

VIABILITY ASSESSMENT THROUGH TZ TEST OF *Jatropha curcas* SEEDS, COLLECTED FROM DIFFERENT AGRO CLIMATIC ZONES OF INDIA**HARSHAL KUMAR^{a1}, J. RADHAMANI^b AND R. K. SARBHOY^c**^{ab}National Bureau of Plant Genetic Resources, IARI PUSA, New Delhi, India^cAgra College, Department of Botany, Dr. B.R. Ambedkar University, Agra, U.P., India**ABSTRACT**

Despite some studies on some *Jatropha curcas* provenances, a systematic documentation of morphological & physiological characteristics of *Jatropha curcas* seed available in different agro climatic regions in India, is lacking. Many specific questions about its production, commercialization and genetic improvement work are still in their infancy. Moreover before exploiting any plant for industrial application, it is imperative to have complete information about its seed viability pattern so that the potential of plant could be utilized maximally. Result of the present investigation revealed that potential viability, as estimated by TZ test is significantly and positively correlated with actual germination percentage in *Jatropha* seeds. So the TTC staining method can be recommended for evaluation of seed viability in *Jatropha* seed.

KEYWORDS : Seed Viability, TTC Staining Method, *Jatropha* Seeds , Germination Rate

Currently due to gradual depletion of world petroleum reserves and the impact of environmental pollution of increasing exhaust emissions, there is an urgent need to develop alternative energy resources, such as biodiesel fuel. Vegetable oil is a promising alternative because it has several advantages, it is renewable, environment-friendly and produced easily in rural areas, where there is an acute need for modern forms of energy. Therefore, in recent years several researches have been studied to use vegetable oils as fuel in engines as biodiesel. (Pramanik, 2003, Bozbas, 2005). With no competing food uses, this characteristic turns attention to *Jatropha curcas*, which grows in tropical and subtropical climates across the developing world (Openshaw,2000).The fact that *Jatropha* oil cannot be used for nutritional purposes without detoxification makes its use as energy or fuel source very attractive as biodiesel. *Jatropha curcas* (Linnaeus) is a multipurpose bush/small tree belonging to the family of Euphorbiaceae. It is a plant with many attributes, multiple uses and considerable potential.

Despite some studies on some *J. curcas* provenances, a systematic documentation of morphological & physiological characteristics of *Jatropha curcas* seed available in different agro climatic regions in India, is lacking. Many specific questions about its production, commercialization and genetic improvement work are still in their infancy. Moreover before exploiting any plant for industrial application, it is imperative to have complete information about its seed viability pattern so that

the potential of plant could be utilized maximally. Thus the main motive of this research will be a step towards filling this knowledge gap about correlation between Germination percentage and Viability Assessment through 2,3,5 Triphenyl Tetrazolium chloride test of *Jatropha curcas* seeds, collected from different agro climatic zones of India.

The TZ test was developed in Germany in the early 1940's by Georg Lakon and introduced in the United States after World War II. The use of the test has increased and expanded since then because it can be completed quickly, usually within a few hours. This is a faster method of determining seed viability than a standard germination test and TZ results are commonly used in place of germination test results. The procedure is also used to determine the viability of ungerminated seed at the end of a germination test. The TZ test measures the activity of the dehydrogenase enzymes used in the respiration process. Respiration is the cellular process of breaking down sugars to produce energy, carbon dioxide, and water, using oxygen. The enzymes react with substrates releasing hydrogen ions to the soluble tetrazolium chloride salt solution. The salt solution is reduced by the hydrogen ions. The colourless TZ salt solution is changed into an insoluble reddish compound called formazan. If the embryo and possibly endosperm or secondary nutrient reserve or storage tissues are actively respiring, formazan will be present and the tissues will stain red. The estimated viability of a seed lot is based on the number of seeds per testing sample that display the required staining, necessary to classify a seed as viable. A viable seed

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is considered to be capable of producing normal seedling under favourable conditions.

MATERIALS AND METHODS

The fruits of *Jatropha curcas* were assembled from NBPGR, Delhi and its regional stations at different agro climatic zones of India (Table, 1). The excised seeds without any mechanical damage were used for the experiment. The passport data of the assembled seeds from different zones of India has been depicted in Table 1. The passport data contained the information on *Jatropha* collected from FOUR different zones of India, viz Haryana, Uttaranchal, Rajasthan & Andhra Pradesh. All these seed collections (Small-S; Medium-M; Large-L) were assembled from the wildy distributed trees. Uttaranchal state represented the Western Himalayan region of India with temperate, cold conditions while Haryana represented the Transgangetic plain region of India. Similarly Rajasthan covered the Western dry region, and Andhra Pradesh represented the Vth zone i.e. East coast plains and hill region. The seeds were selected according to their condition where damaged seeds were discarded before seeds in good condition were cleaned and de-shelled.

The standard germination test was conducted by

placing the seeds in replicates of 50 seeds each in the tests and the seed samples were kept at $27 \pm 2^{\circ}\text{C}$ with 80-90 percent relative humidity in the seed germinator. In the fully mature seeds, the radical started emerging on the third day of plating. The seeds were considered germinated only when both radical and plumule emerged completely. The percentage germination was calculated based on the count of normal seedlings. Seedling with stunted growth or very slow growth was considered abnormal. Germination percentages were recorded as per ISTA (2003) guidelines. The normal and the abnormal seedlings were also counted separately for calculating the viability percentage. The seedlings were evaluated on the 15th day of plating and the speed of germination was calculated by counting the number of fresh emergence on each day till the final count.

RESULTS

The studies of TTC staining pattern suggested the use of 0.1 to 1% solution of TTC stain for the testing of seed viability in various crops viz. (Grabe, 1976, Moore, 1986) whereas the present studies indicated for jatropha seeds 0.1% TZ SOLUTION at 30 C followed by 24 hours of pre-moistening of seeds are suitable conditions for staining as the potential viability which was at par with the laboratory

Table 1: Seed Viability Determination By Germination and Triphenyl Tetrazolium Chloride (TTC) Test in Different Accession of *Jatropha curcas* Seeds

S. N.	Category (S/M/L) *	National ID	Viability by TTC (%)	Seed Germination (%)	SOURCE STATE
1	S-1	IC 551437	50	52	Haryana, Hissar
2	S-2	IC 545449	50	48	Uttaranchal
3	M-1	IC 551450	65	60	Haryana, Hissar
4	M-3	IC 312447	70	70	Jodhpur, Rajasthan
5	M-2	IC 545457	25	20	Uttaranchal
6	L-1	IC 551454	90	90	Haryana, Hissar
7	L-2	IC 545446	46	50	Uttaranchal
8	M-4	IC 550844	100	90	Hyderabad, (A.P)

*Category(S/M/L):- S: - Small; M: - Medium; L: - Large

Source State; Different Agro Climatic Zones

- 1).Uttaranchal - Western Himalayan region of India with temperate, cold conditions
- 2). Haryana - Transgangetic plain region of India.
- 3). Rajasthan - Western dry region, and
- 4). Andhra Pradesh - Vth zone i.e. East coast plains and hill region

germination test (ISTA, 1993). The evaluation of seeds under different categories based on the staining pattern is supported by seed germination test where staining of all essential structures of seeds is recognized as necessary for development of a normal seedling. There was no significant difference between the potential viability through TTC test and actual germination in seeds. Results of seeds revealed that potential viability, as estimated by TZ test is significantly and positively correlated with actual germination percentage. So the TTC staining method can be recommended for evaluation of seed viability in *Jatropha* seeds. Similar reports are observed in *Quercus falcate* and *Pinus elliottii* where significant correlation of TZ test result with germination was found (Bonner, 1984, Alvarez et.al., 1992).

In correlating seed viability by TTC & germination method there is a direct relation between viability & germination in *Jatropha* seeds. When the viability was <60% the germination was found to be more than 60%. When the viability was less than 60% germination also decreased. The viability in all the accessions was found to be in the range of 40-90%, whereas the larger seeds recorded maximum viability compared to the small and medium size seeds.

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