesis. Ontogeny traditionally involves the development of an individual organism, while the general patterns of morphogenesis refer to the formation and differentiation of organs and tissues. The problem is to separate the unique characteristics of the seeds and emphasize the morphogenesis of specific cultures and varieties. Therefore, the qualitative result of the study should be based on a large sample of events. A computer program automatically holds must process the images to identify the types of latent defects of the seed, to select seeds with different types of errors, and conducts automatic reporting and report generation in a separate file. The main disadvantage of currently developed computer programs is the lack of versatility.

Using the DR images, we can determine seeds germination faster, than the visual view. The Graph. 3. demonstrate two approaches for determining a seed germination ratio.



Graf. 3. Dva pristupa za izračunavanje stepena klijanja semena paradajza
Graph. 3. Two approaches for calculation a degree of seed tomato germination

The first approach lies in calculating a percent ratio R_{gL} between seed's germ dimension a and a linear seed's dimension b ($R_{gL}=\frac{b-a}{b}\cdot 100\ \%$). The second approach lies in calculating a percent ratio R_{gS} between germ section square $S_1 + S_2 + S_3$ a and a whole seed's section square S_s ($R_{gS}=\frac{S_s-(S_1+S_2+S_3)}{S_s}\cdot 100\ \%$).

All values for both approaches can be easily calculated automatically (Table 1.):

Tabela 1. Procentualni odnos R_{gL} i R_{gS}
Table 1. The percent ratio R_{gL} and R_{gS}

Linearni pristup *Linear approach*			Kvadratni pristup *Square approach*		
		R_{gL} (%)	S_1	0,67 mm²	*R_{gS} (%)*
a	0,90 mm	45,78	S_2	0,50 mm²	49,33
b	1,66 mm		S_3	0,72 mm²	
			S_s	3,73 mm²	

Both values (*R_{gL} and R_{gS}*) have a 10 % tolerance, but the square approach is slightly better for program realization.

Conclusion

Its digital radiography method can guarantee the high-quality hybrid seeds without a single flawed internal structure of the seed. The technique makes it possible to determine the presence of mechanical trauma, a hidden population, and damage by insects, inner sprouting, and other defects and anomalies of the internal structure of seeds shown on the various types in varying degrees of intensity (Klyuchka et al., 2019).

Using the Digital roentgenography, it is possible to determine the tomato seed germination by two approaches. The first approach is a percent ratio between the linear seed's dimension (R_{gL} = 45,78 %), and the second approach, more appropriate, is a percent ratio between the section square seed's dimension (R_{gS} = 49,33 %). Both values have a 10% tolerance.

Further evolution of the method can rapid assessment result in a high-speed X-ray separation of the seeds for which will further the maintenance of a data Bank for X-ray signs of vegetable seeds. With growing applicability due to reduced costs and increased computer processing capabilities images, 3-D X-ray imaging of the seeds will find its way not only in seed laboratories but also for modeling and design of various systems with artificial microclimate. As a result, their commercial value will only increase. It is expected that the methods and treatments using the technology of three-dimensional Roentgenography in the future will become part of the certification and standardization of seeds in regulations or guidance ISTA.

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DIGITALNA RENTGENOGRAFIJA JE INOVATIVNA METODA PROCENE KVALITETA SEMENA

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Izvod: Eksperimentalna studija, koja započinje ocenom kvaliteta semena Digitalnom roentgenografijom, doneće značajne uštede za organizaciju istraživanja, povećaće tačnost i obnovljivost naučnih eksperimenata. Digitalnom roentgenografijom moguće je utvrditi klijavost semenki paradajza pomoću dva pristupa. Prvi je procentualni odnos između linearne dimenzije semena (R_{gL} = 45,78%), a drugi je procentualni odnos između kvadratne dimenzije (površine) semena (R_{gS} = 49,33%). Obe vrednosti imaju 10% tolerancije.

Ključne reči: Digitalna rentgenografija, seme, sadnica, procena kvaliteta semena, paradajz

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U n i v e r z i t e t u K r a g u j e v c u ,
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T e h n i č k i u r e d n i c i

Dr Pavle Mašković, vanr. prof.; Miloš Petrović, istraživač pripravnik;
dipl. inž. Dušan Marković, asistent

T i r a ž : 150 primeraka

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PREDGOVOR

Promene koje se ubrzano dešavaju na globalnom i lokalnom nivou, od naučnih, klimatskih, ekonomskih, pa do političkih, podstiču potrebu da proučimo njihov uticaj na živi svet i na jednu od najvažnijih ljudskih delatnosti - proizvodnju hrane.

Naša poljoprivreda, selo, poljoprivredni proizvođači nisu danas to što su bili pre trideset ili četrdeset godina, srpsko selo se danas više nego ikad ubrzano i u hodu menja. Poljoprivredna nauka mora preuzeti deo odgovornosti u pogledu proizvodnje dovoljne količine kvalitetne hrane za ljudsku ishranu, jer prolaze vremena kada se za svaku lošu žetvu traže opravdanja u klimi.

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