SOME BIOCHEMICAL ASPECTS DURING BROAD BEAN SEEDS GERMINATION

Conf dr. Marcel AVRAMIUC
Conf.dr. Liviu FĂRTĂIŞ
Universitatea "Ştefan cel Mare" Suceava

Rezumat

Patru eşantioane de seminţe cu capacităţi germinative apropiate valoric, aparţinând unei varietăţi de bob (*Vicia faba* L. var. *major*), au fost studiate în timpul germinaţiei pentru a stabili dacă există vreo legătură între valorile indicilor exsudatului seminal şi activitatea unor enzime. Analizând activitatea catalazei şi a α-amilazei la anumite intervale de timp, pe parcursul a 168 ore de germinaţie, s-a putut stabili că activitatea celor două enzime a fost într-o relaţie indirectă, mai ales cu efluxul transmembranar de zaharuri dacât cu valorile aminoacizilor sau ale conductivităţii electrice.

Résumé

Quatre échantillons de semences avec capacités germinatives ressemblantes, appartenant à une variété de fève (*Vicia faba* L. var. *major*), ont été étudiés pendant la germination pour établir s'il existe une liaison entre les valeurs des indices d' exsudate séminale et l'activité de quelques enzymes. Analysant l'activité de la catalase et de α-amylase aux certains intervalles de temps, en cours de 168 heures de germination, on a pu établir que l'activité de ces deux enzymes a été dans une relation indirecte, surtout avec le flux transmembranaire de sucres qu'avec les valeurs des aminoacides ou de la conductivité électrique.

Abriss

Vier Samenproben von Ackerbohne (*Vicia faba* L. var. *major*) mit annähernden Keimungsfähigkeiten wurden studiert um festzustellen ob eine Verbindung zwischen die Werte dem seminalen Exsudat und die Tätigkeit einigen Enzyme gibt. Es wurden die enzymatischen Tätigkeiten der Katalase und der α-Amylase wägend 168 Keimstunden und wurde festgestellt dass die Tätigkeit den zwei Enzyme ist in einer indirekten Verbindung, besonders mit den transmembranaren Ausfluss des Zuckers als mit den Werten den Aminosäuren oder der elektrischen Leitungsfähigkeit.

Introduction

Not always a high germination capacity is a sure sign of seed structural and functional integrity. The damage of seed can lead to modifications of metabolic process, of content and activity of enzymes involved both in degrading the nutritive reserves and in synthesis of new forming organism components.

Seed structural and functional decline is frequently related to some changes such as: the diminution of germination, the seedling growth rate and the membrane permeability increase (Abdul-Baki, 1969).

The aim of this paper is to evidence some biochemical aspects in seeds belonging to four broad bean samples during germination, and to establish a relationship between the seed exudate indices and the activity of peroxidase and α-amylase.

Materials and methods

Four seed samples belonging to a broad bean variety (*Vicia faba* L. var. *major*) were used as biological material in this work. All samples have represented the crop of the same year, but coming from four different places of Suceava county. Some data, including seed exudate indices (electrical conductivity, sugars and aminoacids content), of the samples are reproduced in the Table 1.

Table 1

Characteristics of the used biological material

Samples	G (%)	WTG (g)	SMC (%)	EC (μS/g)	S (μg/g)	AA (μg/g)
B1	93	1040	12, 45	31,85	263,40	88,25
B2	92	1044	12, 20	52,40	152,85	131,08
B3	95	1042	12, 25	49,17	340,35	113,70
B4	90	1050	12, 50	45,30	145,10	105,35

B1...B4= seed samples; **G**= germination percentage; **WTG** = weight thousand grain; **SMC** = seed moisture content; **EC**= electrical conductivity; **S**= sugars; **AA**= aminoacids

As seen, the four seed samples has close values as to germination percentage, weight thousand grain and seed moisture content. Comparatively, the seed exudate indices have registered higher differences between values especially to sugars content.

The germination percentage was estimated using 4 replicates of 50 seeds for each sample. Petri dishes, with special filter paper, have been used and medium of germination was distilated water. The maximum term of the test evaluation was 7 days (Anghel et al., 1959; Ellis at al., 1985).

The indirect evaluation of the membrane damages was carried out by measuring of electrical conductivity as well as sugar and free aminoacid losses. Conductivity of samples was expressed as micro Siemens/g seed (μS/g seed). For sugars and free aminoacids efflux were used colorimetrical methods, and the results were expressed as micrograms (μg) at 1 g seed (Villegas and Mertz, 1975; Schell, 1980).

The peroxidase activity was investigated, according to a colorimetric method (Artenie and Tănase, 1981), at different times of germination, within 0-168 hours interval, and was expressed as IU/g DM.

The α-amylase activity was evaluated according to Noelting-Bernfeld method (Artenie and Tănase, 1981) and was expressed as μM maltose at 1 g seeds (flour).

Results and discussion

Table 2 includes the peroxidase activity in four broad bean samples during germination.

Along the whole analysed interval, one can see an increasing of the enzime activity once with germination time increasing. The lowest enzyme values were found at 0 and 24 hours of germination, and the highest ones at 96 and 168 hours.

Comparing the four seed samples between them, within each germination time interval, it can find that two samples (B1 and B3) have registered the lowest values, especially after 48, 96 and 168 hours of germination.

Although from one germination interval to another the peroxidase activity evolution was sometimes oscillating, there were two time intervals where the difference between samples was highest, namely at 96 and 168 hours of germination.

Table 2

The mean values of peroxidase activity in broad bean seed samples, at different periods of germination

	Peroxidase activity (IU/g DM)					
Time	**0 h**	**24 h**	**48 h**	**72 h**	**96 h**	**168 h**
Samples						
B1	6,50	11,25	20,38	28,40	32,04	26,50
B2	5,60	9,08	40,59	37,25	46,05	52,36
B3	5,25	8,67	17,07	16,85	28,02	30,68
B4	8,46	10,30	28,74	35,26	56,48	45,00

IU = international units; **DM** = dry matter

The α-amylase activity values are reproduced in the table no. 3.

Table 3

The mean values of α-amylase activity in broad bean seed samples, at different periods of germination

	μM malt / g DM					
Time	**0 h**	**24 h**	**48 h**	**72 h**	**96 h**	**168 h**
Samples						
B1	22,15	20,08	27,19	30,25	68,39	65,15
B2	18,60	26,45	40,82	38,70	65,24	70,39
B3	10,95	20,50	32,40	30,18	58,44	63,09
B4	26,26	18,85	42,26	43,84	52,75	65,23

μM malt= micromoles maltose; **DM**= dry matter

As seen in the table 3, the α-amylase activity has generally had a constant increasing from zero to 168 hours of germination. Except B1 sample, the highest enzyme values were registered after 168 hours.

Like in peroxidase case, the same samples (B1 and B3) has had, comparatively to the other samples, the lowest activity of α-amylase. But here, the biggest differences between above mentioned samples and the other ones were at 48 and 72 hours.

On the other hand, analysing the table 1 it can see some differences between samples mainly as for seed exudate indices.

Thus, if the electrical conductivity and the free aminoacids leakage do not register high differences between samples, the sugars efflux shows the highest values for B1 and B3.

Trying to find a relationship between the two enzyme activity and the seed exudate index values, it seems that the more sugars leakage the lower peroxidase and α-amylase activity is.

It is possible that some structural damages, expressed through increasing of seed exudate index values, to influence the activity of certain enzymes like peroxidase and α-amylase.

Conclusions

1.Analysing the peroxidase and α-amylase activity in four different broad bean seed samples, with close germination capacities, there was found an increasing of enzyme values along the investigated germination interval (0-168 hours).

2.The lowest enzyme activities have been registered by seed samples with highest sugar leakage values.

3.It seems that more than electrical conductivity increasing and aminoacids transmembrane loss, the sugars leakage is a signal of membrane damage leading to seed structural and functional decline.

References

1. Abdul-Baki A.A., 1969 – *Relatioship of glucose metabolism to germinability and vigour in barley and wheat seeds.* Crop Sci., 9.

2. Anghel G., Raianu Maria, Matei C., Bucurescu N., Rădulescu I., Anganu I., Velea C., 1959 - *Determinarea calităţii semînţelor,* Edit. Acad. R.P.R., Bucureşti.

3. Artenie V., Tănase Elvira, 1981 - *Practicum de biochimie generală - Univ. " Al. I. Cuza " Iaşi.*

4. Ellis, R.H., Hong, T.D., Roberts, E.H., 1985 - *Handbook of Seed Technology for Genebanks.* International Board for Plant Genetic Resources, vol. II, Rome.

5. Schell H., 1980 - *Biochimie.* Edit. Did. şi Ped., Bucureşti.

6. Villegas E., Mertz T.E., 1975 – *Simple chemical and biological methods used at Purdue University to evaluate cereals for protein quality.* St. Bull. no. 70, U.S.A.