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# DEPENDENCE OF SEEDS GERMINATION OF HEMP INBRED LINES ON GENERATION AND DURATION OF STORAGE

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*One of aspect of inbreeding (self-pollination) influence over proving of depression of signs of germinative energy and germination of seeds of modern hemp (Cannabis sativa L.) varieties is given in this article. It was proved, that germinative energy and germination is reduces due to increasing in the duration of storage of seeds. Germinative energy decreases during the self-pollination from $I_0$ to $I_5$. This regularity is typical for all investigated lines (samples). Substantial variation limits (h is from 1 to 68 and from 2 to 68) point to genotypic features of germinative energy and germination in the lines.*

***Keywords:*** *hemp, inbred lines, germinative energy, germination, depression*

**Formulation of the problem.** Different species of plants have different degrees of manifestation of autbreeding and inbreeding. The extreme degree of manifestation of inbreeding is self-pollination. It is characteristic for a large number of plants. There are biological and genetic barriers to interbreeding between closely related individuals in many species. Genotypes of these species can have self-incompatibility alleles [2]. Inbreeding is used very much in the most allogamy crops in the breeding and agricultural production today.

Inbreeding increases dominant signs and eliminates recessive genes. Due to inbreeding we can find new forms in the populations of allogamy crops which are useful for humanity because of they have recessive signs. Freely crosses populations have such signs in hidden form. The main importance of inbreeding is creation of homozygous generation in a short time. An increasing productivity of plant on the basis of heterosis effect is a result of hybridization of inbred lines [7].

Inbreeding of hemp (Cannabis sativa L.) was investigated by Сизов И.А., Степанов Г.С., Мигаль Н.Д., Лайко И.М., Ситник В.П., Вировец В.Г., Fruwirth C., Hirata K., Hoffmann W., Fleischmann R., Bócsa I., Crescini F., Wichert-Kobus J., Tran Van Lai et al. Their contribution is described in a review paper [5]. These studies were not conducted in full because of cytoplasmic male sterility is not found and inbred lines were not used to create heterotic hybrids. The problem of complex and comprehensive study of biological and selection signs of inbred lines is very important. The problem of studying of dependence of seeds germination of hemp inbred lines on generation and duration of storage is actual too.

**Analysis of last investigations and publications, in which the problem was discussed.** Степанов Г.С. reported about a significant reduction of seed productivity as a result of inbreeding. According to his experimental data the original form had 360-380 seeds per inflorescence and plant $I_1$ –

10–16 (2.7–4.1%), $I_2$ – 3–6 (2.2–3.1%), $I_3$ – 5–6 (1.3–1.5%), $I_4$ – 3–6 (0.8–1.5%), $I_5$ – 2–5 pieces (0.5–1.2%) [6]. He concludes that the phenomenon of reduction germination is a result of inbreeding depression. Already the first generation had 5.5–10.0% of germination and in the future it was lowered to 3.3–5.5% ($I_5$) from generation to generation. Fertility was lost in many cases. Offspring in the fourth and the fifth generations can not be obtained [6]. Research of seed quality, including germinative energy and germination in inbred lines of modern varieties of different origin and maturity groups is a problem today.

Quality seeds have great importance in biological plant growing. It gives possibility don't use additional costs for fertilizers, pesticides, retardants, defoliants. It provides plant growth, reduces or minimizes the negative impact of weeds, diseases, pests and increases crop yield, quality of products, improves the ecological conditions of the field, crop rotation and agricultural landscapes as a whole [1]. Cleanliness, weight of 1000 seeds, moisture content, germinative energy, germination laboratory are the main indicators of seed quality [1].

Number of normally germinated seeds during definite time, which is expressed as a percentage to its total is germination. The laboratory germination is determined by germination under optimal conditions during period which is individual for each culture. The field germination is lower than the laboratory, because of the field has worst conditions for seed germination [4]. Germinative energy is being established simultaneously with germination. Number of normally germinated seeds for the first 3–4 days of germination is germinative energy. This indicator is characteristic feature for seed ability to give good shoots in field conditions. The good shoots provide high survival of plants during the growing season [4].

**The aim of research** is detection of the impact of extreme form of inbreeding (self-pollination) to reproductive ability of hemp, including germination and germinative energy.

**Problems of research** are:

– to identify the relationship of sowing qualities of seeds of inbred lines of modern varieties of hemp various ecological and geographical origin on the duration of storage and generation,

– to discover presence of genotypic conditionality studied traits.

**Materials and methods of research.** The study was conducted from 2007 to 2012 on the basis of Research Station of Bast Crops of the Institute of Agriculture North-East NAAS of Ukraine. The plants of monoecious hemp varieties of Srednerussky eco-geographic type Glukhivski 58 (Victoria), Hlyana, Hlesiya and the South type Zolotoniski 15 were self-fertilization. It was done in a glass house. We used individual isolators which were made from agrovolokno. Clean seeds were stored in paper packages in the laboratory conditions. Indicators of quality seeds of 2008–2012 were determined in 2012 in a month after harvest (hemp seeds germinate without dormancy period).

The seed was germinated by 20°C of temperature on filter paper. Germinative energy was determined after the third day. Germination was determined after the seventh day according to recommendations [4]. The study was conducted in twofold repetition (100 pcs. seeds). The study of individual inbred lines was performed in a single repetition (3 lines). Statistical processing of the data were performed according to methods of field experiments [3].

**Results.** Germinative energy and germination is reduces due to increasing in the duration of storage of seeds (Table 1). This feature is characteristic for all the studied varieties and inbred lines without exception. For example, the germinative energy of seeds variety Glukhivski 58 (harvest in 2008) is 1, 2009 – 36, 2010 – 78, 2011 – 80, 2012 – 81% and the germination is 4, 42, 81, 82, 92% respectively. Germinative energy of seeds variety Zolotoniski 15 (harvest in 2008) is 1, 2009 – 51, 2010 – 58, 2011 – 85, 2012 – 86% and the germination is 2, 60, 78, 92, 93% respectively. Germinative energy and germination in $I_1$ Glukhivski 58 in the different years are 0 and 3, 26 and 28, 77 and 80, 64 and 81, 76 and 90% and in I1 Zolotoniski 15 are 0 and 1, 46 and 56, 50 and 72 81 and 82, 84 and 85% respectively. Germination of seeds is very sharply reduced after 3 years of storage. Seeds lost germination in 4 years under normal storage.

***1. Dependence of germinative energy and seed germination (%) of hemp inbred lines from generation and storage period (2008-2012)***

| Variety, line | Year | | | | |
|---|---|---|---|---|---|
| | 2008 | 2009 | 2010 | 2011 | 2012 |
| Glukhivski 58 | 1/4 | 36/42 | 78/81 | 80/82 | 81/92 |
| $I_1$ Glukhivski 58 | 0/3 | 26/28 | 77/80 | 64/81 | 76/90 |
| $I_2$ Glukhivski 58 | – | 22/33 | 60/80 | 62/80 | 75/84 |
| $I_3$ Glukhivski 58 | – | – | 50/70 | 52/81 | 70/85 |
| $I_4$ Glukhivski 58 | – | – | – | 51/70 | 65/70 |
| $I_5$ Glukhivski 58 | – | – | – | – | 57/77 |
| Hlyana | – | – | – | – | 94/99 |
| $I_1$ Hlyana | – | – | – | – | 93/99 |
| Hlesiya | – | – | 55/67 | 88/95 | 91/100 |
| $I_1$ Hlesiya | – | – | 50/60 | 86/92 | 97/97 |
| $I_2$ Hlesiya | – | – | – | 80/95 | 87/95 |
| $I_3$ Hlesiya | – | – | – | – | 70/70 |
| Zolotoniski 15 | 1/2 | 51/60 | 58/78 | 85/92 | 86/93 |
| $I_1$ Zolotoniski 15 | 0/1 | 46/56 | 50/72 | 81/82 | 84/85 |
| $I_2$ Zolotoniski 15 | – | 20/21 | 50/70 | 51/75 | 80/80 |
| $I_3$ Zolotoniski 15 | – | – | 49/68 | 49/80 | 79/79 |
| $I_4$ Zolotoniski 15 | – | – | – | 46/79 | 79/85 |
| $I_5$ Zolotoniski 15 | – | – | – | – | 78/80 |

Note. Numerator – germinative energy, denominator – germination.

Self-pollination is cause for declining of germinative energy. This regularity is typical for all investigated variants. This sign decreases in the population of variety Glukhivski 58 from 36 to 22% in $I_2$ (2009), from 78 to 50% in $I_3$ (2010), from 80 to 51% in $I_4$ (2011), from 81 to 57% in $I_5$ (2012), in a variety Hlesiya from 55 to 50% in $I_1$ (2010), from 88 to 80% in $I_2$ (2011), from 91 to 70% in $I_3$ (2012) and in the population variety Zolotoniski 15 from 51 to 20% in $I_2$ (2009), from 58 to 49% in $I_3$ (2010), from 85 to 46% in $I_4$ (2011), from 86 to 78% in $I_5$ (2012). There is intsuht-depression in this case. Intsuht-depression consists in reducing power seed embryo development, the ability to germinate, which is a consequence of suppressed formation of seeds after pollination and fertilization, including the development of the endosperm.

Germination of seeds reduces only in some variants. Clear regularity isn't observed. We can propose the following hypothesis. Field germination and features of shoots of inbred lines will be worse because of the changing of grade germinative energy. This will have to take into account in the breeding.

The equation of linear regression accoding to the presence of a clear dependence of the studied signs in interval measurement scales and normal distribution of values was constructed by us (Figure 1). The equation of linear regression is used for predicting of values of signs in a generation of inbred lines.

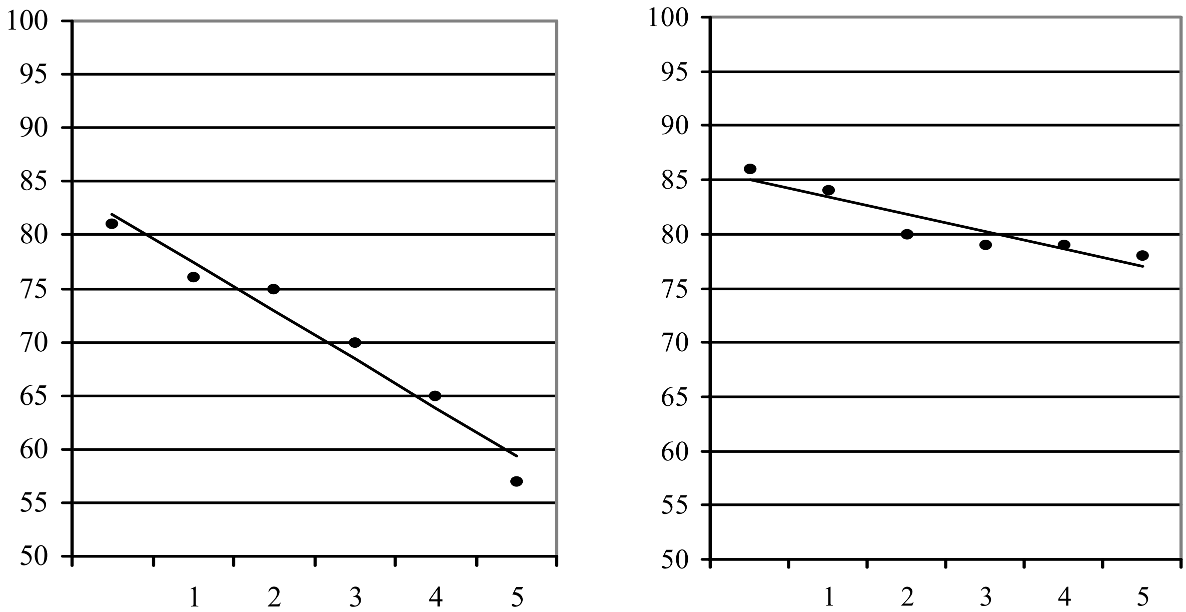


***Figure 1. Dependence of germinative energy of hemp seeds from generation of inbred line (left – for a lines of variety Glukhivski 58, the equation of linear regression is Y = 81.953 – 4.514X, right – for the lines of variety Zolotoniski 15, the equation of linear regression is Y = 85.000 – 1.600X )***

The limits of variation of signs of germinative energy and germination of seeds of different families inbred lines were analyzed by us too (Table 2). Substantial variation limits (h is from 1 to 68 and from 2 to 68) point to genotypic features of germinative energy and germination in the lines (samples).

***2. The limits of variation of signs of germinative energy and germination of seeds (%) of different***

***families of hemp inbred lines***

| Year | Line | Germinative energy | | Germination | |
|---|---|---|---|---|---|
| | | Min–Max | h | Min–Max | h |
| 2008 | $I_1$ Glukhivski 58 | 0–1 | 1 | 2–9 | 7 |
| | $I_1$ Zolotoniski 15 | 0–1 | 1 | 0–2 | 2 |
| 2009 | $I_1$ Glukhivski 58 | 9–77 | 68 | 10–78 | 68 |
| | $I_2$ Glukhivski 58 | 1–57 | 56 | 3–63 | 60 |
| | $I_1$ Zolotoniski 15 | 1–39 | 38 | 1–40 | 39 |
| | $I_2$ Zolotoniski 15 | 1–52 | 51 | 1–54 | 53 |
| 2012 | $I_1$ Glukhivski 58 | 52–84 | 32 | 83–100 | 17 |
| | $I_2$ Glukhivski 58 | 50–76 | 26 | 59–93 | 34 |
| | $I_3$ Glukhivski 58 | 60–90 | 30 | 60–90 | 30 |
| | $I_4$ Glukhivski 58 | 60–70 | 10 | 70–75 | 5 |
| | $I_5$ Glukhivski 58 | 30–70 | 40 | 60–90 | 30 |
| | $I_1$ Hlesiya | 90–100 | 10 | 98–100 | 2 |
| | $I_2$ Hlesiya | 59–97 | 38 | 50–100 | 50 |
| | $I_3$ Hlesiya | 50–90 | 40 | 50–100 | 50 |
| | $I_1$ Zolotoniski 15 | 75–90 | 15 | 90–97 | 7 |
| | $I_2$ Zolotoniski 15 | 75–86 | 11 | 78–88 | 10 |
| | $I_3$ Zolotoniski 15 | 61–90 | 29 | 64–100 | 36 |
| | $I_4$ Zolotoniski 15 | 75–90 | 15 | 80–90 | 10 |
| | $I_5$ Zolotoniski 15 | 77–80 | 3 | 79–81 | 2 |

**Conclusions.** 1. Germinative energy and germination is reduces due to increasing in the duration of storage of seeds. 2. Germinative energy decreases during the self-pollination from I0 to $I_5$. This regularity is typical for all investigated lines (samples). 3. Substantial variation limits (h is from 1 to 68 and from 2 to 68) point to genotypic features of germinative energy and germination in the lines.

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