EFFECT OF SOAKING TIME ON IMBIBITION, WEIGHT LOSS AND RESPIRATION RATES OF SOYBEAN, CORN AND BARLEY SEEDS[1]

I. Rouhani, A. Bassiri and I. Poostchi[2]

ABSTRACT

A comparative study was made to investigate the effect of soaking on percent dry weight loss, imbibition and respiration rates of seeds of soybeans (*Glycine max* L.), corn (*Zea mays* L.) and barley (*Hordeum vulgare* L.) and also to find the respiration rates of seedlings of these species at different stages of germination.

After 4 hours of soaking the amount of water imbibed by soybean, corn and barley seeds were 105, 32 and 23 percent of their original weights, respectively. The percent dry weight loss of seeds was dependent on at least three main factors; the soaking time, the respiration rate and the species.

Manometric measurements of gas exchange showed that the oxygen consumption of seeds reached a maximum of 180, 69 and 56 $\mu l\, O_2$/hr/g fresh weight after six hours of soaking for soybean, barley and corn, respectively. When seeds were germinated and their respiration rates measured at different stages, it was found the maximum oxygen uptake of 1250, 770 and 465 $\mu l\, O_2$/hr/g fresh weight for barley, soybean and corn, respectively, cccurred when radicles had attained a length of 6 to 10 mm and the primary stem was completely green in color.

INTRODUCTION

Germination process is known to require an adequate supply of water, a suitable

1. Joint contribution, Departments of Horticulture and Agronomy, College of Agriculture, Pahlavi University, Shiraz, Iran.
2. Assistant Professor of Horticulture, and Assistant Professor and Professor of Agronomy, respectively.

temperature, favorable amount of gases in the atmosphere and light for certain seeds (2, 7). Kidd and West (3) have shown that conditions of the seed during the early stages of germination could "predetermine" subsequent growth of the plant. Woodstock and Grabe (13) reported that seed metabolism during the first few hours of germination could be related to the rate of seedling growth several days later. This interdependence of seed metabolism and growth due to initial synthetic processes and to the use of energy from respiration reflects a high level of metabolic activity (8, 10, 11).

A seed must reach a certain moisture level before it can germinate. Lyles and Fanning (6), working on the sorghum hybrid RS 610, found that the soil moisture should rise to 25 percent before the emerging radicles become visible. Similarly Clark *et al.* (2) reported that seeds of the non-dormant "Shallu" cultivar of sorghum germinated when the seed moisture rose to 32 to 34 percent.

Imbibition characteristics of seed species during germination have been greatly neglected in the past. The chief component which imbibes water in seeds is protein. Mucillage of various kinds, cellulose and pectic substances also imbibe water to a lesser degree (2). As these substances begin to attract water and cause it to enter through the seed coat the seed starts to swell.

Due to high content of protein, seeds of the legume family imbibe a relatively large amount of water during germination. Gains of 150 to 200 percent of their original dry weight are not uncommon for seeds of this family (4).

In the present investigation attempts were made to find the effects of water imbibition on the percent dry weight loss, moisture content and on the respiration rate of soybean (*Glycine max* L.), corn (*Zea mays* L.) and barley (*Hordeum vulgare* L.) seeds. The respiration rates of seedlings of these species were also compared at different stages of germination.

MATERIALS AND METHODS

Commercial seed lots of corn, soybean and barley of at least 95 percent germination were chosen. Samples consisted of 30 seeds for corn and soybean and 100 for barley. Samples of each of these species were placed in cheesecloth bags, weighed and surface sterilized according to the method described by Larson and Leanga (5).

Bags were then submerged in beakers containing 100 ml of distilled water at 20 C and removed after soaking periods of 0, 2, 4, 6, 8, 10 and 24 hours. The control bags, representing zero soaking period, were not submerged in water. The soaked seeds were immediately dry-blottered, weighed and either incubated at 22 ± 2C on moist filter paper to germinate (three samples for each soaking period of each species) or used for oxygen consumption measurements according to manometric techniques described by Umbreit *et al.* (12) using a Warburg apparatus. To be able to compare relative oxygen consumption of monocotyledonous seedlings (corn and barley) with a dicotyledonous seedling (soybean), three samples at following germination stages were used :

Stage 1 : Seeds visibly swollen; radicle up to 1 mm long; plumule or coleoptile (primary shoot) not apparent.

Stage 2 : Radicle 2 to 3 mm long; primary shoot 2 mm long (except in corn); no greening.

Stage 3 : Radicle 4 to 6 mm long; primary shoot yellowish green.

Stage 4 : Radicle 6 to 10 mm long; primary shoot definitely green.

Stage 5 : Radicle 10 to 25 mm long; appearance of secondary roots; shoot completely green.

Stage 6 : Secondary roots 10 to 20 mm long (except in barley); seed coat separated (soybean).

The dry weight was determined by weighing comparable samples and placing them in a forced draft oven at 102 C for 48 hours. At the end of this period the seeds were reweighed. The difference between the dry weights of non-soaked and soaked seeds of each period was designated as the dry weight loss.

The data on dry weight losses were analyzed according to a split-plot design having species as the main plots and time of soaking as the sub-plots. The treatment means were compared by the use of the Duncan's New Multiple Range Test at 5% probability level. Simple correlations between the measured characters were then calculated on the treatment means.

RESULTS AND DISCUSSIONS

As the soaking time increased, there was an increase in the amount of weight loss in the three species (Table 1). After 24 hours, these losses in soybean, corn and barley seeds were 3.87, 7.10 and 6.13%, respectively.

TABLE 1. Effect of soaking duration on mean percent dry weight loss in grams for soybean, corn and barley seeds.

Soaking duration, hr	Species			Mean
	Soybean	Corn	Barley	
0	3.17	6.67	5.57	5.07c
2	3.27	6.53	5.67	5.16bc
4	3.33	6.73	5.73	5.34abc
6	3.47	6.80	5.77	5.61ab
8	4.13	6.83	5.87	5.50abc
10	3.63	6.97	5.90	5.70a
24	3.87	7.10	6.13	
Mean	3.52c	6.78a	5.80b	

Means followed by the same letter in the marginal row or column are not significantly different at the 5% probability level.

The analysis of variance for the dry weight loss data showed highly significant F values for both species and soaking levels, although their interaction was not significant. This indicates that the three species are different in weight loss during soaking and that the different soaking periods had different effects on the character measured although all species showed similar weight loss at different levels of soaking. These relations are clearly shown in the marginal row and column of Table 1 using Duncan's New Multiple Range Test.

When correlations were calculated between the soaking time and the dry weight loss, the coefficients were highly significant with values of 0.94, 0.89 and 0.95 for soybean, corn and barley, respectively.

Data in Fig. 1 show the amount of water imbibition as percent of dry weight of seeds at different periods of soaking. Corn and barley seemed to have almost the same rate of water absorption while soybean showed higher (about three times as much) imbibition. After 6 hours of soaking, soybean seeds absorbed water equivalent to 112 percent of their original weight while corn and barley seeds absorbed only 31 and 28%, respectively. The amount of water imbibed seemed to be greatly related to the protein content of the seed.

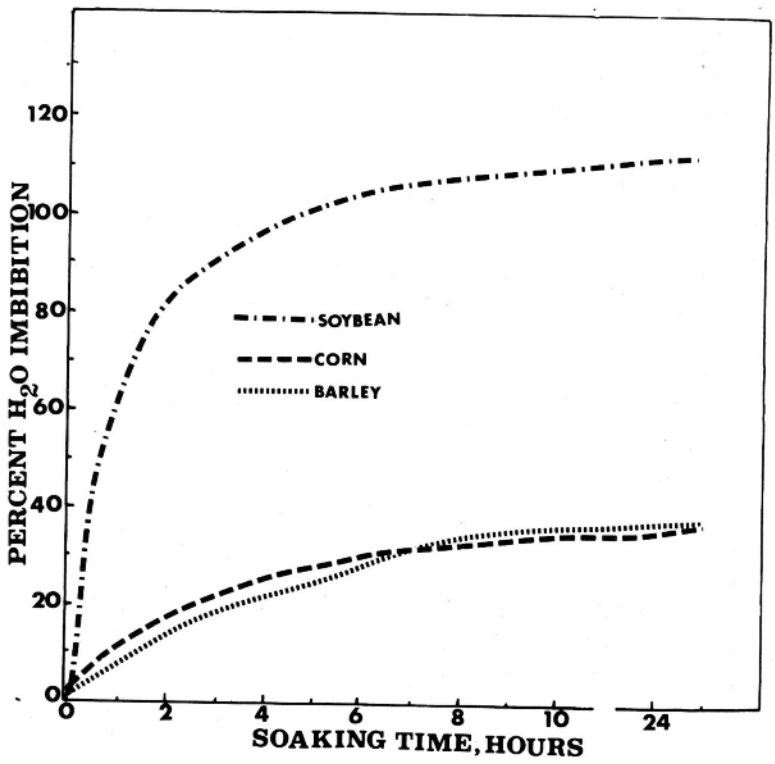

Fig. 1. Percent (of fresh weight) water imbibition of soybean, corn and barley seeds at different soaking periods.

The calculated simple correlation coefficients between the soaking time and the amount of water imbibed were 0.61, 0.76 and 0.84 for soybeans, corn and barley, respectively. These values were statistically significant for corn and barley but not for soybean. This probably is due to the fact that the relationship between soaking period and percent water imbibition was almost linear for corn and barley and not so for soybean (Fig. 1).

The rate of imbibition of soybean seed was 3 to 5 times faster than corn and barley seeds during the first two hours of soaking. Soybean seed imbibed 85 to 90 percent water during the first 2 to 4 hours, while corn and barley seeds imbibed a similar amount of water after 8 to 10 hours.

The respiration rates of seeds, measured in $\mu l\, O_2$/hr/g fresh weight as influenced by different soaking times, are presented in Fig. 2. It can be seen that there was a gradual

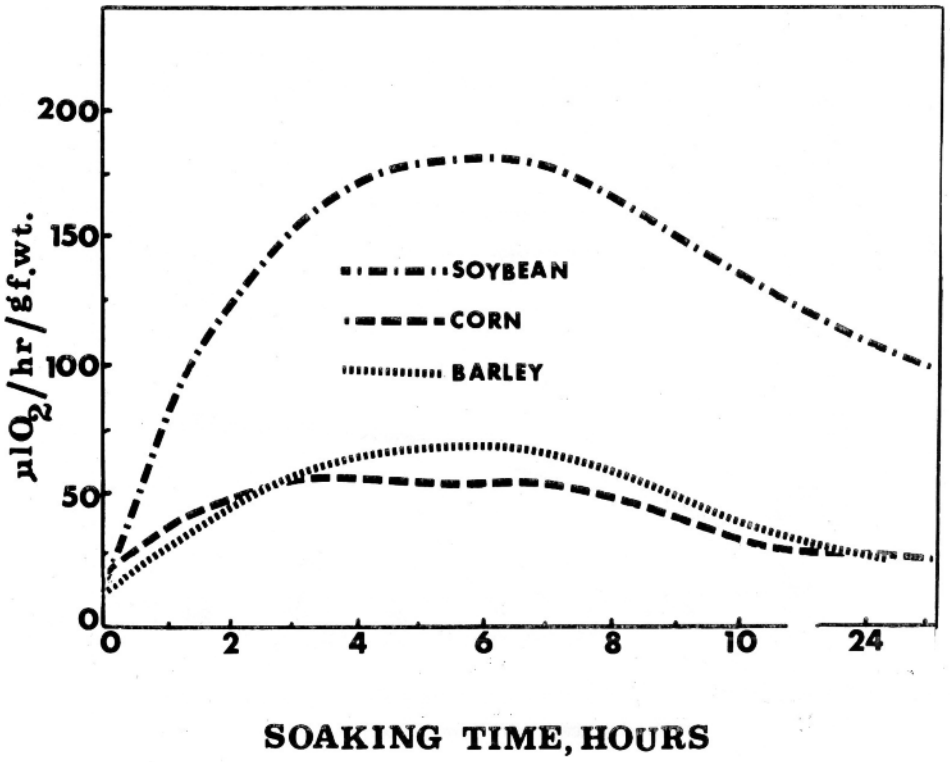

Fig. 2. Influence of soaking period on respiration rates of soybean, corn and barley seeds

increase in the respiration rates of the three species up to 6 hours of soaking, and from 6 to 24 hours, the rate of respiration decreased. Fig. 2 also indicates that soybean had different respiratory rates at each soaking level than barley and corn. At each soaking period, the rate of soybean respiration was 3 to 4 times higher than that of corn or barley.

The correlation coefficients between the soaking period and the rate of respiration were all non-significant, i.e., 0.21, 0.09 and 0.05 for soybean, corn and barley, respectively. These results and the data presented in Fig. 2 point to the fact that there is no direct or linear relationship between these two characters.

Nutile and Woodstock (8) have shown that respiration of sorghum seed during the first 6 hours of water imbibition varied directly with the initial seed moisture. Ragai and Loomis (9) found the respiration of corn seed to increase 100 times when the moisture content of the seed was increased from 14 to 23 percent at 21 C.

The present data show that the amount of dry weight loss depends directly on at least three factors; the soaking time, the respiratory activity of the seed and the seed species. The amount of protein in the seed presumably caused greater imbibitional power (comparing soybean to barley or corn) which up to a point (6 hours of soaking) produced higher respiration rates.

The respiration rates of seeds of soybean, corn and barley at different stages of germination are shown in Fig. 3. As the plumule or the coleoptile started to appear (stage 2) the oxygen consumption rates increased sharply. However, with the onset of chlorophyll formation and photosynthesis, the oxygen consumption rates decreased.

At stages 1 through 4, the respiration process was more active than the photosynthetic process thus causing increasing rates of oxygen consumption. With increased growth of the primary shoot at stages 5 and 6, photosynthesis dominated or equalled the respiration activities thus producing a decreased or constant rate of oxygen consumption. These results are in agreement with those of Bassiri (1) for red clover (*Trifolium pratense* L.) seedlings.

Fig. 3 also demonstrates the inherent variability in respiration for the 3 species tested. Barley seedlings in general, respired 1.6 and 2.0 times more than seedlings of soybean and corn, respectively. Probably barley seedlings synthesized enzymes necessary for

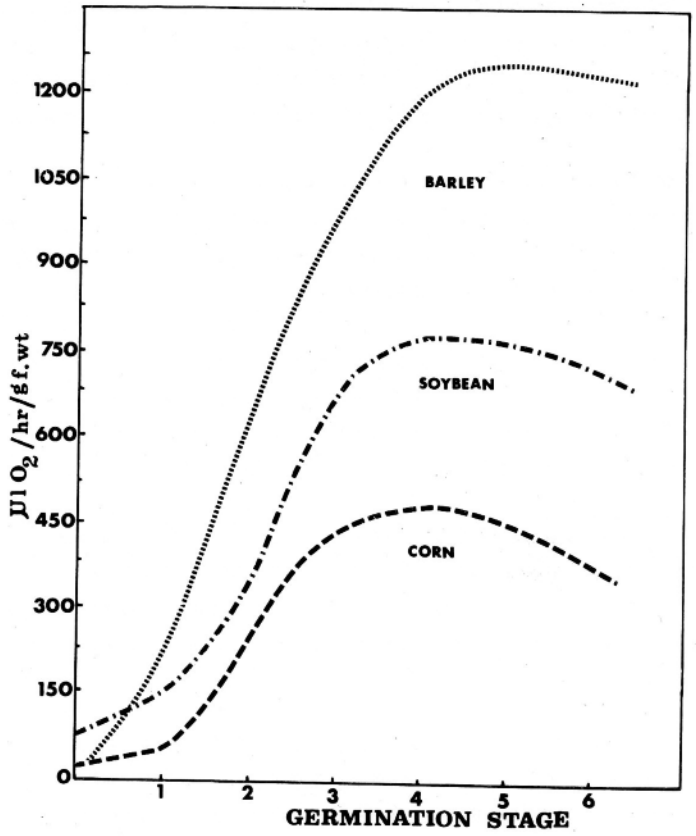

Fig. 3. Comparison of respiration rates of barley, soybeans and corn seeds at different stages of germination.

the respiration process more rapidly than corn or soybean.

LITERATURE CITED

1. Bassiri, A. 1971. Variation in quantitative characters of red clover, *Trifolium pratense* L., after two generations of selfing. Ph. D. Thesis, University of Wisconsin, 135 p.

2. Clark, L.E., J.W. Collier and R. Langston. 1968. Dormancy in *Sorghum bicolor* (L) Moench. II. Effect of pericarp and testa. Crop Sci. 8 : 155 - 158.

3. Kidd, F. and C. West. 1918. Physiological condition of the seed upon the course of subsequent growth and upon the yield. I. The effect of soaking seeds in water. Ann. Appl. Biol. 5 : 1 - 10.

4. Koller, D., A.M. Mayer, A. Poljakoff-Mayber and S. Klein. 1962. Seed germination. Ann. Rev. Plant Physiol. 13 : 437 - 464.

5. Larson, L.A. and K. Leanga. 1959. The effect of prolonged seed soaking on seedling growth of *Pisum sativum*. Can. J. Bot. 47 : 707 - 709.

6. Lyles, L. and C.D. Fanning. 1964. Effect of presoaking, moisture tension and soil salinity on the emergence of grain sorghum. Agron. J. 56 : 518 - 520.

7. Mayer, A.M. and A. Poljakoff - Mayber. 1966. The Germination of Seeds. Pergamon Press, New York. p. 37.

8. Nutile, G.E. and L.W. Woodstock. 1967. The influence of dormancy during desiccation treatments on the respiration and germination of sorghum. Physiol. Plant. 20 : 554 - 561.

9. Ragai, H. and W.E. Loomis. 1954. Respiration of maize grain. Plant Physiol. 29: 49 - 55.

10. Siegel, S.M. and L.A. Rosem. 1962. Effect of reduced 0_2 tension on germination and seedling growth. Physiol. Plant. 15 : 437 - 444.

11. Throneberry, G.D. and F.G. Smith. 1955. Relation of respiratory and enzymatic activity to corn seed viability. Plant Physiol. 30 : 336 - 343.

12. Umbreit, W.W., R.H. Burris and J.F. Stauffer. 1964. Manometric Techniques. Burgess Publishing Co. Minneapolis, Minn. p. 30 - 45.

13. Woodstock, L.W. and D.F. Grabe. 1967. Relationships between seed respiration during imbibition and subsequent seedling growth in *Zea mays* L. Plant Physiol. 42: 1071 - 1076.