

## Dodders (*Cuscuta spp.*): skin repose, seed germination and pre-parasitic life

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**Abstract** Lately in Romania dodders have been the most dangerous parasites on perennial forage bean crops (alfalfa, clover, bird's foot, seeded lawns with bean majority, etc.)

Although vegetatively propagated with fragments of stems, a broader way of spreading is accomplished by means of seeds, which retain their germination capacity for a duration of up to 40 years in storage spaces; and of up to 6-8 years in soil, plant debris or manure.

Dodders offer many curiosities and numerous features which can explain the adaptation of the plant to parasitism, such as: an enormous fertility capacity (up to 10000 seeds per plant); a long period for search of host plants for subsistence; a long germination ability, covering several time periods due to the phenomenon of skin inhibition, etc.

### Key words

Dodder, seed germination, pre-parasitism

### Tegumentary repose, seed germination and pre-parasitic life

The dodder, a parasitic and quarantine plant, induces significant damages on clover cultures and grasslands, attacking them at a frequency and degree of between 10 and 80%. In some cases it damages the crops completely, thus infringing on the reevaluation of crop technologies, which have to be in accordance with the requirements of improvement of natural resources in the context of a sustainable development [8].

Among some of the most significant features of the dodder, let us mention the following ones: lack of a cotyledon; presence of the chlorophyll in all organs, except for the root; lack of a meristem and of a root caul, which determine a solely upward migration of assimilates; endurance of biotic haustoria inside the host plant while the parasite's stem is already dead; haustoria that are able to regenerate the parasitical plant, thus rendering it perennial; etc.

The germination process takes place in April, May or even in later months, given that certain optimal warmth and moisture conditions are fulfilled. It is even possible for it to occur between May and October, therefore covering the entire vegetative period, but only for those seeds in the upper level of the ground or which are located in a depth of up to 10 centimeters [12].

The optimal germination temperature is between 15 and 20 degrees Celsius for small-seeded dodders (*C. trifolii*, *C. epithymum*) and around 20-30 degrees Celsius for dodders with relatively bigger seeds (*C. campestris*). This might thus explain the fact that the former sprout earlier (April-May) than the latter (June-July) [6].

If the growth conditions are favorable, dodders can germinate even if the seeds are contained in the fruit.

The tegumentary repose, an important feature of dodder seeds, is determined by the water imperviousness of the seminal tegument [13].

The interruption of the tegumentary repose has hardly been researched on. However, it is well-known that it can be interrupted by the passing of seeds through the alimentary duct of animals and birds; these representing as well vectors in the spreading of the parasite, either directly, or by means of farmyard manure.

Likewise, such factors as cold weather, certain bacteria and fungi can interrupt the tegumentary repose of dodder seeds. It is well-known that seeds can stay in the soil and keep their germination ability for a duration of up to 10 years [12]; or even longer (between 10 and 40 years) in the case of those seeds that are preserved under low humidity conditions [14].

Under laboratory conditions dodder seeds that have been treated with enriched sulfuric acid boast very rapidly if they are placed on a wet surface. During this stage one can notice in the middle of the reserve tissue, which had meanwhile become more or less translucent, a filiform and spiraled embryo, lacking a cotyledon. The embryo is the starting point of a rapid growth. The root is the first one to show, like a bulge, and slightly embedded in the soil, up to a depth of between one and three centimeters. Especially in this early phase, the plant is extremely sensitive to drought. It has been proved effectively that a drought period happening during the first growth stages of the plant (1-12 days) results in the necrosis of the plant. If any drought occurs during later stages of development, there will be a slight dehydration of the root, but the ulterior progress and the attachment to the host plant will not be influenced.

Any possible cold period (four-five degrees Celsius) occurring during the germination process would only determine a growth deceleration. Different climate conditions as well as the different length of the humid period explain the quite significant variations regarding the time of sprouting [14].

The stem of the dodder is the next one to develop after the root; and it represents an important elongation. If the attachment to the host plant is delayed for any reason, the root starts to necrose, and it dies after approximately one month.

During some experiments carried out in the field, an artificial infestation of an alfalfa culture with dodder seeds and filaments was attempted, but with no result. For this reason, the germinative ability of dodder seeds was tested thereafter under laboratory conditions.

## Material and Method

Vegetal material: dodder seeds gathered within the Sibiu County from several host plants (alfalfa, clover, willow). The tegumentary repose was interrupted by means of dipping the seeds into enriched sulfuric acid (96%; d=1,83) and agitating them. Different dipping durations were used, starting with five minutes for the first sample, and reaching 35 minutes for the last sample. As a next step, the seeds were washed thoroughly with distilled water and placed for germination onto filter paper at temperatures of 20, 22 and 24 degrees Celsius.

A further experiment was carried out with dodder seeds whose tegument had been rubbed against abrasive paper for 20 minutes. Afterwards, the seeds were put into envelopes which had been wet two hours in advance.

## Obtained results

Table 1

**Germination of dodder seeds after their dipping into enriched sulfuric acid**

Germination temperature	Germination percentage after the dipping of seeds into enriched H <sub>2</sub> SO <sub>4</sub> / different duration							
	0 min.	5 min.	10 min.	15 min.	20 min.	25 min.	30 min.	35 min.
20°C	-	-	-	54	53	65	-	-
22°C	-	-	-	63	72	68	-	-
24°C	-	-	-	72	70	67	-	-

The dodders in sulfuric acid germinated after approximately seven days, the best sprouting being recorded in the case of those seeds which were in enriched sulfuric acid for 15, 20 and 25 minutes. Those seeds dipped for less than 15 minutes did not have their tegument cut through, so the sprouting did not occur due to the tegument's water tightness, whereas the seeds which were in H<sub>2</sub>SO<sub>4</sub> for more than 25 minutes did not germinate either. In this case it might be assumed that the viable elements of the embryo were destroyed, thus rendering the sprouting of a new plant impossible.

In order to bring forth the sprouting process, qualitative filter paper was employed as a germination layer. The method used was the so-called B. P. technique (between paper). The seeds were put between paper layers and then placed on trays and bolts, and then finally introduced into germination machines.

From the filter paper several four-layered envelopes were created, two layers being under the seeds, and the other two layers covering them; and the margins were folded inwardly so as to avoid any seed loss.

Water was being kept in the germination layer by introducing the samples into plastic bags. Before being put into the germination machine, the envelopes containing those seeds whose teguments had been chafed against abrasive paper, were deposited in a refrigerator at a temperature of 5-10 degrees Celsius, for a duration of seven days. Low temperature acts on

seed coatings, increasing their water and gas perviousness. After seven days the samples were thus placed in the thermostat, at a temperature of 20 degrees Celsius. After ten days the percentage of germinated seeds was 12%, and after 20 days, 23% of the seeds had already sprouted.

Dodder seeds are therefore characterized by a strong inhibition [13], which can be disturbed under laboratory conditions, by means of corrosion with enriched sulfuric acid [4]; [9].

The germinative ability is indicative of the number of pure seeds – represented in percentages – capable of producing normal sprouts during a certain time-frame and under the optimal conditions, pre-established for every species individually.

Dodder seeds germinate in soil within a period between two and nine days, and at a temperature of between 10 and 25 degrees Celsius; in Romania this temperature corresponding to the month of May. Small dodders (*C. trifolii*) sprout in approximately seven-nine days at a temperature of 11-19 degrees Celsius. Average-sized seeds (*C. campestris*) germinate in six-seven days at a temperature of 19-23 degrees Celsius, whereas the bigger species sprout at a temperature of 22-26 degrees within a time frame of between two and four days. The germinative ability starts decreasing after the sixth year.

It is well-known that the dodder's largest way of spreading is done by means of seeds, which retain their

germination capacity for up to 12-15 or even up to 40 years in storage and up to 6-8 years in soil, plant debris or trash manure. Although it is a quarantine weed, many areas of perennial legumes, especially alfalfa seed is sown with “undoddered” seeds without known origin and biological value.

In 1 kg of alfalfa seed there were found between 70,000 and 150,000 seeds of *Cuscuta* [4]. This large number of seeds reaching the soil germinates in the following proportions: 15-20% in the first two years of vegetation; 30-40% in years 3-4 of vegetation and 10-15% in subsequent years. Due to this mode of germination, an alfalfa crop is affected throughout its entire cropping duration [4]. This tegumentary repose can be considered to be natural hibernation (hibernation class D) according to Baskin and Baskin [3] because it is caused by one or more layers of seminal tegument cells that are impermeable to water [1], [2], [3]. Seeds will remain inactive until a factor renders the coating water permeable. In nature, these factors include fluctuating temperatures, freezing / thawing, passing through the digestive tract of animals, etc.

Hibernation interruption under laboratory conditions is recommended to be done by mechanical or chemical scarification so that the seminal tegument becomes permeable to water and the seeds can germinate under a wide range of environmental conditions. Further treatments, such as the use of light, the use of growth regulators and other chemicals (potassium nitrate (0.2%) as well as thio - urea (0.5 to 3%), or soaking in water for 1-5 minutes to 80°C, are recommended for the discontinuation of the hibernation process in the cases of those seeds where the deterrent factor is the seed embryo itself [15].

The seed hibernation affects the seed germination patterns in natural ecosystems and the persistence of seed crops. It can also change cultural practices for crop plants by delaying germination. Research in this field involves ecological, biochemical and molecular approaches to describe seed hibernation.

## Conclusions

### Phases of the germination process:

a. – imbibition – also called bloating of the seeds – is a physicochemical process, which can take from several minutes to a few hours. The water ingress occurs from the exterior towards the interior, covering the entire surface of the crust. At cell level, water ingresses into the space between the cell walls and then to the storage organelles, which are the first ones to hydrate;

b. – The hydration and activation of enzymes, vitamins as well as growth regulators is supposed to mobilize reserve substances by means of hydrolysis, and to transport them to the growth points, thus systematizing the biochemical transformations that have taken place in this phase, namely: rapid absorption of water by the seed’s colloids; reactivation of the existing enzymatic

system; the respiration providing the necessary energy to the synthesis of enzymes, structural proteins and lipids for chromosomes, nucleoli, membranes and cell walls in the process of growth and development of new cells within the embryo, etc. The intensity of respiration indicates the pace of metabolic processes during this phase;

c. – cell segmentation and growth result in the development of the embryo. The growth occurs in three successive stages: cell enlargement; increase in their number and enhancement of their differentiation degree;

d. – the sprouting of the embryo from its seed is a crucial moment in the germination process; and it takes place after the crust of the seed or even of the fruitlet has been broken through. For most seeds, the sprouting is the effect of embryo development; and the most likely organ to permeate the tegument is the radicle. However, dodders do not have this organ [11];

e. – The process of morphogenesis results in the occurrence and development of those essential structures which show for every seed the species’ capacity of producing a normal plant under favorable soil conditions.

The seeds thus treated and placed onto wet filter paper, at a temperature of 22-24 degrees Celsius, boast rapidly. Consequently, it becomes possible to see the filiform and spiral embryo in the middle of the reserve texture, lacking a cotyledon, and meanwhile become more or less translucent (Fig. 1).

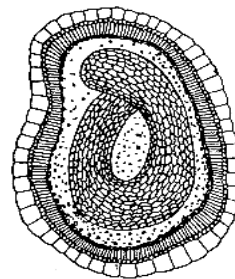


Fig. 1 – Dodder embryo (cf. Rădulescu, 1957)

After consuming part of the reserve texture, the embryo is the starting point of the plant’s development, given optimal temperature and moisture conditions. There is a breach at the level of the micropillar surface; and the tip of the root appears through the aperture. At the top of a long stem there is a hook-like tip, for whose growth and elongation solar light is crucial [10]; [9]. The stem is now characterized by circumnutation movements, which are generated by light and by means of phytochromes [10]; these movements enabling the dodder stem to fix itself onto the host plant. However, if a host plant is not found in due time, the radicle tip starts to mortify. The necrosis then advances towards the stem, and the plant is reduced to a mere fragment of

the former stem, whose length decreases progressively if the necrosis is more rapid than the apical growth. Thus, in the absence of a host plant, the dodder degrades after approximately one month of life.

The young sprout presents itself as a long and thin filament, which ends with a protuberance at its lower end. This bulge is in fact its root, which is very short, thick and with no coleorhiza. It is nonetheless a root characterized by a normal heliotropism and geotropism.

The upper part of the plant starts its growth by means of nutation movements induced by the unequal and variable development of the exterior textures. At its upper end the plant buckles until it forms a completely closed ring, and then it continues its backward course until the stem adopts an almost even trajectory, so as to restart the movement.

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