

EFFECT OF STRATIFICATION ON BIOCHEMICAL COMPOSITION OF *CEDRUS DEODARA*

PARVEZ AHMAD SOFI AND S.D. BHARDWAJ

*Department of Silviculture and Agroforestry,
Dr. Y.S. Parmar University of Horticulture and Forestry,
Nauni-Solan (Himachal Pradesh).*

Introduction

Cedrus deodara, commonly called 'Deodar', is one of the most important group of species of Pinaceae. Deodar is found throughout the Western Himalayas from Afghanistan to Garhwal at elevations ranging from 1,200 to 3,050 m, being most common from 1,800 to 2,600 m. The species is widely distributed in the Western Himalayan states of Jammu & Kashmir, Himachal Pradesh and Uttarakhand. One of the complicating factor which affects the natural regeneration of Deodar forests is dormancy. Coniferous seeds are generally characterized by morpho-physiological dormancy, often complicated by the inhibitory effect of hard seed coat (Nikolaeva, 1990). This type of dormancy is primarily due to underdeveloped embryo or a decreased metabolic activity of the embryo and requires a treatment of cold stratification to produce sufficient levels of enzymes, hormones, soluble metabolites and other compounds needed by the embryo for germination.

Numerous physiological changes take place during the stratification of seeds, during which a large quantity of soluble metabolites are translocated into them and converted into new storage forms such as starch, fats and proteins. Various organic

and inorganic constituents also accumulate in them while still other constituents decrease in amount as seeds are stratified for various durations (Abdul-Baki and Anderson, 1970; Pitel, 1980; Ghosh *et al.*, 1981; Blanche *et al.*, 1990; Singh *et al.*, 1992; Gautam, 1997; Dogra, 2003). No information is available concerning the effect of stratification on biochemical constituents in *Cedrus deodara* seed. Therefore, the objective of the present study was to examine the effect of stratification on the biochemical constituents in Deodar seeds.

Material and Methods

The freshly harvested seeds of Deodar were placed for moist stratification upto 90 days at fifteen days intervals i.e. 15, 30, 45, 60, 75 and 90 days. The stratification was done in sand in different plastic petriplates and kept at 2 $^{\circ}$ C. The stratifying media i.e. sand was kept moist during entire period of stratification. The stratified seeds of Deodar were rinsed twice in distilled water and then dried to constant weight in an oven at 50 $^{\circ}$ C. One gram of dried sample of each treatment were placed in 20-25 ml of boiling ethanol (80%) for 10 minutes and decanted. Another 10-15 ml of boiling ethanol was added to the residue. Thereafter the two

extracted samples filtered and combined. The final volume was made 50 ml. This alcohol extract was used for estimation of total sugars while the residue was used for determination of starch. Dubois *et al.* (1951). For the estimation of sugars, the alcoholic extract was evaporated to dryness and residues were dissolved in 3 ml of distilled water and centrifuged to 4000 rpm for 15 minutes. The supernatant was decanted and used for estimation of sugars. Total sugar in the samples were estimated by phenol-sulphuric acid method given by Dubois *et al.* (1951). One ml of extract was taken in the test tube, to which one ml of 5 per cent phenol solution was mixed. This was then thoroughly shaken with 5 ml of concentrated H_2SO_4 and cooled under tap water. The transmittance of emerged yellow orange colour was measured at 490 nm wave length on spectrophotometer. The total sugars were determined from the standard curve drawn using known strengths of glucose solution and expressed as mg/g of dry weight. The residue left from the alcohol extract of dry samples were hydrolyzed by adding 5 ml of distilled water and 6 ml of 52 per cent perchloric acid. Contents were stirred for 30 minutes followed by addition of 20 ml distilled water, thereafter filtered and volume made to 100 ml. Glucose in samples was determined by phenol sulphuric acid method (Dubois *et al.*, 1951) and starch content was calculated by multiplying the glucose value with conversion factor 0.9 and expressed as mg/g dry weight. Soluble proteins were estimated by the method prescribed by Lowry *et al.* (1951).

Results and Discussion

The results of the effect of stratification on biochemical constituents

Table 1

Biochemical constituents of seeds as affected by stratification duration

Stratification duration (days)	Soluble protein (%)	Starch (%)	Total sugar (%)
No stratification (control)	5.20	8.50	5.58
15	5.77	8.44	5.79
30	5.87	8.29	5.86
45	6.29	7.50	6.11
60	6.58	7.61	6.78
75	6.23	6.97	6.08
90	5.52	6.78	5.76
CD _{0.05}	0.28	0.24	0.30

of seed viz., soluble protein, starch and total sugar are presented in Table 1.

It is evident that the soluble protein content and total sugar increased in seeds upto 60 days of stratification duration and thereafter decreased (Table 1). Statistical traits, indicated that the effect of stratification duration is statistically significant in respect of soluble protein, starch and total sugar. The maximum soluble protein (6.58%) and total sugar (6.78%) was recorded in seeds stratified for 60 days duration and the minimum soluble protein (5.20%) and total sugar (5.58%) was registered in seeds under control. The starch content shows a decreasing trend with the increase in stratification duration. The maximum starch (8.50%) was recorded in control which was statistically at par with 8.44% and 8.29% in seeds stratified for 15 and 30 days of stratification duration. The minimum starch content (6.78%) was exhibited in seeds

stratified for 90 days of stratification duration.

The rise in the soluble protein has been demonstrated in a number of species during natural and artificial aging (Abdul-Baki and Anderson, 1970; Pitel, 1980; Ghosh *et al.*, 1981; Blanche *et al.*, 1990). The peak in soluble protein coincided with the start of the decline in germination and so, the level of soluble protein could perhaps be used as a marker of seed vigour. Gautam (1997) reported that soluble proteins increased steadily from 30 days to 75 days of stratification and thereafter, recording a decline upto 120 days in seeds of *Quercus leucotrichophora*. Singh *et al.* (1992) also reported that protein content in *Pinus gerardiana* seeds decreased with increasing storage time.

The total sugars in *Cedrus deodara* seeds also exhibited a trend like soluble proteins. This may be due to conversion of starch into sugar during the stratification. Singh *et al.* (1992) reported that the reducing sugar content in seeds of *Pinus gerardiana* increased during first six

months after the storage period and decreased later showing a fluctuating trend. Dogra (2003) also reported the accumulation of total sugars in silver fir seeds increase steadily upto 60 days and showing a declining trend thereafter upto 90 days under dry stratification.

Starch content shows a declining trend with increasing stratification duration. The results are well in conformity with the observations of Blanche *et al.*, 1990 who reported that with the increase in artificial aging, the starch content gradually declines. The results are similar with that of Gautam (1997) who reported that starch content gradually declines from 30 days to 120 days in *Quercus leucotrichophora* seeds.

The biochemical constituents in seeds play a significant role in determining the viability and germination behaviour of the seeds. Since maximum soluble protein and total sugars were found at 60 days of stratification, therefore stratification for 60 days in Deodar seeds is recommended.

SUMMARY

Cedrus deodara (Deodar) seeds collected from Lolab area of Kashmir valley after subjecting to different stratification durations were quantitatively examined for biochemical parameters. There was increase in the soluble protein and total sugar content of seeds upto 60 days of stratification and thereafter decreased while the starch content showed a continuous decline with the increase in stratification duration. Stratification of seed for 60 days resulted in an increase of 1.38 per cent soluble protein and 1.20 per cent total sugar than control.

सीडरस डिओडारा की जैवरसायनिक रचना पर स्तरीकरण का प्रभाव
परवेज अहमद सोफी व एस०डी० भारद्वाज
सारांश

विभिन्न अवधियों तक स्तरीकरण करने के बाद कश्मीर उपत्यका के लोलब क्षेत्र से संग्रहीत देवदारु बीजों का उनके जैव रसायनिक परिमाण जानने को मात्रात्मक परीक्षण किया गया। 60 दिनों तक स्तरीकरण करने पर बीजों के विलेय प्रोटीन और कुल शर्करा तत्व में वृद्धि हुई किन्तु तदुपरान्त ये घट गए किन्तु स्तरीकरण की अवधि बढ़ाते जाने के साथ-साथ इनके मण्ड तत्व में निरन्तर कमी होती चली गई। बीजों का 60 दिन स्तरीकरण कराने से नियामक की तुलना में उनकी विलेय प्रोटीन में 1.38% की और कुल शर्करा में 1.20% की वृद्धि हुई।

References

- Abdul-Baki, A.A. and J.D. Anderson (1970). Viability and leaching of sugars from germinating barley. *Crop Sci.*, **10**: 31-34.
- Blanche, C.A., W.W Elam and J.D. Hodges (1990). Accelerated aging of *Quercus nigra* seed: biochemical changes and applicability as a vigour test. *Can. J. For. Res.*, **20**: 1611-1615.
- Dogra, Shivani (2003). Effect of stratification and plant bioregulators on the germination of Fir and Spruce. *M.Sc. Thesis*, Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan (H.P.).
- Dubois, M., K. Giles, J.K. Hamilton, P.A. Rebers and F. Smith (1951). A colorimetric method for the determination of sugars. *Nature*, **168**: 167.
- Gautam, J. (1997). Studies on presowing seed treatment on germination and seedling development of *Quercus leucotrichophora* Camus ex. Bahadur. *M.Sc. Thesis*, Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan, H.P.
- Ghosh, B., J. Adhikary and N.C. Banerjee (1981). Changes in some metabolites in rice seeds during aging. *Seed Sci Technol.*, **9**: 469-473.
- Lowry, O.H., N.J. Rosebrough, A.L. Fan and R.J. Randal (1951). Protein measurement with Folin-phenol reagent. *J. Biol. Chem.*, **193**: 265-275.
- Nikolaeva, M.G. (1990). Characteristics of seed germination in gymnosperms. *Botanicheskii Zhurnal.*, **75**(12): 1648-1656.
- Pitel, J.A. (1980). Accelerated aging studies of seeds of Jack pine (*Pinus banksiana* Lamb.) and Red oak (*Quercus rubra* L.). *Proc. International symposium on forest tree seed storage*, Patawawa. pp. 40-54.
- Singh, P.L., M.N. Gupta and A.L. Singh (1992). Deterioration of physico-chemical properties of Chilgoza (*Pinus gerardiana* Wall.) seed during storage. *Ind. J. Pl. Physiol.*, **35**(3): 231-237.
-