EFFECT OF GERMINATION ON HEMP (*CANNABIS SATIVA L.*) SEED COMPOSITION

— research paper —

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Abstract: This study investigates the quantitative transformation that take place in hemp (*Cannabis sativa L.*) seeds during germination: dry matter, ash, protein, lipid, fiber and carbohydrates, the Germinative Energy, the Germinative Capacity over eight days of germination. The results show that different components of the seed undergo transformations during germination. Germ grows by accumulating proteins and fiber and by consuming lipids and carbohydrates. Weight loss occurs.

Keywords*:* seed, germination, Cannabis sativa L.

INTRODUCTION

Germination is a complex biochemical process which leads to profound transformation in the qualitative and quantitative composition of hemp (*Cannabis sativa L.)* seeds (Mediavilla et al, 1998). As a result of reserve compound mobilization and intense biosynthesis that takes places when the new plant is formed, a significant accumulation of active biological compounds takes place (Turner et al., 2006), influencing positively cellular activity and its equilibrium.
Seed germination is a way of seed processing that leads to an important growth of their nutritive value due to the increase of nutritive compounds bioavailability, but also due to the increase of the bio active compounds content (Urbano et al., 2005) (Harmuth-Hoene et al., 1987). Previous researches of the authors (Albu et al., 2008) studied the effect of germination

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on seed minerals absorption in animal organism, **t**he results indicating a normalization of the parameter value showing that germinated seeds consumption ameliorates serum calcium, magnesium and iron values.
In this research, the effects of germination on some important characteristics of hemp: Germinative Energy, Germinative Capacity, dry matter, ash, protein, lipid, fiber and carbohydrates during eight days of germination are investigated.

MATERIALS AND METHODS

Germinating the seeds

For germination the seeds have been soaked 30 min in a disinfectant solution (0.7 g/l sodium hypochlorite in water) to prevent microorganisms contamination. After the seeds have been washed with distilled water they have been soaked for another 5h 30 min in clean distilled water. The operation was carried out in a controlled environment at 20^0C, water having the same temperature. After inhibition the seeds are placed on perforated plastic trays in the germination chamber. The process takes place in a controlled temperature, humidity and light environment.
Seeds are sprayed with distilled water every 4h with a programmable electronic device. The tested samples were germinated for 0, 48, 72, 96, 144 and 192 h.

Germinative Energy and the Germinative Capacity

For Germinative Energy and the Germinative Capacity four successive determinations were made. The final result is the arithmetic mean of those values.

Germination losses

Dry matter (DM) determination was made trough the gravimetric method. The samples were dried until constant mass at 105^0C (at least 24 h) in a drying chamber. Dry matter determination was made initially for the mature seeds. Next step was to germinate 5 samples (100 g of dry mass, each) for 2, 3, 4, 6 respectively 8 days.

Ash determination

Ash was determined according to the official method of the AOAC (Association of Official Analytical Chemists) 942.05 (*).

Two grams from the sample are calcined at 600°C porcelain caster for over two hours. The caster is passed to an exicator, cooled down and immediately weighted.

Lipid determination

Lipids were determined according to the official method of the AOAC (Association of Official Analytical Chemists) 996.06 (**).

Hydrolysis-Extraction is one of the most commonly used method and combines an acid hydrolysis (with boiling HCl 5M using a 1047 Hydrolyzing Unit – Soxtec System Foss Tecator) with a petroleum ether extraction (using a 1043 Extractor Unit – Soxtec System Foss Tecator).

Crude fiber determination

Crude fiber was determined according to the official method of the AOAC (Association of Official Analytical Chemists) 962.09 (***).

This method determines crude fiber which is the organic residue remaining after digesting with 1,25% H_2SO_4 şi 1,25% NaOH. The compounds removed are predominantly protein, sugar, starch, lipids and portions of both the structural carbohydrates and lignin.

Crude protein determination

Proteins were determined according to the official method of the AOAC (Association of Official Analytical Chemists) 954.01. (****)

An estimation of the total protein content is obtained with the Kjeldahl method. The sample is treated with concentrated sulphuric acid in the presence of a catalyst. This leads to the nitrogenous conversion in to ammonium sulphate (the nitrogenous from nitrates are only partial converted). The obtained liquid is cooled down, diluted with distillated water and neutralized with sodium hydroxide so the dissociation of the ammonium sulphate to take place. The ammonia is separated through distillation and determinate through titration. Since most proteins from nature had an 16% N content, the protein content of the sample is obtained multiplying the obtained nitrogenous concentration with 6.25 (correction factor resulting from: 1/16=6,25).

Carbohydrates determination

Carbohydrates determination was obtained through difference as follow:

Carbohydrates = 100 – (proteins + lipids + ash + fiber)

RESULTS AND DISCUSSION

After 72 h of germination, the Germinative Energy has the value 96.5% and the Germinative Capacity of the tested hemp seeds is 98% after 98 h of germination. These data shows that seeds with good germinative properties were used, which is a warranty for the reliability of the following tests results.
In the germination process seeds composition undergoes a complex transformation. On germination, seeds lose from their initial mass. Those loses depend of the seed reserves composition. Carbohydrates and lipids are the principal components of those reserves and they are use consequently as an energy source for respiration and other life maintenance processes. In the cases in which the starch is the main seed reserve germination losses are greater in comparison with those with a high lipid reserve. The explanation resides in the high energetic density of lipids towards starch and there for a smaller quantity is required for the same vital activity.

In figure 1 are presented the dry matter (DM) variation during germination of the *Cannabis sativa* seeds.

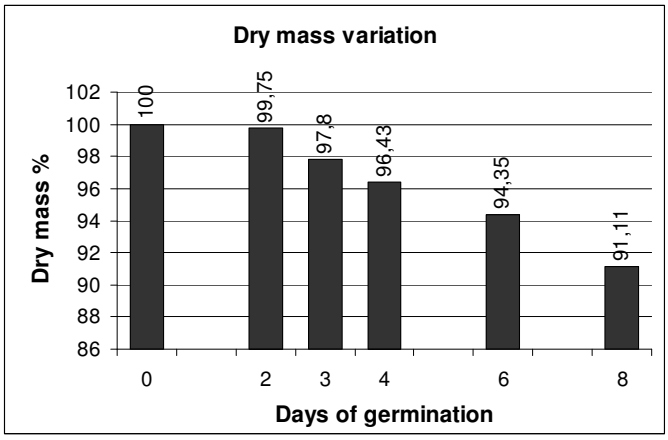

Figure 1. Dry matter (DM) variation during germination of the *Cannabis sativa L.* seeds

The results present the variation occurred during germination in a lot of seeds with an initial 100 grams of DM. Loss of mass during germination are small in the first 3 days and it intensifies from day four. The loss is tightly connected with the carbohydrates level in the seed. Seeds rich in

carbohydrates score high losses during germination as oppose to those rich in lipids.

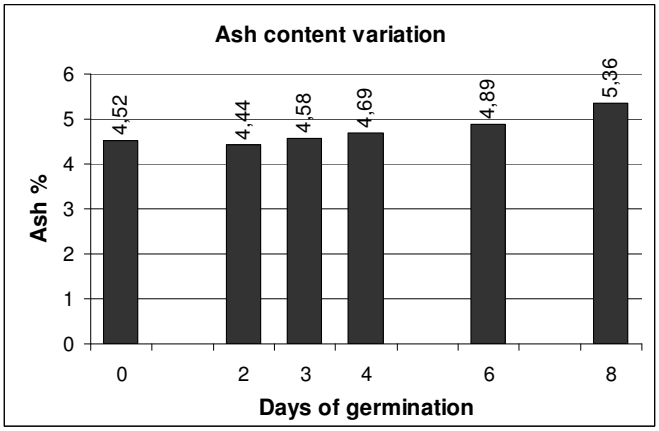

Figure 2. Ash variation during germination of the *Cannabis sativa L.* seeds

As observed in Figure 2, ash has a slight growth tendency upon germination. This change can be mainly caused by the initial alteration of the seeds component proportion.

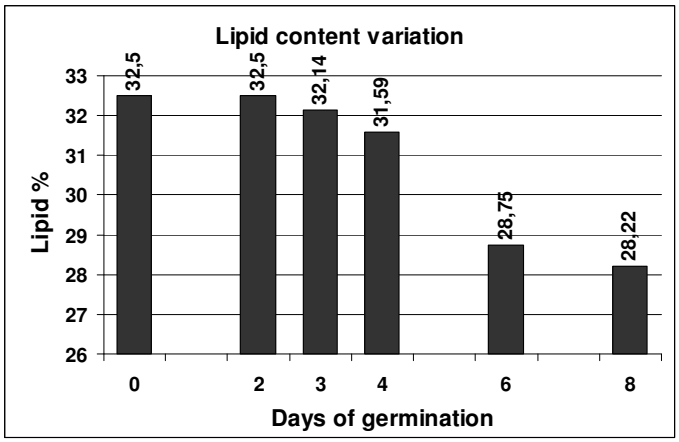

Figure 3. Lipid variation during germination of the *Cannabis sativa L.* seeds

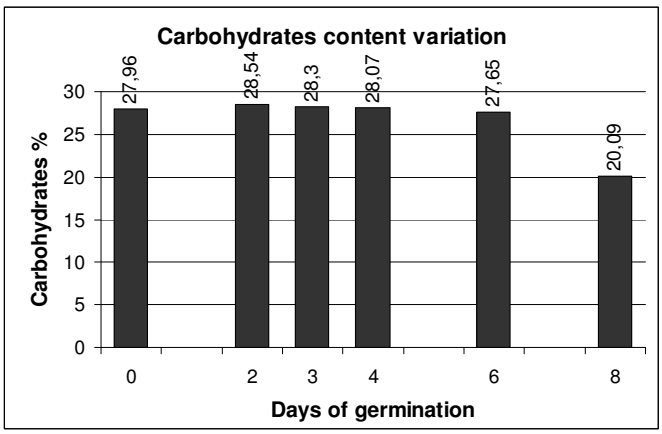

Figure 4. Carbohydrate variation during germination of the *Cannabis sativa L.* seeds

The lipid and carbohydrates content (Figure 3 respectively Figure 4) tends to run down especially in the final part of the germination. The catabolic component is defining in their cases.

The protein content (Figure 5) grows significantly during the germination especially in the final part of the process. This is explained mainly trough the initial alteration of the seeds component proportion (the run down of the seeds reserve), than trough the *de novo* synthesis.

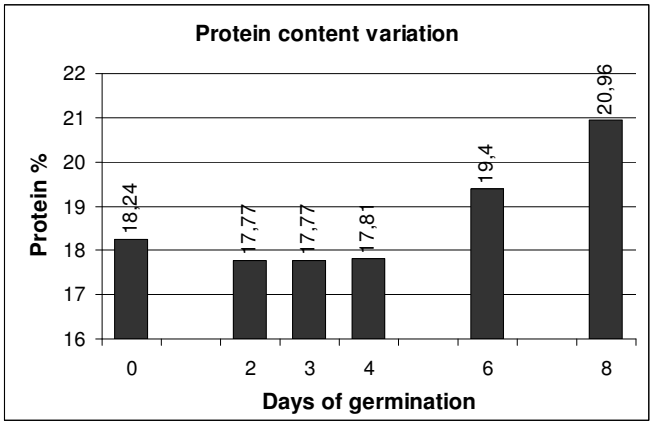

Figure 5. Protein variation during germination of the *Cannabis sativa L.* seeds

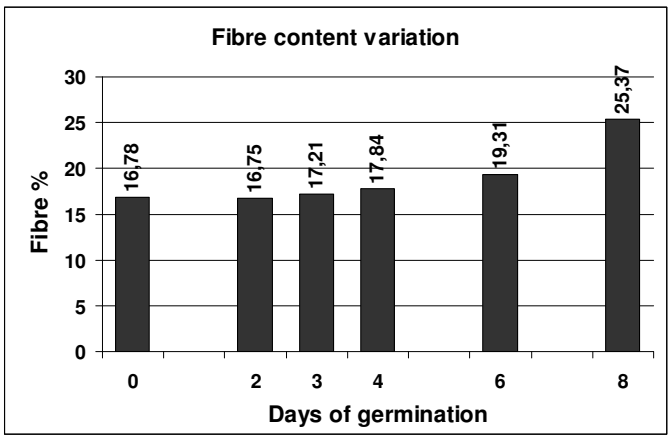

Figure 6. Fiber variation during germination of the *Cannabis sativa L.* seeds

The fiber content (Figure 6) grows significantly during the eight days of germination especially in the final part of the process. This behavior is explaining trough new cell formation. The nuts shells were not removed for those experiments.

CONCLUSIONS

All components present in the seed under enzyme action are put trough a complex degradation and re-synthesis process.
During germination of hemp seeds, weight loss occurs, principally due to the use of lipids and carbohydrates in the respiratory process. The increase of the mineral content is apparent, as result of the alteration of the seeds component proportion.
The lipid content of seeds undergoes important quantitative and qualitative modifications. During germination of *Cannabis sativa L.* the lipid content decreases.
But the increase of proteins and fibers content is real and reflects the reconstruction of germ. Fiber content increases due to the germination period (8 days) especially in the last few days. Fiber quantity resulted mainly because of new cell formation these being the main constituents of cellular walls (cellulose, lignin and hemicelluloses). Also the protein content increases in Cannabis sativa L. seed on germination. The polypeptidic pattern undergoes important transformations with a positive effect upon the biological qualities of proteins and upon their nutritive efficiency.

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