

Laboratory Manual

Testing Seed Viability of Native Plants of Kuwait

(Revised Edition)

S. Jacob, R. R. Thomas, and M. K. Suleiman

Plant Physiology Laboratory
Food Security Program
Environment and Life Sciences Research Center

Kuwait Institute for Scientific Research
P. O. Box 24885, Safat 13109, Kuwait

May 2025

Laboratory Manual

Testing Seed Viability of Native Plants of Kuwait

(Revised Edition)



S. Jacob, R. R. Thomas, and M. K. Suleiman

Plant Physiology Laboratory
Food Security Program
Environment and Life Sciences Research Center

Kuwait Institute for Scientific Research
P. O. Box 24885, Safat 13109, Kuwait

May 2025

For Use in **Plant Physiology Laboratory, KISR**

Prepared and Compiled by

Sheena Jacob

Principal Research Technician

Food Security Program

Kuwait Institute for Scientific Research

Rini Rachel Thomas

Research Associate

Food Security Program

Kuwait Institute for Scientific Research

Under the Guidance of

Dr. Majda Khalil

Coordinator, Kuwait Environmental Remediation Program

Food Security Program

Kuwait Institute for Scientific Research

*This manual is prepared based on the ISTA guidelines and enriched by years of experience in native plant projects; it serves as a reference for future generations. It can be updated as new knowledge and experience are gained.

Cite this Manual as

Jacob, S., Thomas, R. R., Khalil, M. K. 2025. Laboratory Manual on Testing Seed Viability of Native Plants of Kuwait, Kuwait Institute for Scientific Research, Kuwait.

Designed by: **Oudah Orabi**

Acknowledgment

The authors would like to express their sincere gratitude to the Kuwait Foundation for the Advancement of Sciences (KFAS) for funding the study (FA125C) and to the management of the Kuwait Institute for Scientific Research (KISR) for its unwavering support. The authors would like to thank Dr. N. R. Bhat, Dr. Arvind Bhatt, and Dr. Sara Al Ateeqi for their invaluable guidance, support, interest, and encouragement.

The team also extends its heartfelt appreciation to Mr. Processo Ramos, a former KISR employee, for generously sharing his extensive knowledge on Kuwait's native plants. His passion for plants was inspiring, and his patience and willingness to clarify doubts, despite his busy schedule, were truly commendable. The authors would also like to thank Ms. N. J. Anupama, for her consistent assistance in the laboratory work.

Table of Contents

Introduction.....	1
Viability Assessment through TTC Method	1
Principle of TTC Test.....	1
Material Requirement.....	2
Methodology.....	3
Viability Assessment through the Germination Method.....	5
List of Native Plants Covered	6
<i>Acacia pachyceras</i> (Al Talha, الطلحة)	7
<i>Aeluropus lagopoides</i> (Ikrish, عكرش)	9
<i>Anvillea garcinii</i> (Nugd, نقد)	10
<i>Brassica tournefortii</i> (Harraizah, حريزة)	11
<i>Calligonum polygonoides</i> (Artah, أرطى)	15
<i>Cenchrus ciliaris</i> (Subbat, Thumum, سبط ثمام).....	18
<i>Convolvulus oxyphyllus</i> (Ethris, Udhris, عخرس)	21
<i>Deverra triradiata</i> (Hazhas,).....	25
<i>Farsetia aegyptia</i> (Al Lbanah, اللبانة)	28
<i>Halocnemum strobilaceum</i> (Theluth, ثليث)	30
<i>Halothamnus iraquensis</i> (Qadqad, قضقاظ), <i>Haloxylon salicornicum</i> (Al Rimth, الرمث), <i>Salsola imbricata</i> (Mulaih, Gathraf, خذراف, مليح), and <i>Seidlitzia rosemarinus</i> (Al Shinan الشنان).....	33
<i>Helianthemum lippi</i> (Rag-rug, رقروق)	40
<i>Horwoodia dicksoniae</i> (Khuzamah, خزامة).....	43
<i>Lycium shawii</i> (Awsaj, العوسج)	46
<i>Nitraria retusa</i> (Ghardaq, الغردق).....	49

<i>Ochradenus baccatus</i> (Gurdi, غرضي).....	51
<i>Panicum turgidum</i> (Thamam, ثمام).....	54
<i>Peganum harmala</i> (Harmal, الحرمل).....	59
<i>Pennisetum divisum</i> (Thumam, ثمام).....	61
<i>Prosopis cineraria</i> (Ghaf, الغاف الرمادية).....	64
<i>Rhanterium epapposum</i> (Arfaj, العرفج).....	67
<i>Tamarix aucheriana</i> (Athal, Tarfa, أثل، طرفاء).....	70
<i>Ziziphus spina-christi</i> (Sidr, سدر).....	73
<i>Zygophyllum qatarense</i> (Harm, هرم).....	79
References.....	82
Appendix A: Germination Test Sheet	85

List of Figures

Figure 1. Seeds of <i>Acacia pachyceras</i> .	8
Figure 2. Stained viable seeds of <i>Acacia pachyceras</i> .	8
Figure 3. Spikes and extracted seed of <i>Aleuopus lagopoides</i> .	9
Figure 4. Capitulum and extracted seeds of <i>Anvillea garcinii</i> .	10
Figure 5. Stained viable seeds of <i>Anvillea garcinii</i> .	10
Figure 6. Seeds of <i>Brassica tournefortii</i> .	12
Figure 7. Seed of <i>Brassica tournefortii</i> .	12
Figure 8. Dry and imbibed seeds of <i>Brassica tournefortii</i> .	13
Figure 9. Unstained and stained viable embryo of <i>Brassica tournefortii</i> .	14
Figure 10. Germination of <i>Brassica tournefortii</i> .	14
Figure 11. Seeds of <i>Calligonum polygonoides</i> .	15
Figure 12. Longitudinally split and stained viable seed of <i>Calligonum polygonoides</i> .	16
Figure 13. Germinated <i>Calligonum polygonoides</i> seed.	17
Figure 14. Spikelet and extracted seed of <i>Cenchrus ciliaris</i> .	19
Figure 15. Germinated seed of <i>Cenchrus ciliaris</i> .	20
Figure 16. Stained seed of <i>Cenchrus ciliaris</i> after TTC test.	20
Figure 17. Seeds of <i>Convolvulus oxyphyllus</i> .	22
Figure 18. Germinated seedling of <i>Convolvulus oxyphyllus</i> .	23
Figure 19. Stained viable seed of <i>Convolvulus oxyphyllus</i> (with folded cotyledons).	24
Figure 20. Seeds of <i>Deverra triradiata</i> .	25
Figure 21. Mericarp of <i>Deverra triradiata</i> .	26
Figure 22. Dry and soaked mericarp of <i>Deverra triradiata</i> .	27
Figure 23. Unstained and stained viable mericarp of <i>Deverra triradiata</i> .	27
Figure 24. Winged seed of <i>Farsetia aegyptia</i> .	28
Figure 25. Germinated seedling of <i>Farsetia aegyptia</i> .	29
Figure 26. Dry and stained viable seed of <i>Farsetia aegyptia</i> .	29

Figure 27. Seeds of <i>Halocnemum strobilaceum</i> .	30
Figure 28. Seed of <i>Halocnemum strobilaceum</i> .	31
Figure 29. Dry and imbibed seed of <i>Halocnemum strobilaceum</i> .	31
Figure 30. Germinated seed and seedling of <i>Halocnemum strobilaceum</i> .	32
Figure 31. Dry and stained viable seed of <i>Halothamnus iraqensis</i> .	34
Figure 32. <i>Halothamnus iraquensis</i> perianth with and without wings under microscope.	35
Figure 33. Dry and stained viable seed of <i>Haloxylon salicornicum</i> .	36
Figure 34. Dry and stained viable seed of <i>Salsola imbricata</i> .	37
Figure 35. Winged and de-winged perianth of <i>Seidlitzia rosemarinus</i> .	38
Figure 36. Perianth and coiled embryo of <i>Seidlitzia rosemarinus</i> .	39
Figure 37. Seeds of <i>Helianthemum lippi</i> .	40
Figure 38. Dry and imbibed seeds of <i>Helianthemum lippi</i> .	41
Figure 39. Seed of <i>Helianthemum lippi</i> .	41
Figure 40. Germinated seed and seedling of <i>Helianthemum lippi</i> .	42
Figure 41. Winged fruit of <i>Horwoodia dicksoniae</i> .	43
Figure 42. <i>Horwoodia dicksoniae</i> seed extracted from the winged fruit.	44
Figure 43. <i>Horwoodia dicksoniae</i> seed.	44
Figure 44. Stained seed of <i>Horwoodia dicksoniae</i> .	45
Figure 45. Seeds of <i>Lycium shawii</i> .	46
Figure 46. Microscopic view of <i>Lycium shawii</i> seed.	47
Figure 47. Germinated seed of <i>Lycium shawii</i> .	47
Figure 48. Unstained and stained seed of <i>Lycium shawii</i> .	48
Figure 49. Seeds of <i>Nitraria retusa</i> .	49
Figure 50. Stained seeds of <i>Nitraria retusa</i> .	50
Figure 51. Seeds of <i>Ochradenus baccatus</i> .	51
Figure 52. Stained seed of <i>Ochradenus baccatus</i> .	52
Figure 53. Germinated seedling of <i>Ochradenus baccatus</i> .	53
Figure 54. Seeds of <i>Panicum turgidum</i> .	55

Figure 55. Seed of <i>Panicum turgidum</i> enclosed in the husk.....	56
Figure 56. De-husked seed of <i>Panicum turgidum</i>	56
Figure 57. Non-viable and viable seed of <i>Panicum turgidum</i>	57
Figure 58. Viable seed of <i>Panicum turgidum</i>	57
Figure 59. Germination of <i>Panicum turgidum</i>	58
Figure 60. Seeds of <i>Peganum harmala</i>	59
Figure 61. Germinated seedlings of <i>Peganum harmala</i>	59
Figure 62. Stained seed of <i>Peganum harmala</i>	60
Figure 63. <i>Pennisetum divisum</i> spikelet and seeds	61
Figure 64. <i>Pennisetum divisum</i> seeds extracted from the spikelet.....	62
Figure 65. Stained seed of <i>Pennisetum divisum</i>	62
Figure 66. Germinated seed of <i>Pennisetum divisum</i>	63
Figure 67. Seeds of <i>Prosopis cineraria</i>	65
Figure 68. Seed of <i>Prosopis cineraria</i>	66
Figure 69. Unstained and stained viable embryo of <i>Prosopis cineraria</i>	66
Figure 70. <i>Rhanterium epapposum</i> capitulum and seed extracted from capitulum.....	68
Figure 71. Germination test in <i>Rhanterium epapposum</i>	69
Figure 72. Stained seed of <i>Rhanterium epapposum</i>	69
Figure 73. Fruit of <i>Tamarix aucheriana</i>	70
Figure 74. Seeds of <i>Tamarix aucheriana</i>	71
Figure 75. Germinated seedling and TTC stained seed of <i>Tamarix aucheriana</i>	72
Figure 76. Endocarp of <i>Ziziphus spina-christi</i>	74
Figure 77. Extraction of <i>Ziziphus spina-christi</i> seed from the endocarp.	75
Figure 78. Seed of <i>Ziziphus spina-christi</i>	76
Figure 79. Unstained and stained viable seed of <i>Ziziphus spina-christi</i>	76
Figure 80. Germination of <i>Ziziphus spina-christi</i>	77
Figure 81. Germination and separation of seedlings in <i>Ziziphus spina-christi</i>	78

Figure 82. <i>Zygophyllum qatarense</i> seeds.....	79
Figure 83. Dry and stained seed of <i>Zygophyllum qatarense</i>	80
Figure 84. Germinated seedling of <i>Zygophyllum qatarense</i>	81



Introduction

Seed viability test is used to determine whether individual seeds in a sample lot are dead or alive. This test is crucial for assessing the germination potential of seeds, which is significant for determining seed rates in field planting/revegetation, and prior to research experiments or germination studies. It is also important for monitoring the quality of seeds before storage. Though the viability can be tested through germination, viable seeds of some species may not germinate immediately after maturation, even under favorable conditions, due to dormancy (physical, chemical, or physiological).

The methodology outlined in this manual aligns with the protocol suggested by the International Seed Testing Agency (ISTA) in the International Rules for Seed Testing, 2007 Edition. The methodology for checking the seed viability of food crops is well established by many testing agencies in the world and is widely practiced. However, the protocol for checking the seed viability of Kuwait's native desert species remains undocumented or unknown. Hence, the authors have compiled and presented these protocols for the viability testing of Kuwait's native plants based on their laboratory experience with native seeds, aiming to provide a guideline for future seed technologists involved in the storage and revegetation of native species in Kuwait.

Generally, seed viability can be tested using either the 2, 3, 5 Triphenyl Tetrazolium Chloride Test (TTC test) or the germination test. Testing the viability of seeds through the germination method can be challenging for seeds that exhibit dormancy. Hence, for such species with dormancy, the TTC test offers a more reliable method for assessing viability. This manual provides an optimized protocol for testing the viability of 27 native seeds, including a detailed methodology, supported by pictorial documentation.

Viability Assessment through TTC Method

Principle of TTC Test. The TTC test relies on the principle that a living, viable tissue of an embryo reduces TTC to red formazan due to the action of dehydrogenase enzymes (ISTA, 2007). In this method, seed viability is evaluated based on the staining pattern and color intensity (i.e., completely stained, partially stained, or completely unstained seeds). In this test, the reduction process takes place only in the living cells, and the viable seeds show staining across all tissues, which is necessary for normal seedling development. The position and size of the stained areas are also critical factors in assessing seed viability.

Material Requirement.

Reagents Required. The reagents required for viability testing and their preparation procedures are detailed subsequently.

1. 2, 3, 5 –Triphenyl tetrazolium chloride (TTC) solution: A 1% TTC solution with a pH of 6.5 to 7.5 is effective for viability testing. However, for some species, 0.1% solution may be more suitable. To prepare 1% TTC solution, 1 g of TTC should be dissolved in 100 ml of distilled water. If the pH of the solution has to be adjusted to the desired level, a phosphate buffer solution should be used. If buffer solution is used, 1% TTC solution has to be prepared by dissolving 1 g of TTC in 100 ml of the buffer solution. However, in most of the cases, buffer solutions are not required. Since TTC is light- sensitive, solutions must be stored at 4 °C in dark glass bottles. It is advisable to wrap glass bottles with aluminum foil to prevent light exposure. Avoid using metallic containers, as TTC might react with certain metals.
2. Buffer solution (ISTA, 2007) can be prepared as follows:
 - a. Solution 1: Dissolve 9.078 g of potassium dihydrogen phosphate (KH_2PO_4) in 1000 ml of distilled water.
 - b. Solution 2: Dissolve 9.472 g of disodium hydrogen phosphate (Na_2HPO_4) or disodium hydrogen phosphate dihydrate ($\text{NaHPO}_4 \times 2\text{H}_2\text{O}$) in 1000 ml of distilled water.
 - c. Mix two parts of solution 1 and three parts of solution 2 to obtain a phosphate buffer solution with the desired pH value of 6.5 to 7.5.
 - d. Gibberellic acid (GA_3) solution is used for conducting dormancy-breaking treatments to determine seed viability through the germination method. Seeds exhibiting physiological dormancy are treated with a GA_3 solution of a specific pre-determined concentration (varies with species), prior to further treatments. To prepare the solution, dissolve the appropriate amount of GA_3 in distilled water to achieve the desired concentration. For example, dissolving 750 mg of GA_3 in 1000 ml of distilled water produces 750-ppm solution.

Glassware and Other Items.

1. Petri dishes
2. Dark glass bottles to store TTC
3. Measuring cylinders, volumetric flask, and beakers
4. Aluminium foil
5. Sieves
6. Wash bottle
7. Forceps
8. Blades/Scalpel
9. Permanent markers to mark the species name and details on the Petri dish
10. Soil medium for germination tests
11. Filter paper
12. Magnifying glass
13. Plastic trays and jiffy pots

Equipment.

1. Refrigerator
2. Growth chamber
3. Weighing scale
4. pH meter
5. Hot plate with magnetic stirrer
6. Stereo microscope with camera*

The photographs in this manual were taken using LEICA stereo microscope EZ4D.

Methodology.

1. Pre-moistening of Seeds. Seeds need to be pre-moistened prior to staining, due to the following reasons:
 - i. Imbibed seeds are easier to cut or puncture.
 - ii. Moist seeds facilitate uniform staining, simplifying evaluation.

Pre-moistening can be achieved through one of the following methods:

- a. Slow moistening: Place seeds between or on top of moist filter papers to allow gradual imbibition. This method is suitable for species that may be damaged by direct soaking in water, as

well as for old and dry seeds.

b. Soaking in water: Fully immerse the seeds in water until they are completely imbibed. Usually, the soaking period is 24 hours, but for species that require longer durations, change the water after 24 hours.

Some species, such as *Farsetia aegyptia*, produces sticky mucilage immediately, when it comes in contact with water during pre-moistening. This issue can be addressed by either of the following:

- a. Drying the seed surface by rubbing with a cloth or using filter paper.
- b. Soaking the seeds in a 1–2% solution of aluminium potassium sulphate ($\text{AlK}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$) for 5 minutes.

Species, such as *Acacia pachyceras*, have hard seed coats, which prevent water imbibition. Hence, the seed coat must be scarified before staining to facilitate imbibition. Sandpaper can be used for this purpose, but care should be taken to avoid damaging the hilar region during scarification.

2. Exposing the Tissues Prior to Staining. It is necessary to expose embryonic tissues prior to staining to facilitate the penetration of TTC and allow accurate assessment of staining patterns. Seed coats can be opened or removed prior to staining. Pre-moistened seeds are exposed at non-essential areas using a needle or sharp scalpel, without causing any damage to the essential areas such as the embryo and the hilar region. Either a longitudinal or a transverse cut can be used to expose the embryo. The method and type of seed exposure vary by species. Immediately after exposing the embryo, **the seeds should be kept moistened until they are immersed in the TTC solution.**

Methods of Seed Embryo Exposure:

- a. Piercing the non-essential part of the seed prior to staining.
 - b. Longitudinal cutting through the middle of the embryonic axis and through three quarters of the endosperm.
 - c. Transverse cutting through non-essential tissues of the seed.
3. Staining. The prepared seeds (with exposed embryo) should be soaked in either 0.1% or 1% TTC solution, normally for 24 hours, depending on the species for staining (staining time may vary with species). The seeds in each replication should be placed in separate Petri dishes and covered with aluminum foil to prevent the exposure of solution to light. At the termination of the staining

period, the seeds have to be rinsed with water to remove any excess stain before examination.

4. Evaluation. The stained and rinsed seeds should be carefully observed with or without using a microscope, depending on their size, to determine the viability. A seed is considered viable if all essential parts required for the emergence and development of a normal seedling are stained. For partially stained seeds, if the essential tissues are stained, it is still considered viable.

Usually, there should not be significant difference between viability measured by the TTC test and the germination test, especially for non-dormant seeds or those without hard seed coats, leading to physical dormancy, provided that optimal growth conditions are met and there is no pest damage or other factors leading to seed quality deterioration.

5. Calculation of Viability Percentage. The number of stained seeds in each replicate should be recorded, and the average viability percentage has to be calculated as follows, to get the final result:

$$\text{Viability (\%)} = \frac{(\text{Total Number of seeds stained})}{(\text{Total number of seeds used})} \times 100$$

6. Experimental Design: It is preferable to have four replications of 25 seeds each or five replications of 20 seeds each for assessing the viability.

Viability Assessment through the Germination Method

The germination method simplifies the accurate determination of viability for species with extremely small seeds. For some species, this method can be used to precisely record the viability due to their large size or other specific factors. The germination method is best suited for seeds that do not exhibit any type of dormancy. This handbook explains the most effective techniques for estimating the viability of the species listed subsequently.

List of Native Plants Covered

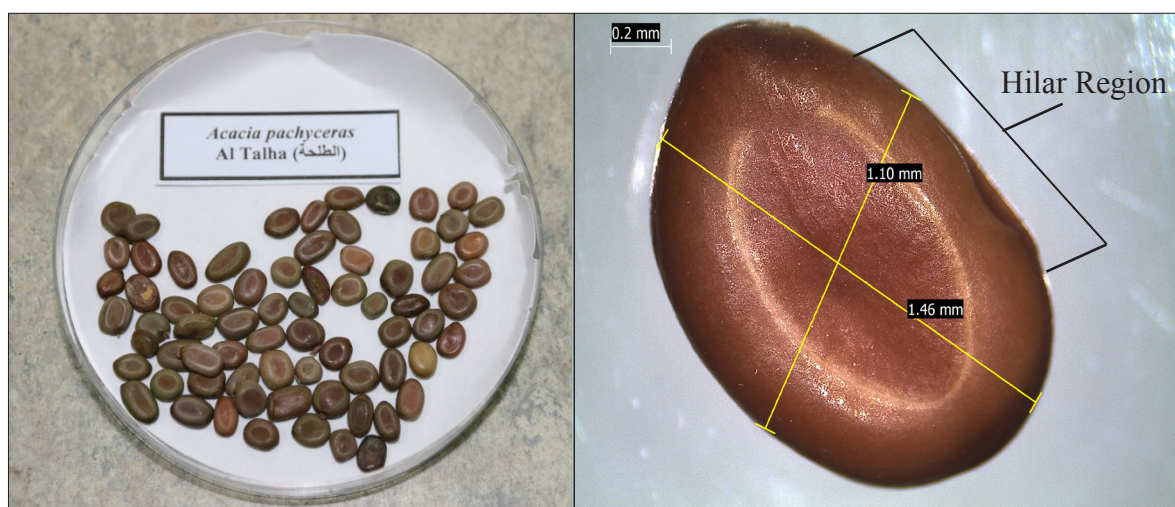
01. *Acacia pachyceras*
02. *Aeluropus lagopoides*
03. *Anvillea garcinii*
04. *Brassica tournefortii*
05. *Calligonum polygonoides*
06. *Cenchrus ciliaris*
07. *Convolvulus oxyphyllus*
08. *Deverra triradiata*
09. *Farsetia aegyptia*
10. *Halocnemum strobilaceum*
11. *Haloxylon salicornicum*
12. *Halothamnus iraqensis*
13. *Helianthemum lippi*
14. *Horwoodia dicksonia*
15. *Lycium shawii*
16. *Nitraria retusa*
17. *Ochradenus baccatus*
18. *Panicum turgidum*
19. *Peganum harmala*
20. *Pennisetum divisum*
21. *Prosopis cineraria*
22. *Rhanterium epapposum*
23. *Salsola imbricata*
24. *Seidlitzia rosemarinus*
25. *Tamarix auheriana*
26. *Ziziphus spina-christi*
27. *Zygophyllum qatarense*

Acacia pachyceras (Al Talha, الطلحة)

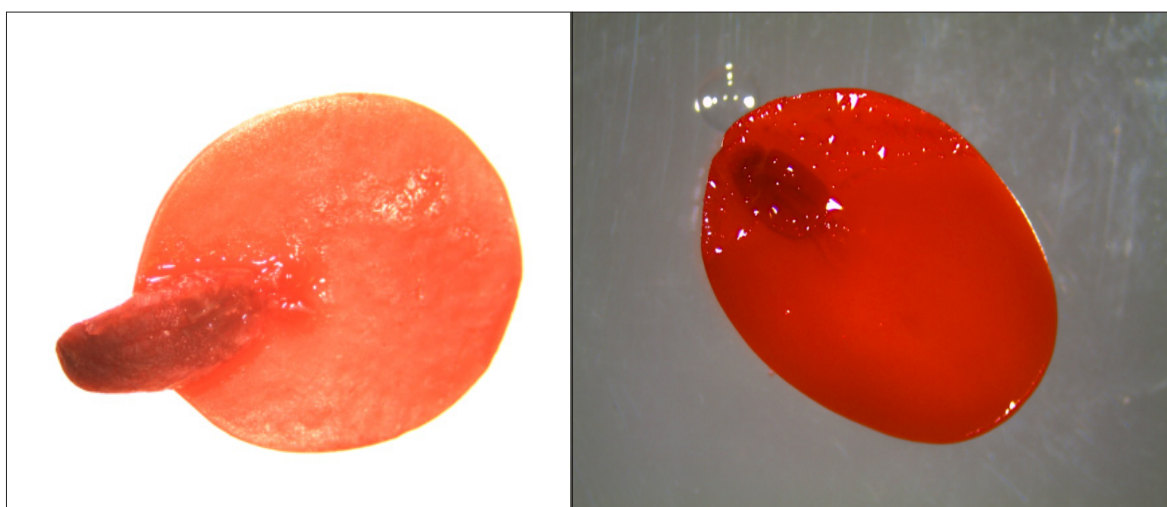
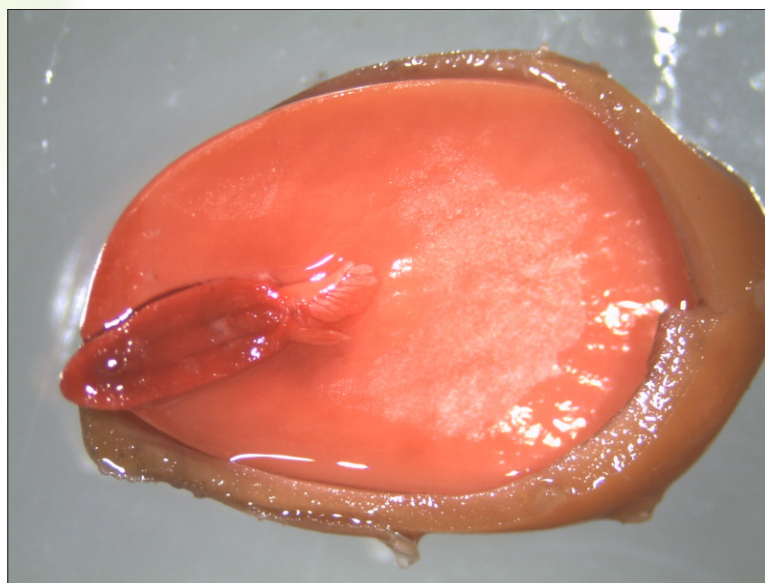
Family: Fabaceae

Preferred Method: TTC

Acacia pachyceras has a hard seed coat that prevents water imbibition (physical dormancy). Rupturing or softening the seed coat through scarification facilitates water imbibition (Suleiman et al., 2018). Refer to the methodology section for experimental details. To overcome physical dormancy, the seeds should be mechanically scarified using sandpaper, cautiously preventing any damage to the hilar region (Figure 1). Imbibition of the scarified seeds can be facilitated by soaking them in water for 24 hours. Longitudinally cut pre-moistened seeds can then be soaked in 1% TTC solution in petri plates. The Petri plates should be covered with aluminum foil, to prevent light exposure, and incubated at room temperature (25°C) for 24 hours. After 24 hours, the seeds should be rinsed and observed for staining to determine viability (Figure 2). Re-confirmation of viability can be achieved by the germination method. The scarified and imbibed seeds should be placed in a Petri dish, lined with moistened filter paper, for germination, which will start after 24 hours and might continue for up to a week.



► Figure 1. Seeds of *Acacia pachyceras*.



► Figure 2. Stained viable seeds of *Acacia pachyceras*.

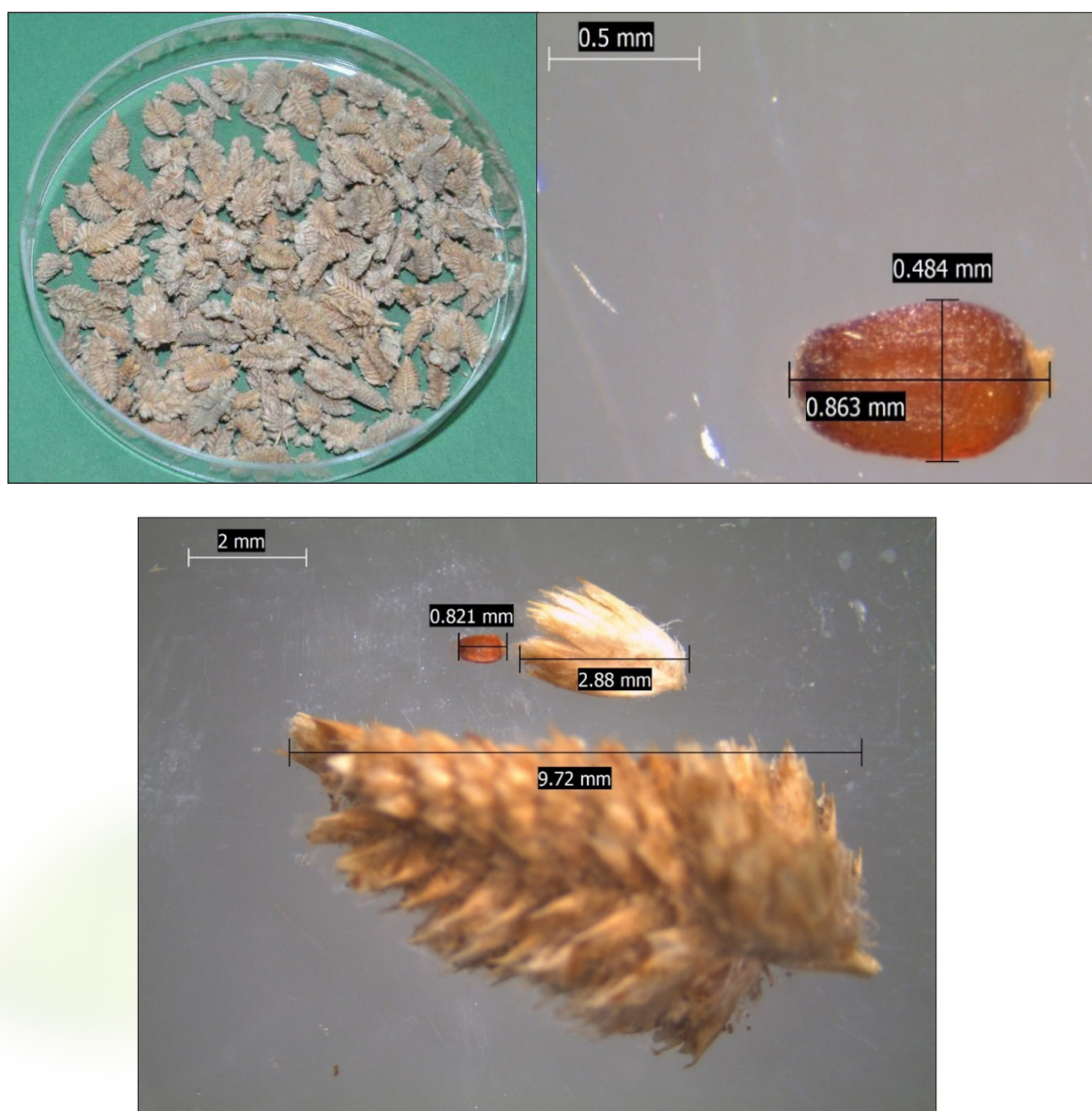
Aeluropus lagopoides (Ikrish, عكرش)

Family: Poaceae



Preferred Method: TTC

Because of its small size, intact seeds of *Aeluropus lagopoides* extracted from spikes are used for the TTC test (Figure 3). After imbibing, the seeds should be removed from water and soaked in 1% TTC solution in Petri dishes. The Petri dishes, covered with aluminum foil, should be incubated at room temperature (25°C) for 24 hours. Following incubation, seeds should be rinsed thoroughly with distilled water to remove excess stain and examined under the microscope to record the number of stained seeds.



► Figure 3. Spikes and extracted seed of *Aeluropus lagopoides*.

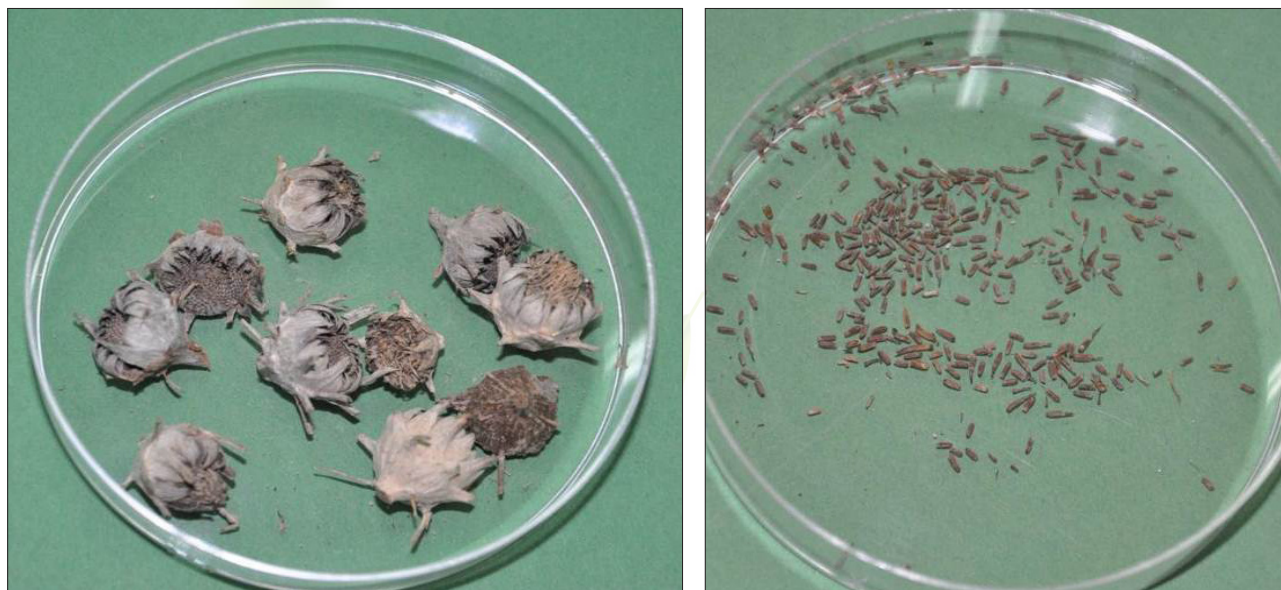
Anvillea garcinii (Nugd, نقاد)

Family: Asteraceae

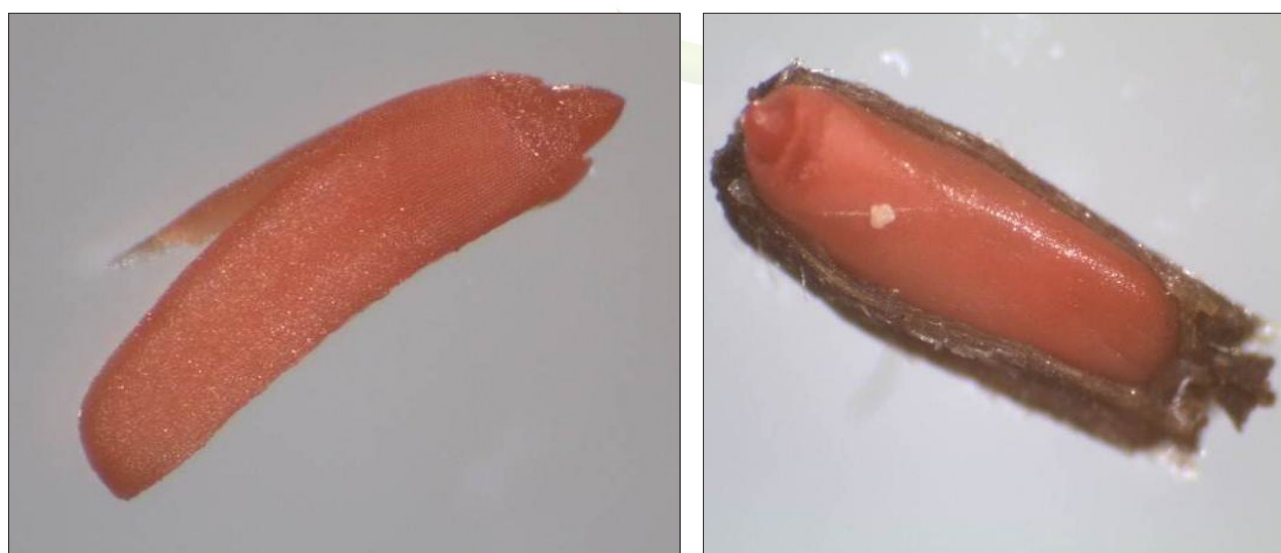


Preferred Method: TTC

Seeds extracted from the capitulum should be split longitudinally to expose the embryo and then immersed in distilled water overnight (Figure 4). Moist seeds should be removed from water and soaked in 0.1% TTC solution in Petri dishes. The Petri dishes should be covered with aluminum foil and kept at room temperature (25°C) for 24 hours. Following incubation, seeds should be washed thoroughly with distilled water to remove excess stain and examined under the microscope to count the number of stained seeds (Figure 5).



► Figure 4. Capitulum and extracted seeds of *Anvillea garcinii*.



► Figure 5. Stained viable seeds of *Anvillea garcinii*.

Brassica tournefortii (Harraizah, حريزة)

Family: Brassicaceae

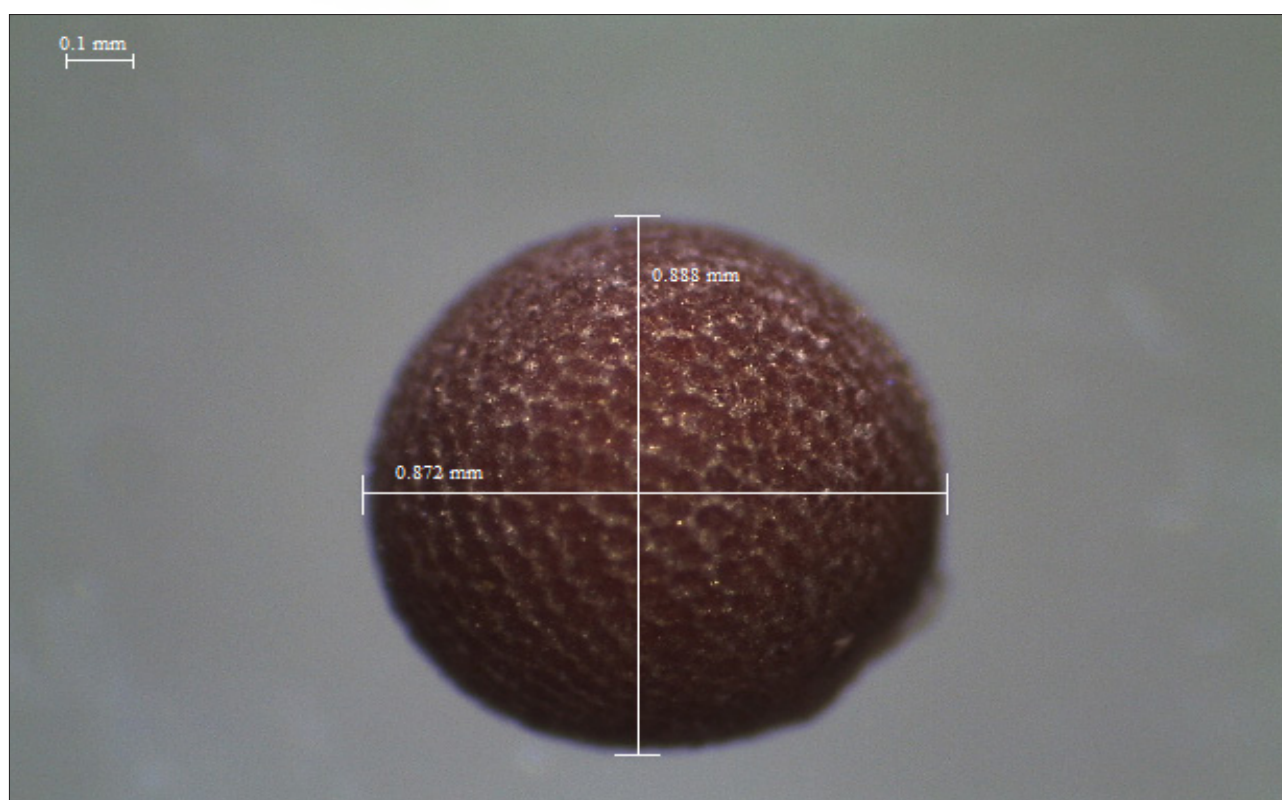
Preferred Method: TTC

Brassica tournefortii exhibits physiological dormancy immediately after ripening. Post-harvest, dormancy may vary depending on biotypes (exomorphological aspects and ecophysiological adaptations) and maternal environment (Mahajan et al., 2018). Light and temperature are other factors that influence germination. However, dormancy is not an issue in the viability testing of seeds stored for over a year. Hence, TTC is the preferred method for assessing seed lot viability of this species. The seeds of *Brassica tournefortii* (Figures 6 to 8) are small and glabrous, and a mucilaginous layer forms around the seed when the seeds come into contact with water, making it difficult to handle. Hence, cutting pre-moistened seeds to expose the embryo for the TTC test can be challenging (Figure 9). Pre-soaked seeds should be held firmly with forceps and cut longitudinally using a sharp scalpel prior to treating with 1% TTC (Refer to the section on Viability Assessment through the TTC Method for detailed methodology).

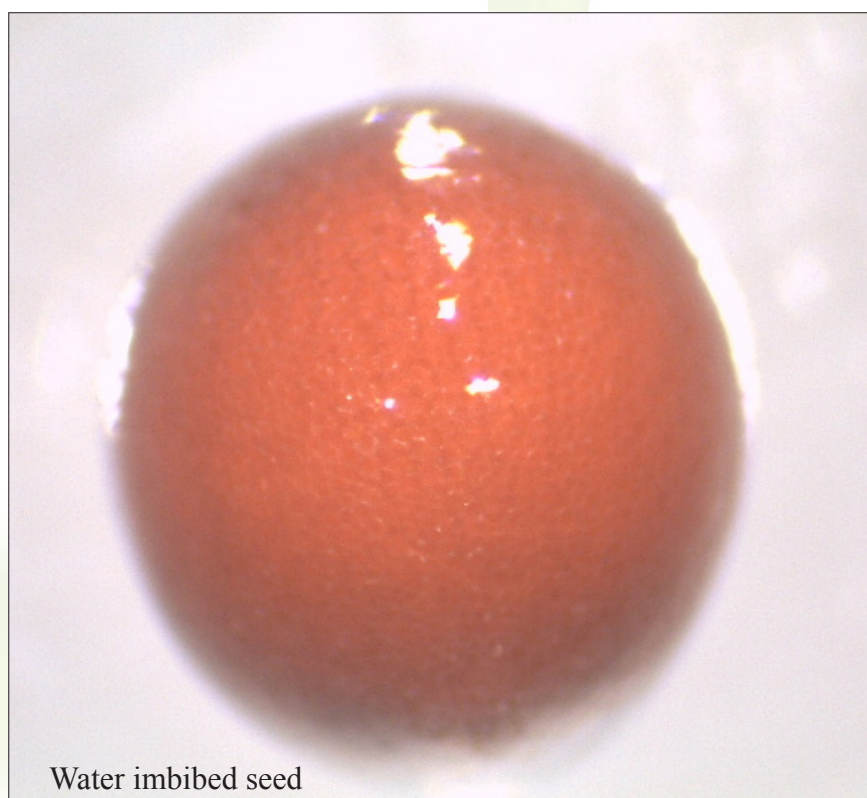
The viability of *Brassica tournefortii* can be confirmed using the germination test. Pre-soaked seeds can be germinated in a Petri dish lined with filter paper and incubated in the dark at 25°C (Figure 10).



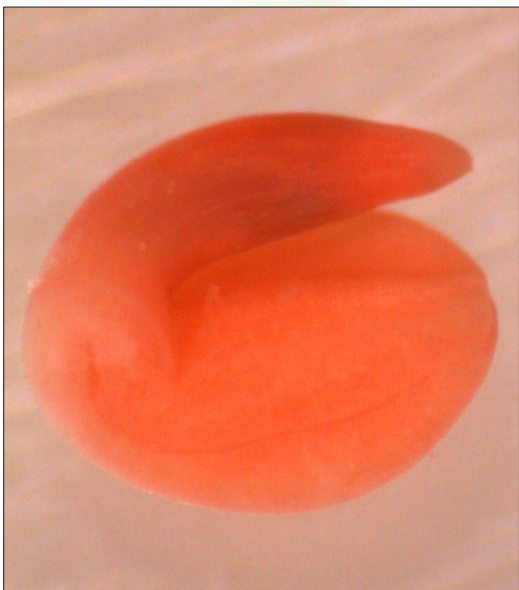
► Figure 6. Seeds of *Brassica tournefortii*.



► Figure 7. Seed of *Brassica tournefortii*.



► **Figure 8. Dry and imbibed seeds of *Brassica tournefortii*.**



▶ Figure 9. Unstained and stained viable embryo of *Brassica tournefortii*.



▶ Figure 10. Germination of *Brassica tournefortii*.

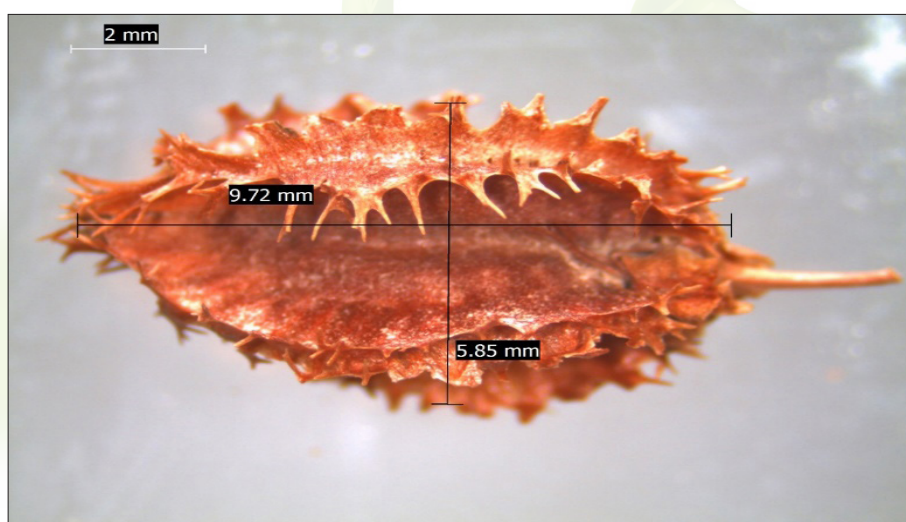
Calligonum polygonoides (أرطى, Artah)

Family: Polygonaceae

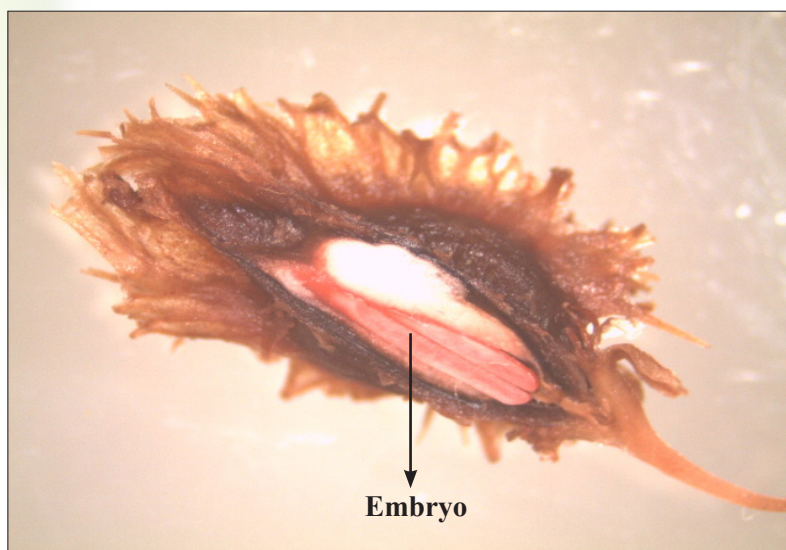


Preferred Method: TTC

Seeds of *Calligonum polygonoides* have a hard, impermeable testa (Figure 11), which prevents the imbibition of water and oxygen, thereby inhibiting germination (Dhief et al., 2012; Derbel and Chaieb, 2013). After pre-soaking the seeds in distilled water overnight to soften the seed coat, they should be split longitudinally to expose the embryo. The moist seeds should be removed from water and soaked in 1% TTC solution in Petri dishes, covered with aluminum foil, and kept at room temperature (25°C) for 24 hours. After incubation, the seeds should be washed thoroughly with distilled water to remove excess stain and examined under the microscope to record the number of seeds stained with TTC (Figures 12). An image of germinated seedling is provided below for reference (Figure 13)



► Figure 11. Seeds of *Calligonum polygonoides*.



► **Figure 12. Longitudinally split and stained viable seed of *Calligonum polygonoides*.**



► Figure 13. Germinated *Calligonum polygonoides* seed.

Cenchrus ciliaris (Subbat, Thumum, سبط ثمام)

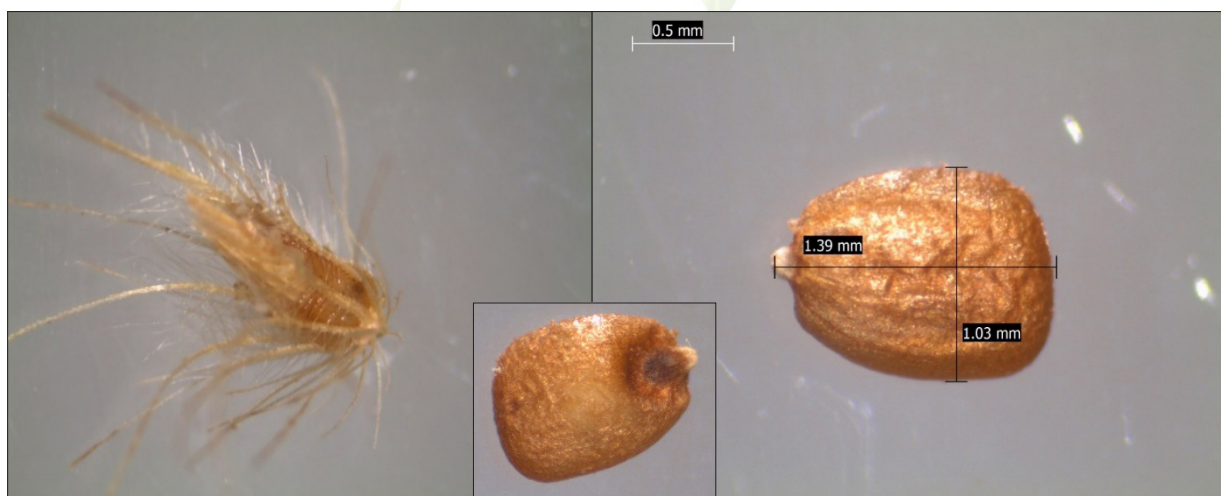
Family: Poaceae



Preferred Method: Germination

The inflorescence of *Cenchrus ciliaris* consists of a cluster of spikelet arranged along the central axis called rachis. Inside each spikelet is a caryopsis, a one-seeded fruit where the seed coat is tightly fused with the fruit wall (Figure 14). The individual seed is called a caryopsis, and the enclosed fruiting structure is called the spikelet (Chakravarty and Kalkani, 2016). The seeds of *C. ciliaris* can be extracted by gently rubbing the spikelet between two layers of sandpaper without damaging the seed. The seeds must be separated from the spikelet to estimate the germination/viability, as not all spikelet contain seeds. The seed filling percentage, calculated as $(\text{number of seeds extracted}/100 \text{ spikelet}) * 100$, provides an indication on the quality of the seed lot. Dry seeds extracted from spikelet should be sown on Petri plates lined with moistened filter paper. The seeds begin to germinate within 24 hours (Figure 15) and the germination percentage can be calculated as the viability percentage. Recording of germination can be continued for up to a week (Appendix A).

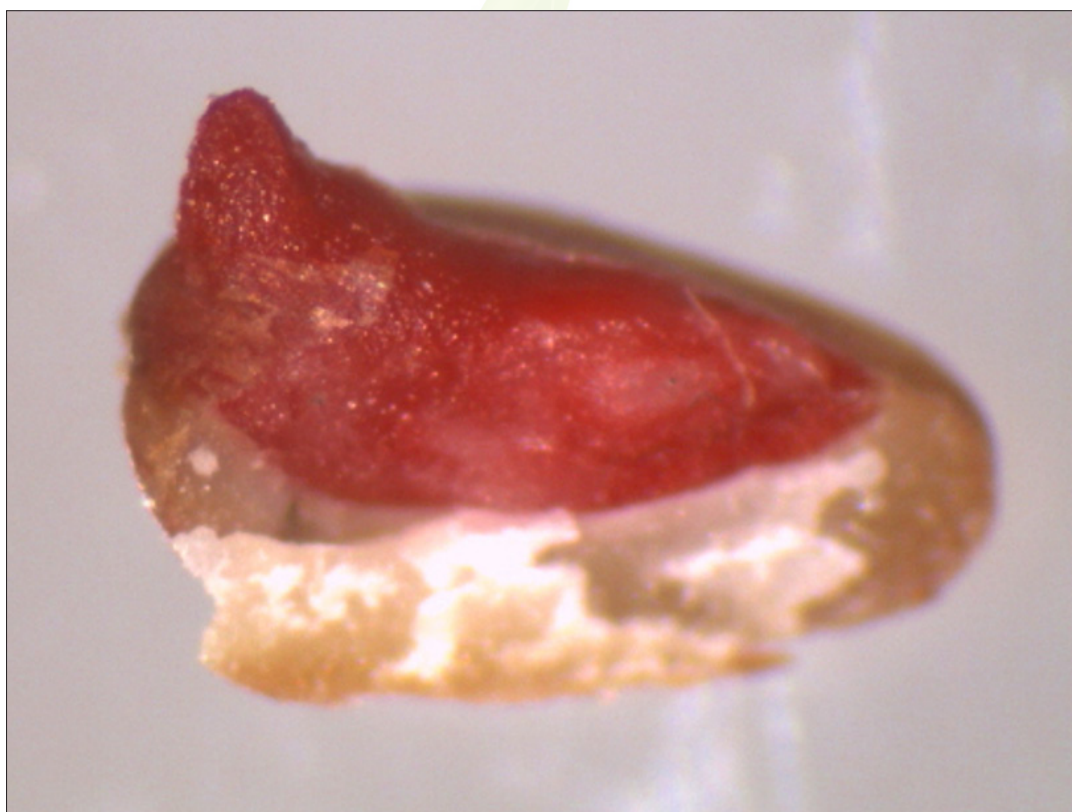
For TTC testing, dehusked and pre-soaked seeds should be given a slanting cut to expose the embryo and soaked in 1% TTC solution. The stained seeds can be used to determine viability (Figure 16).



► Figure 14. Spikelet and extracted seed of *Cenchrus ciliaris*.



► Figure 15. Germinated seed of *Cenchrus ciliaris*.



► Figure 16. Stained seed of *Cenchrus ciliaris* after TTC test.

Convolvulus oxyphyllus (Ethris, Udhris, عضرس)

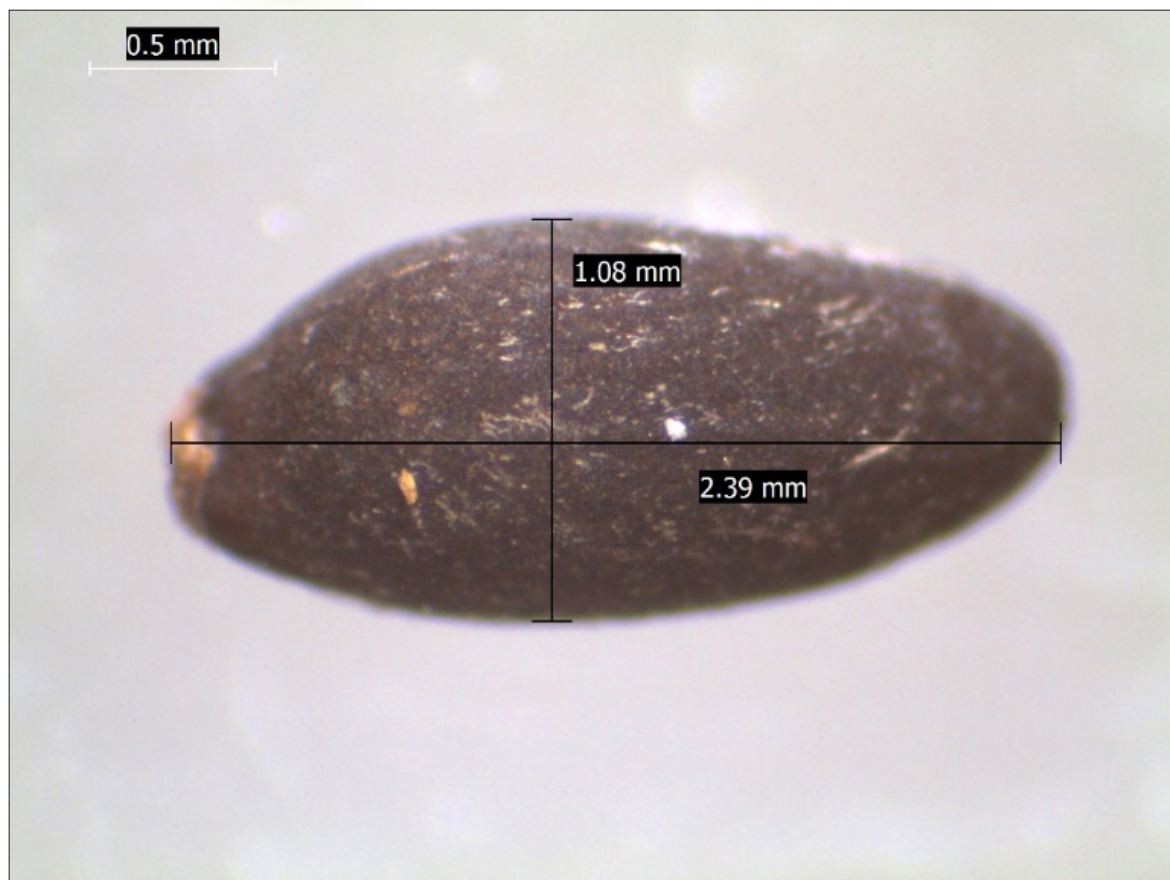
Family: Convolvulaceae



Preferred Method: Germination

Convolvulus oxyphyllus seeds possess a hard seed coat (physical dormancy) that requires scarification (Figure 17). Before placing the seeds on moistened filter paper for germination, they must be scarified using sandpaper. The scarified seeds start to germinate within 3 hours and the germination will continue over the course of a week (Figure 18).

For the TTC test, the scarified seeds should be soaked for 1.5 hours and cut longitudinally prior to soaking in 1% TTC solution. The embryo has folded, bi-lobed cotyledons. The seeds should be kept at room temperature for 24 hours. After incubation, the stained seeds should be rinsed and examined under a microscope to assess their viability (Figure 19).



► Figure 17. Seeds of *Convolvulus oxyphyllus*.



► **Figure 18.** Stained viable seed of *Convolvulus oxyphyllus* (with folded cotyledons).



► Figure 19. Stained viable seed of *Convolvulus oxyphyllus* (with folded cotyledons).

***Deverra triradiata* (Hazhas, حَزَز ذَنِيَّات الجدي)**

Family: Apiaceae

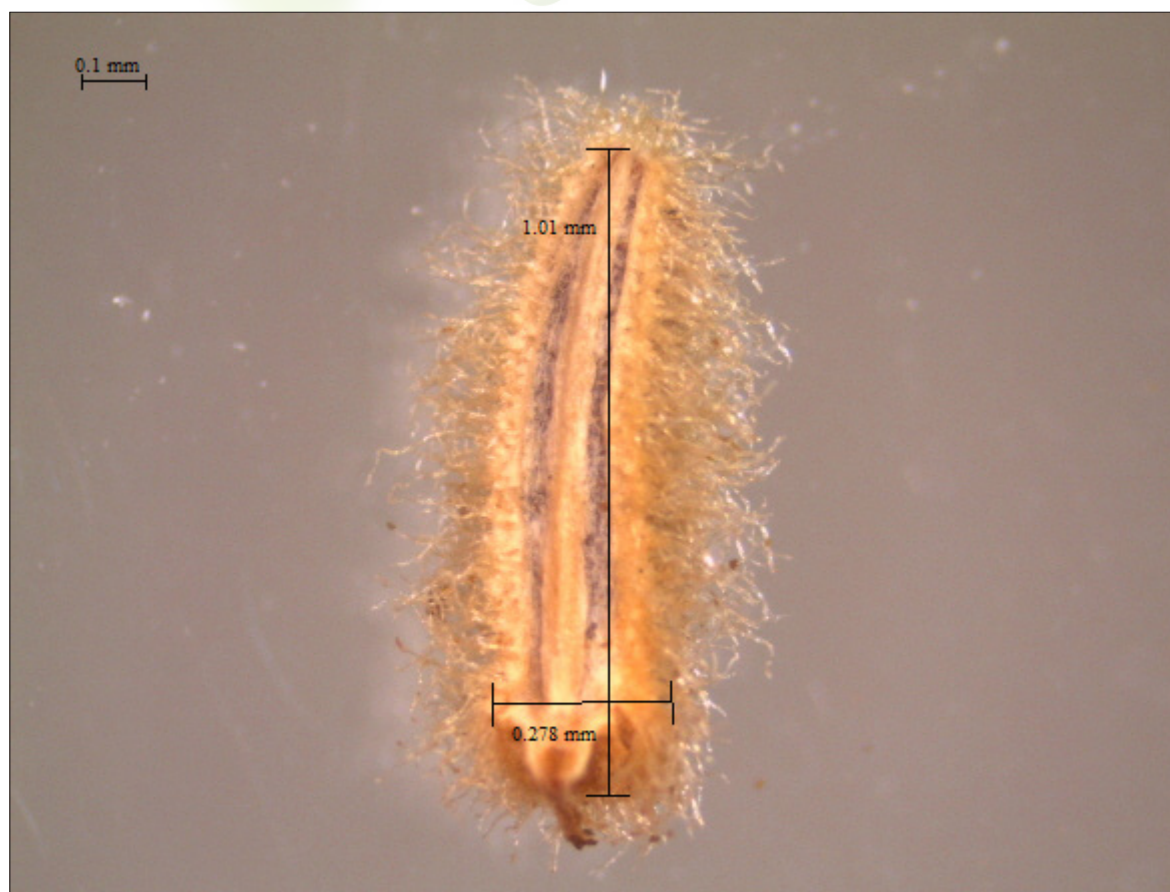


Preferred Method: TTC

This species has a schizocarp fruit with separate indehiscent one seeded mericarp. Each mericarp has three primary ridges with resinous canals, 'vitae', between them (Figures 20 and 21). It is recommended to use a blade for cutting the mericarp of *Deverra triradiata* due to its ease of handling. After soaking the mericarp in water overnight, it should be cut longitudinally to expose the embryo (Figures 22) and treated with 1% TTC for 24 hours. The following day, the stained seeds should be counted to determine viability (Figures 23).



► **Figure 20. Seeds of *Deverra triradiata*.**



► Figure 21. Mericarp of *Deverra triradiata*.

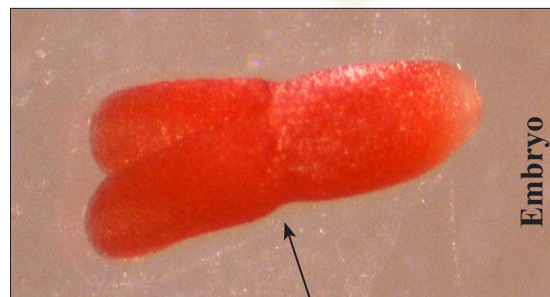
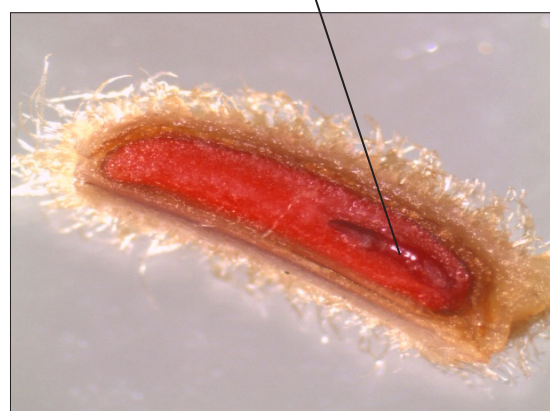


Dry Mericarp



Soaked Mericarp

▲ Figure 22. Dry and soaked mericarp of *Deverra triradiata*.



Embryo

▲ Figure 23. Unstained and stained viable mericarp of *Deverra triradiata*.

Farsetia aegyptia (Al Lbanah, اللبانة)

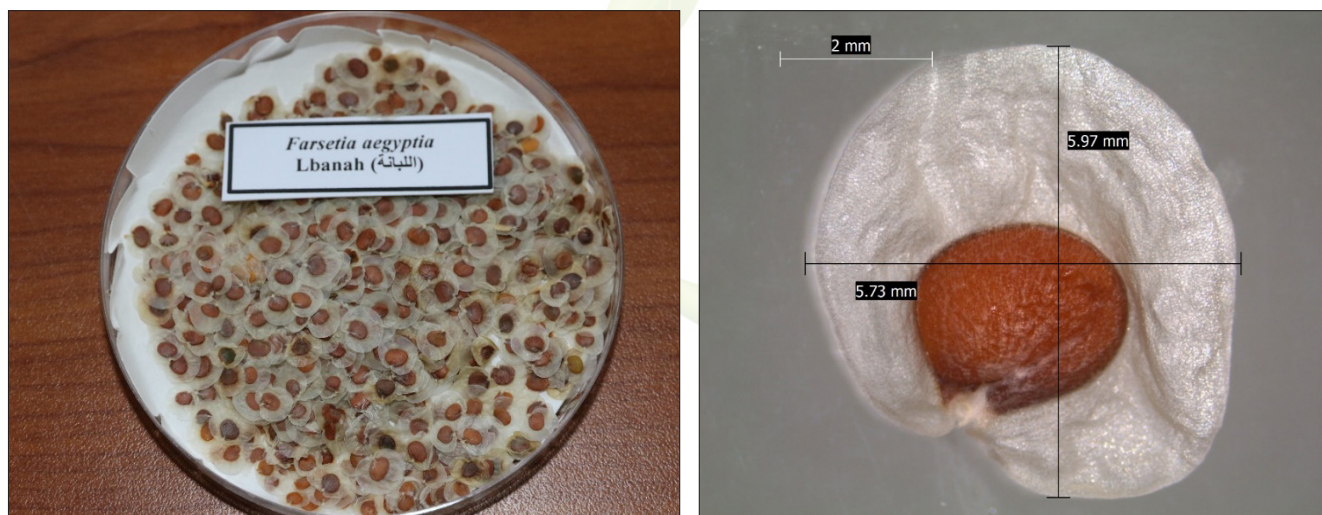
Family: Brassicaceae



Preferred Method: Germination

The seeds of *Farsetia aegyptia* are circular and flattened, surrounded by a membranous, transparent wing (Figure 24), which becomes soggy when soaked in water. Due to the natural red color of the seeds, estimating viability is easier through a germination test rather than TTC. There is no need to soak the seeds before the germination test. The seeds should be sown in Petri dishes lined with either moistened filter paper or sand, and the germination be recorded to assess the viability. Germination begins within 24 hours and continues for up to a week (Figure 25).

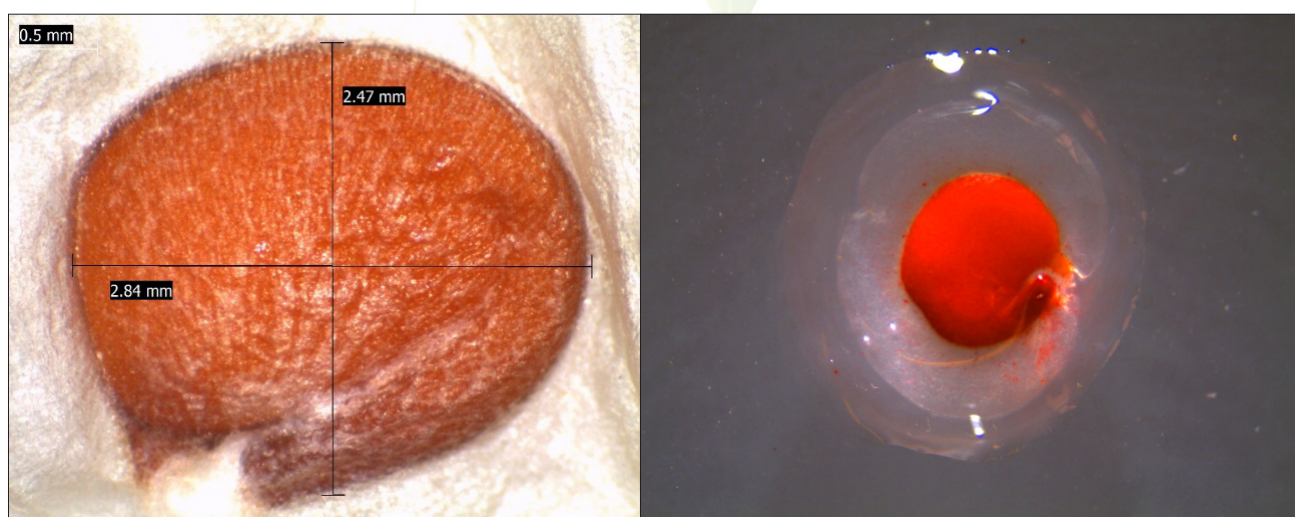
If the TTC test has to be conducted for this species, dry seeds should be soaked directly in 1% TTC solution for 24 hours and observed for staining (Figure 26). There is no need to pre-moisten the seeds prior to TTC treatment.



► Figure 24. Winged seed of *Farsetia aegyptia*.



► Figure 25. Germinated seedling of *Farsetia aegyptia*.



► Figure 26. Dry and stained viable seed of *Farsetia aegyptia*.

***Halocnemum strobilaceum* (Theluth, ثليث)**

Family: Amaranthaceae

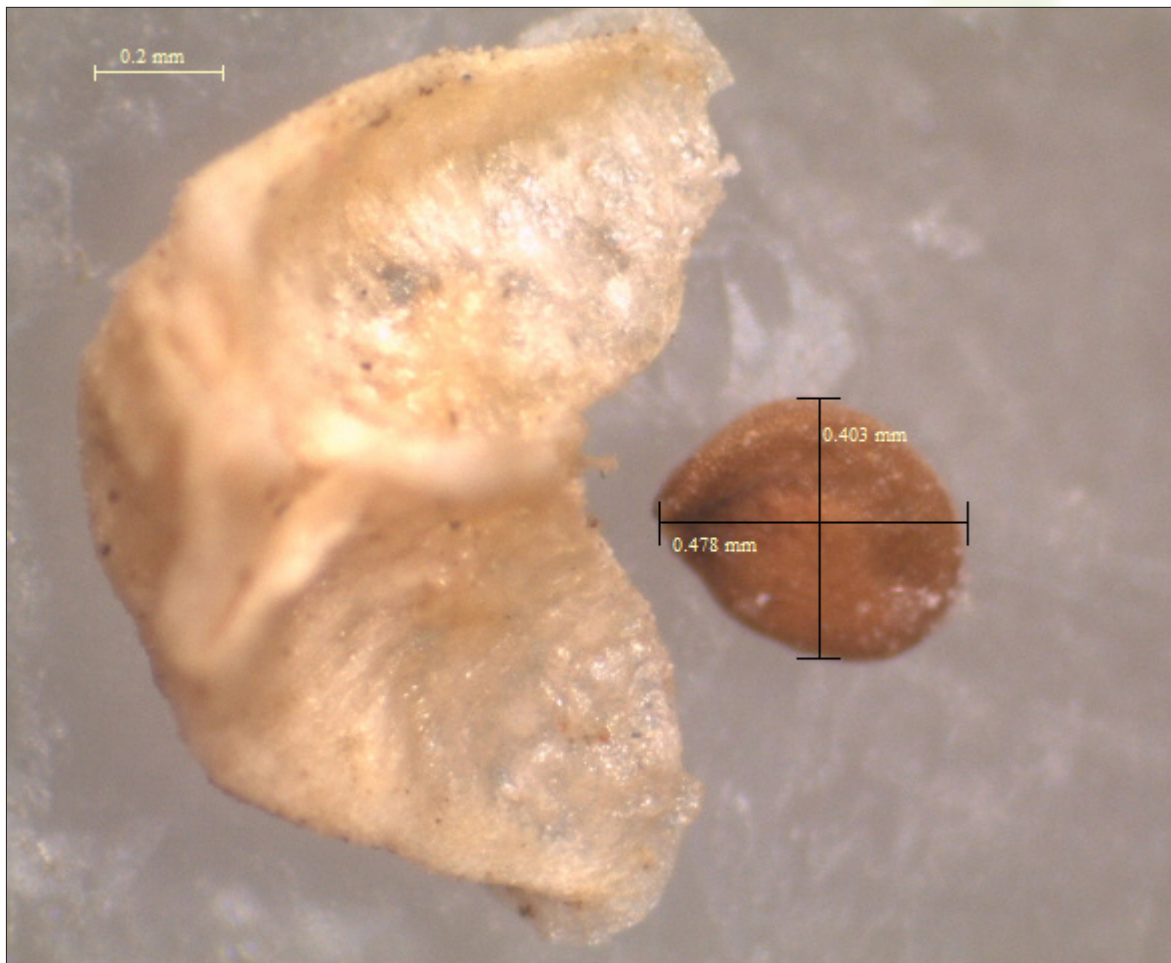


Preferred Method: Germination

As the seeds of *Halocnemum strobilaceum* are non-dormant and the seed size is very small (Figures 27 to 29), it would be easier to assess its viability through the germination method rather than TTC. The seeds should be germinated in a Petri dish lined with moistened filter paper and incubated at 25°C with a 12-hour photoperiod (Figure 30).



► **Figure 27. Seeds of *Halocnemum strobilaceum*.**



► Figure 28. Seed of *Halocnemum strobilaceum*.



► Figure 29. Dry and imbibed seed of *Halocnemum strobilaceum*.



► **Figure 30. Germinated seed and seedling of *Halocnemum strobilaceum*.**

***Halothamnus iraquensis* (Qadqad, قضااض), *Haloxylon salicornicum* (Al Rimth, الرمث), *Salsola imbricata* (Mulaih, Gathraf, مليح, خذراف), and *Seidlitzia rosemarinus* (Al Shinan الشنان)**

Family: Amaranthaceae



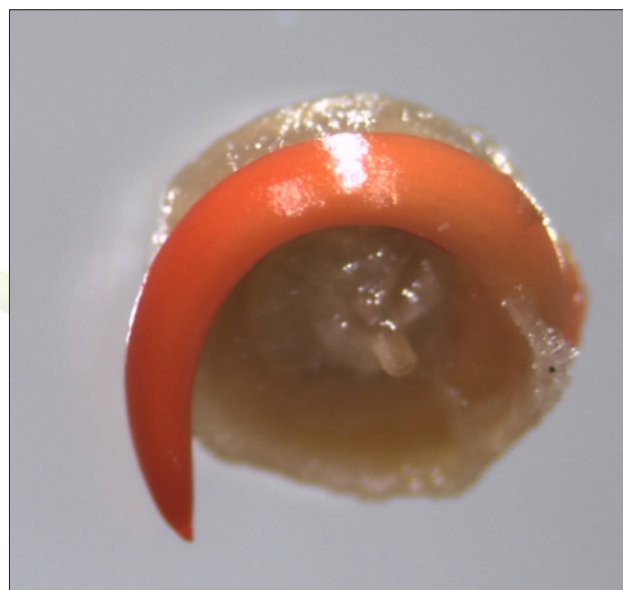
Preferred Method: Germination

Seed structure and germination behavior of *Halothamnus iraquensis* (Figures 31 and 32), *Haloxylon salicornicum* (Figure 33), *Salsola imbricata* (Figure 34) and *Seidlitzia rosemarinus* (Figure 35 and 36) are similar. They have winged perianth with seeds inside, where the coiled embryo is encased in a membranous utricle. Previous studies indicate that the de-winged perianths result in better germination (Bhatt et al., 2019), as the wings can act as a physical barrier to radicle emergence. Hence, it is recommended to use de-winged perianths for testing the viability through germination. The perianths can be de-winged by carefully rubbing them between two sandpapers without damaging the embryo.

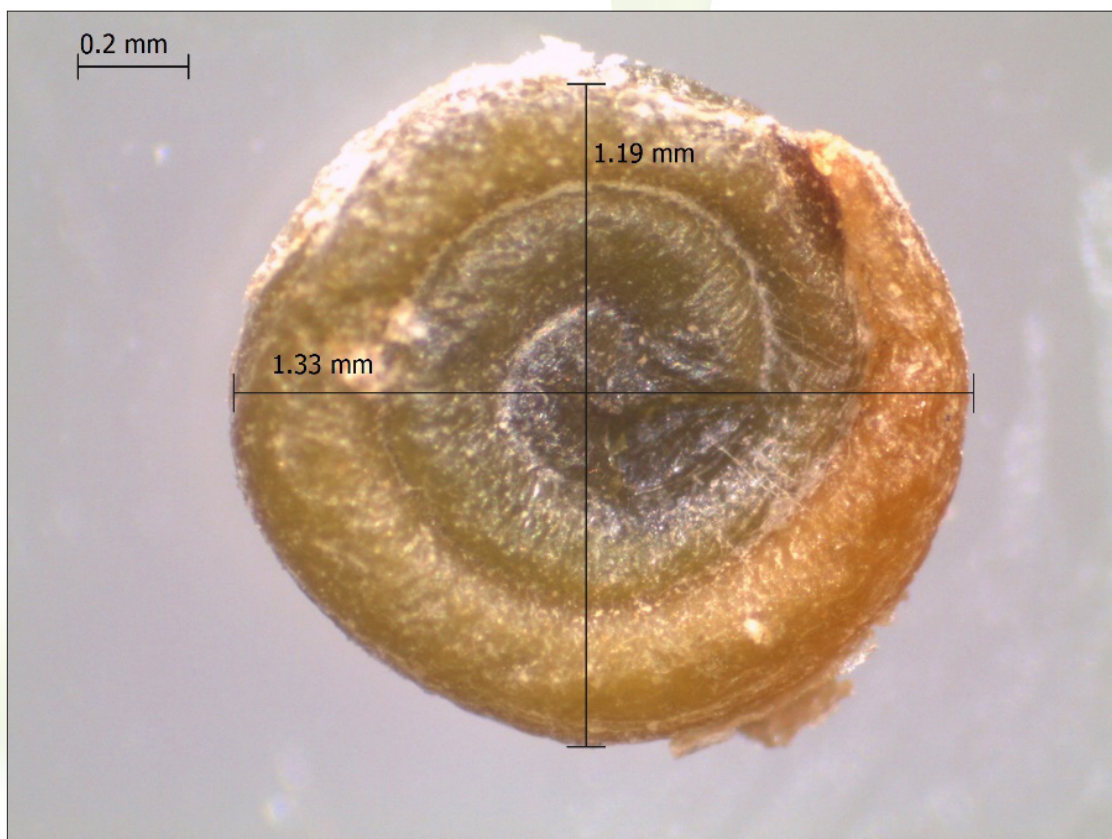
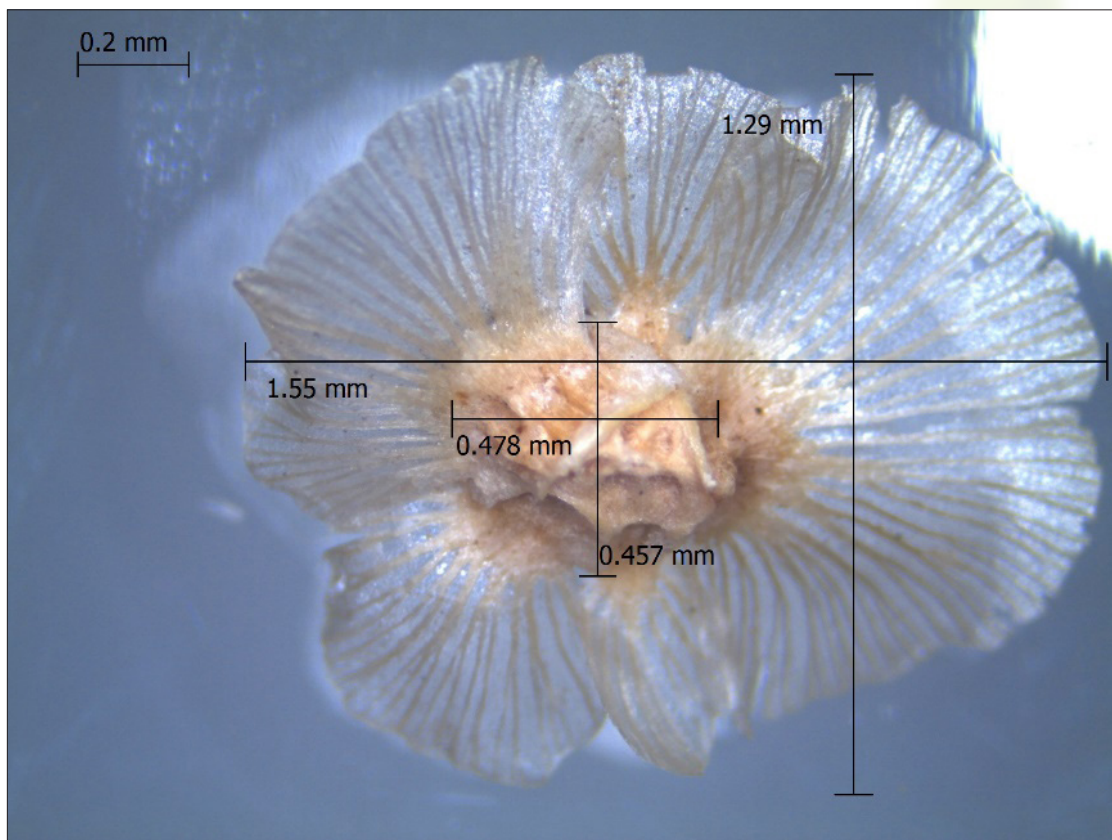
The dry de-winged perianths of these species should be sown on moist sand in a Petri dish, and germination should be recorded to assess the viability. Germination will start within 24 hours and continue for up to a week. The preferred conditions are a temperature of 15/20°C with a 12-hour photoperiod.

If the TTC method is used, the presoaked, de-winged perianths should be given a transverse cut and soaked in 1% TTC in Petri dishes, covered with aluminum foil, for 24 hours. Staining should then be checked to calculate viability. However, it is easier to determine the viability through the germination method.

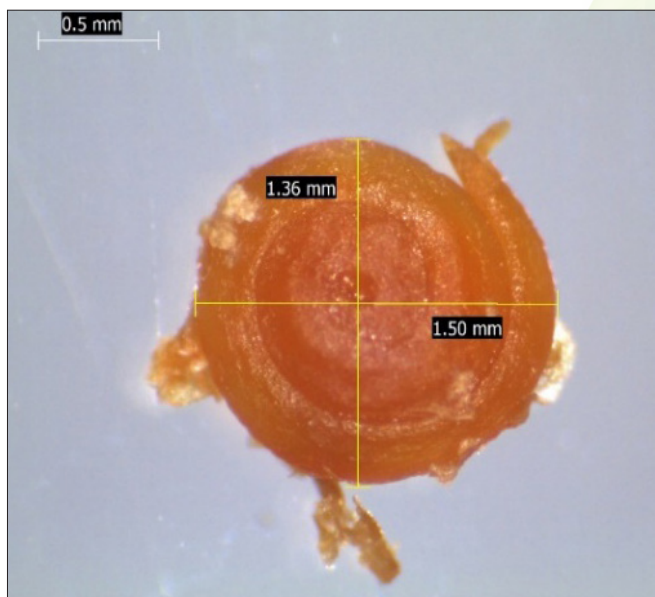
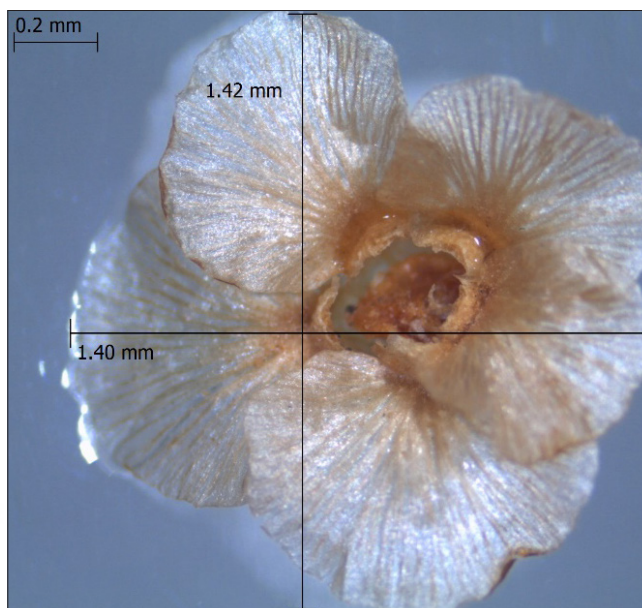
Previous studies indicated that the germination percentage was significantly reduced with prolonged storage and was influenced by the storage temperature (Keblawy, 2013 a). Hence, it is advisable to avoid prolonged storage of these species. Further research is needed to gain deeper insights into this issue.



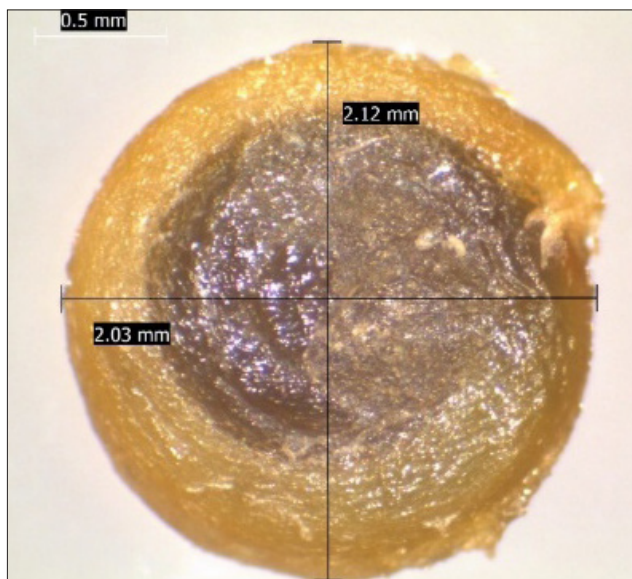
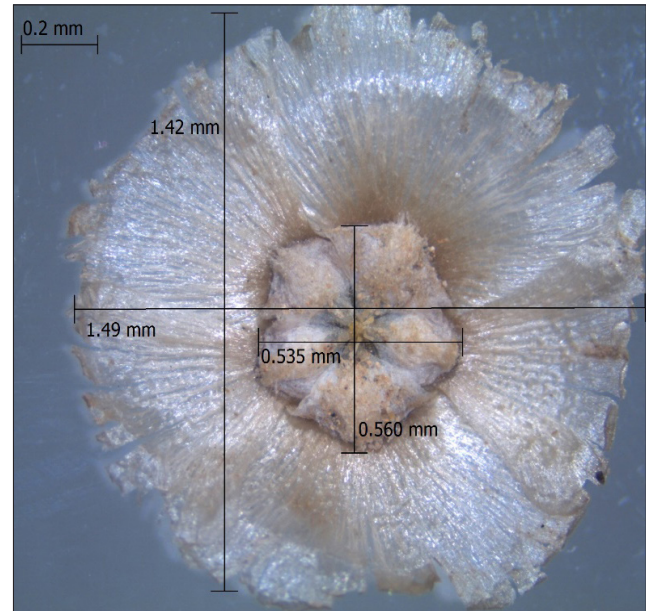
► Figure 31. Dry and stained viable seed of *Halothamnus iraquensis*.



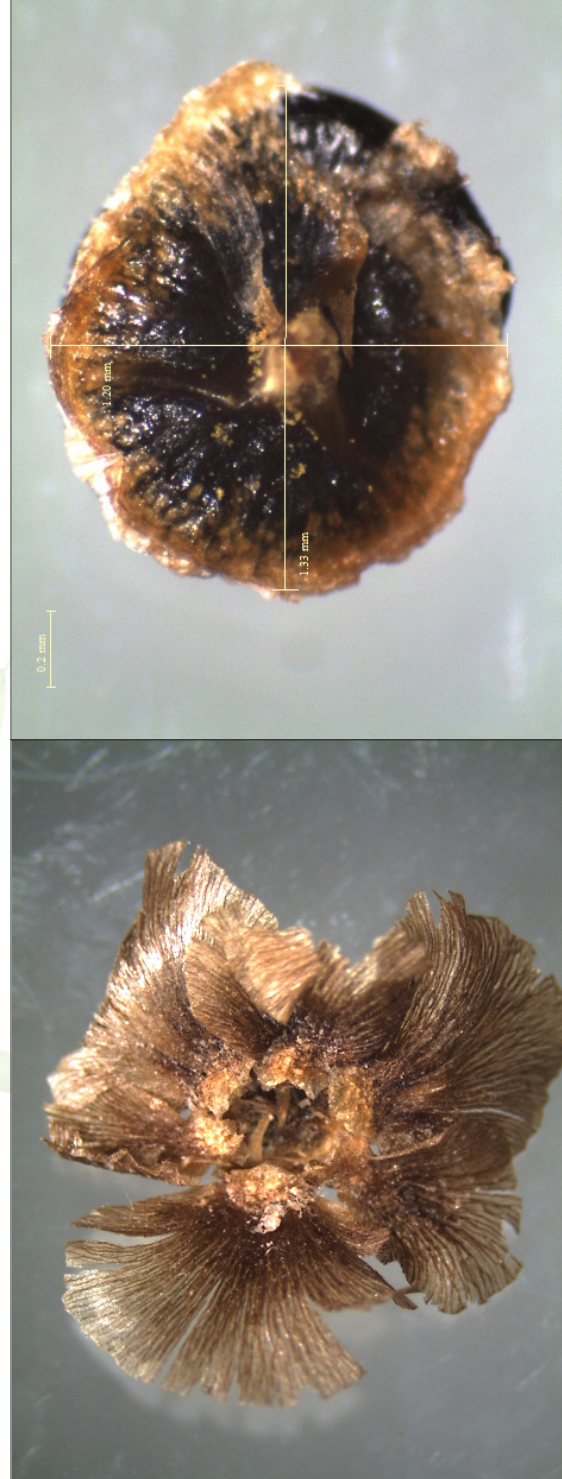
► **Figure 32.** *Halothamnus iraquensis* perianth with and without wings under microscope.



► Figure 33. Dry and stained viable seed of *Haloxylon salicornicum*.



► Figure 34. Dry and stained viable seed of *Salsola imbricata*.



► Figure 35. Winged and de-winged perianth of *Seidlitzia rosmarinus*.



De-winged Dry Perianth



Water Soaked Perianth



Coiled Embryo

Transparent Utricle



► Figure 36. Perianth and coiled embryo of *Seidlitzia rosemarinus*.

Helianthemum lippi (Rag-rug, رقوق)

Family: Cistaceae



Preferred Method: Germination

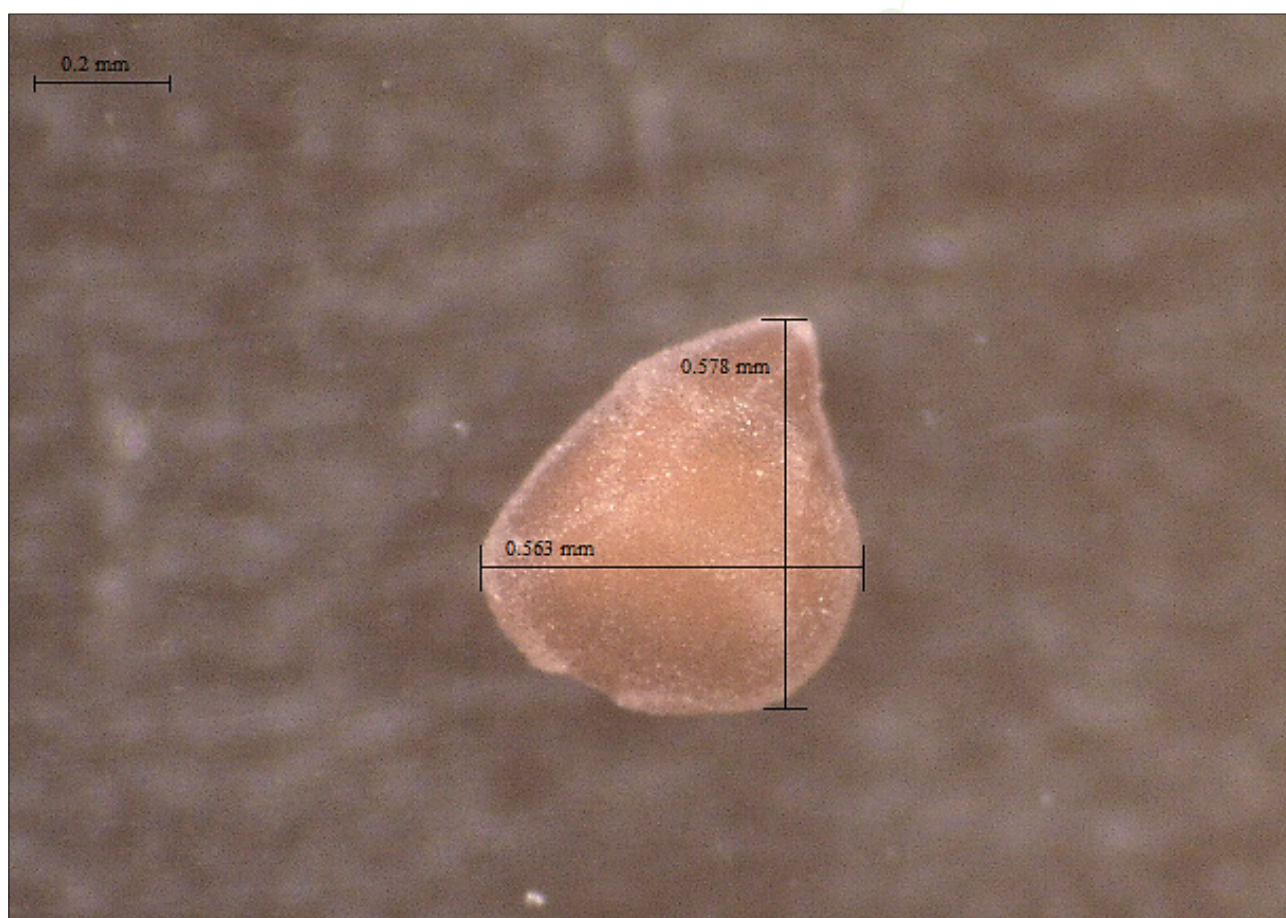
The seeds of *Helianthemum lippi* are extremely small (Figures 37 to 39), making it challenging to dissect them to expose the embryo. It should also be noted that this species exhibits physical dormancy due to a hard seed coat (Zaman et al., 2009). To break the physical dormancy, the seeds should be scarified by rubbing them between two sandpapers. Then, the seeds should be sown in a Petri dish lined with moistened filter paper and incubated at 20⁰C under laboratory conditions. Germination is usually initiated within 24 hours (Figure 40). When the seeds come in contact with water, a waxy material may be observed on the seed coat (Figure 38).



► Figure 37. Seeds of *Helianthemum lippi*.



► Figure 38. Dry and imbibed seeds of *Helianthemum lippi*.



► Figure 39. Seed of *Helianthemum lippi*.



▲ Figure 40. Germinated seed and seedling of *Helianthemum lippi*.

Horwoodia dicksoniae (Khuzamah, خزامة)

Family: Brassicaceae



Preferred Method: TTC

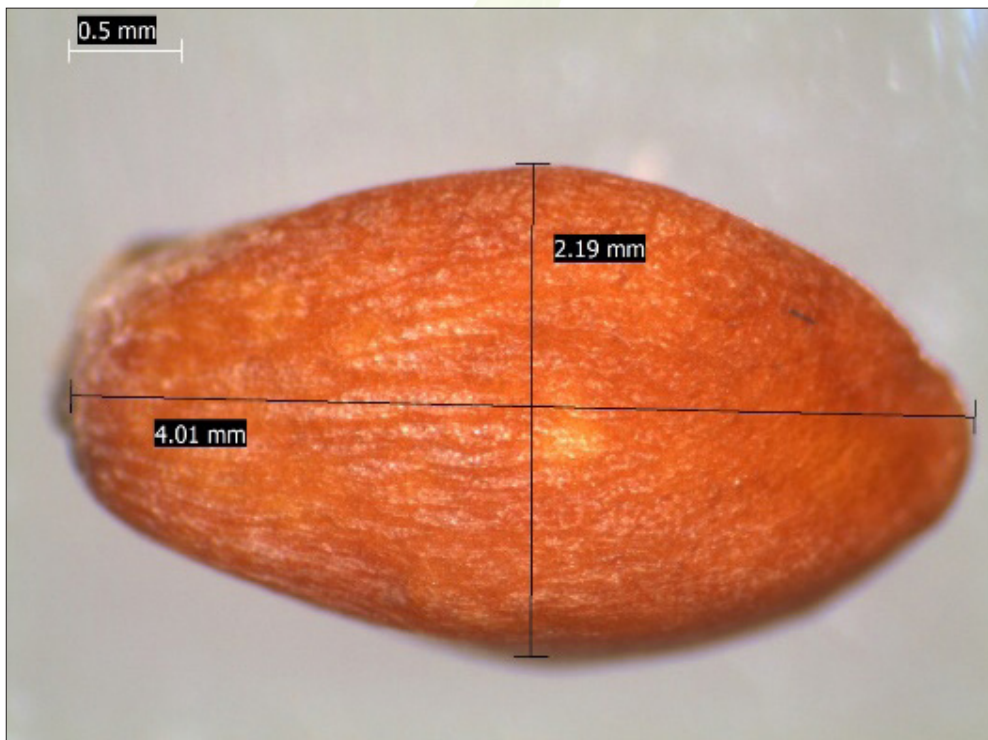
The seeds of *Horwoodia dicksoniae* exhibit physiological dormancy (Suleiman et al., 2009a). To conduct the TTC test, the seeds should first be extracted by splitting open the hard-textured winged fruit (Figures 41 to 43). The seeds should be soaked in distilled water overnight, and the embryos should be exposed by making a longitudinal cut. The seeds should be soaked in 1% TTC solution in Petri dishes covered with aluminum foil and incubated at room temperature for 24 hours (20°C). After incubation, the seeds should be washed thoroughly with distilled water to remove excess stain and examined under a microscope (Figure 44).



► Figure 41. Winged fruit of *Horwoodia dicksoniae*.



► Figure 42. *Horwoodia dicksoniae* seed extracted from the winged fruit.



► Figure 43. *Horwoodia dicksoniae* seed.



► **Figure 44. Stained seed of *Horwoodia dicksoniae*.**

Lycium shawii (Awsaj, العوسج)

Family: Solanaceae



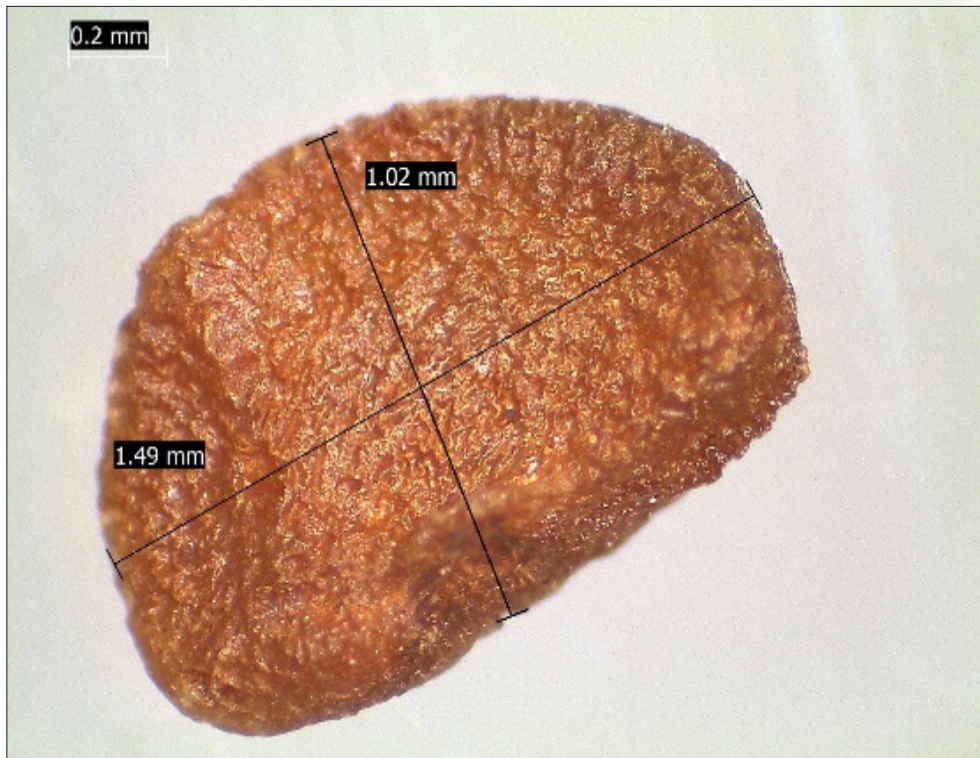
Preferred Method: Germination

Viability of *Lycium shawii* seeds can be effectively tested using the germination method. The dry seeds should be placed in Petri dishes lined with moistened filter paper, ensuring they are periodically moistened. The germination should be recorded daily. Maximum germination is typically observed within 15 days. Although the TTC test can also be used to determine viability, the germination test is preferred due to the small size of the seeds (Figures 45 to 48).

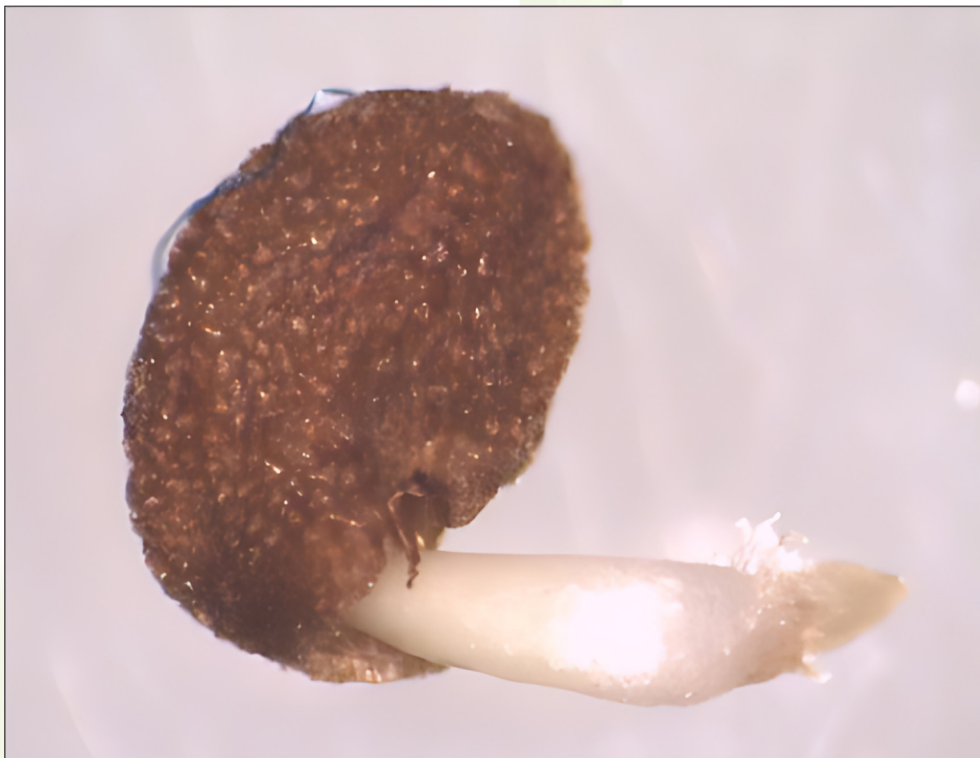
TTC Method: If the TCC method is used to determine viability, the pre-soaking duration of the seeds has to be reduced to 4-6 hours, as the seeds germinate quickly (please refer to the section on Viability Assessment through the TTC Method for detailed methodology).



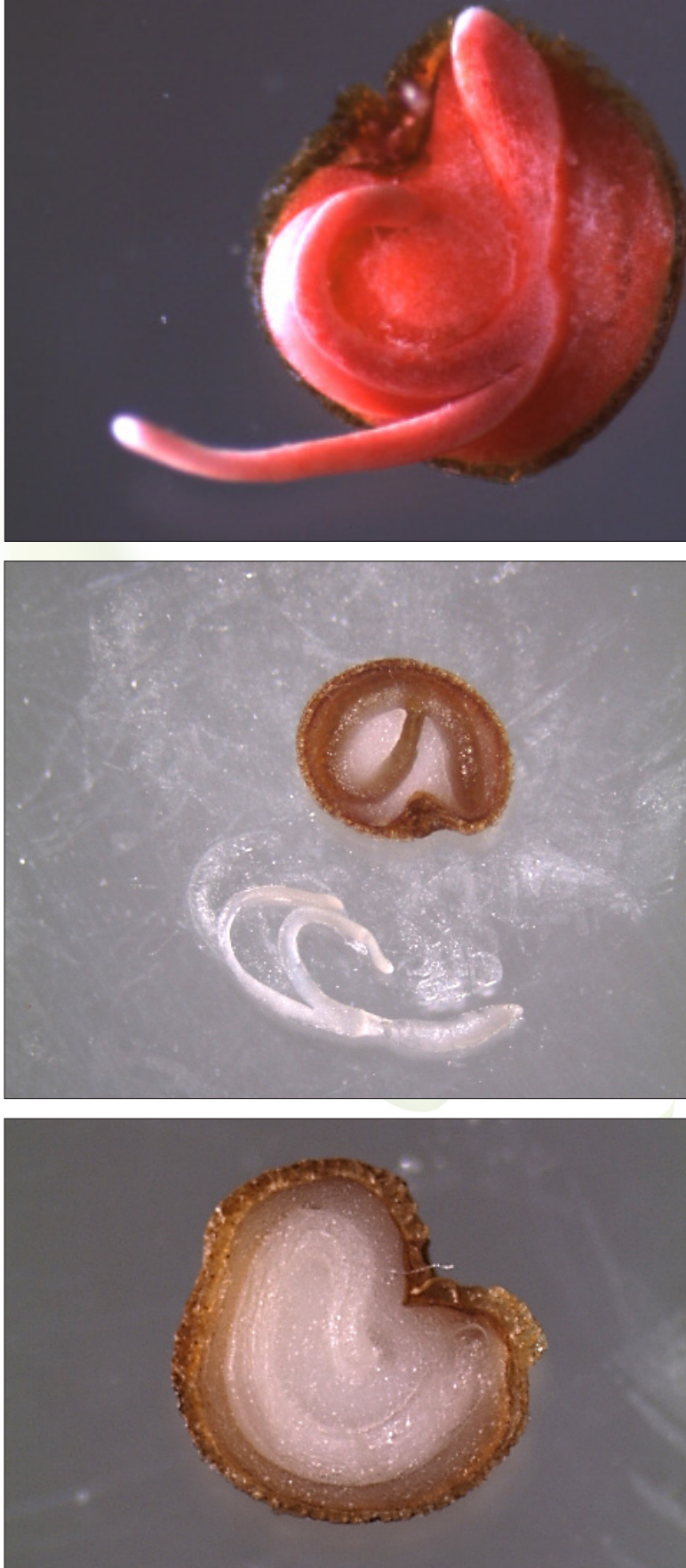
► **Figure 45. Seeds of *Lycium shawii*.**



► Figure 46. Microscopic view of *Lycium shawii* seed.



► Figure 47. Germinated seed of *Lycium shawii*.



▶ Figure 48. Unstained and stained seed of *Lycium shawii*.

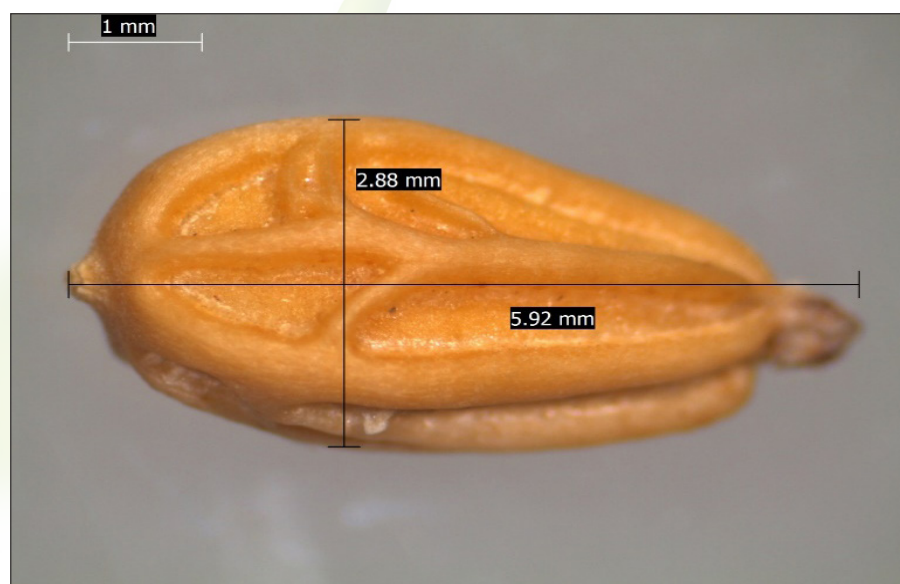
***Nitraria retusa* (Ghardaq, الغردق)**

Family: Nitrariaceae



Preferred Method: TTC

The seeds of *Nitraria retusa* should be soaked in distilled water overnight, and the embryo of pre-moistened seeds should be exposed by splitting the seed with a longitudinal cut. Then, the exposed seeds should be soaked in 1% TTC solution in Petri dishes covered with aluminum foil and incubated at room temperature for 24 hours (20°C). After incubation, the seeds should be washed thoroughly with distilled water to remove excess stain and examined under the microscope (Figures 49 and 50).



► **Figure 49. Seeds of *Nitraria retusa*.**



► **Figure 50. Stained seeds of *Nitraria retusa*.**

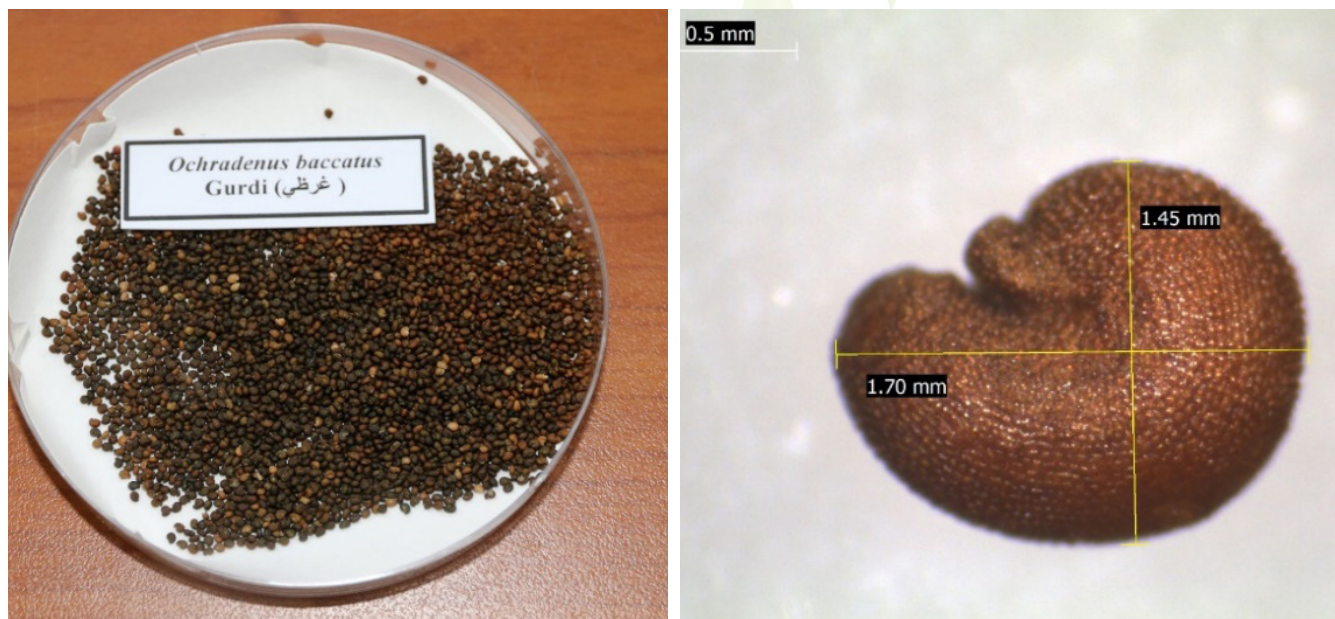
Ochradenus baccatus (Gurdi, غرظي)

Family: Resedaceae

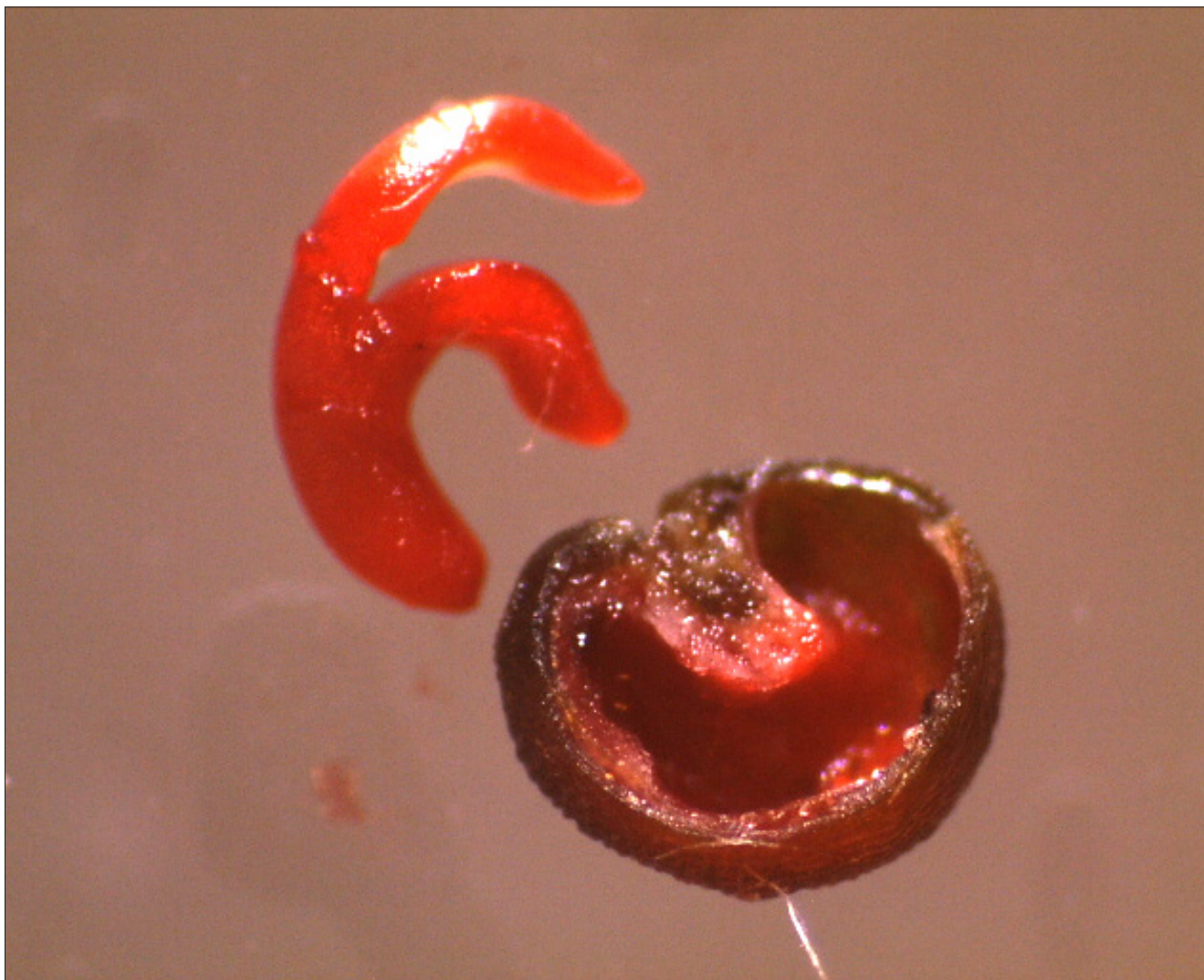


Preferred Method: TTC

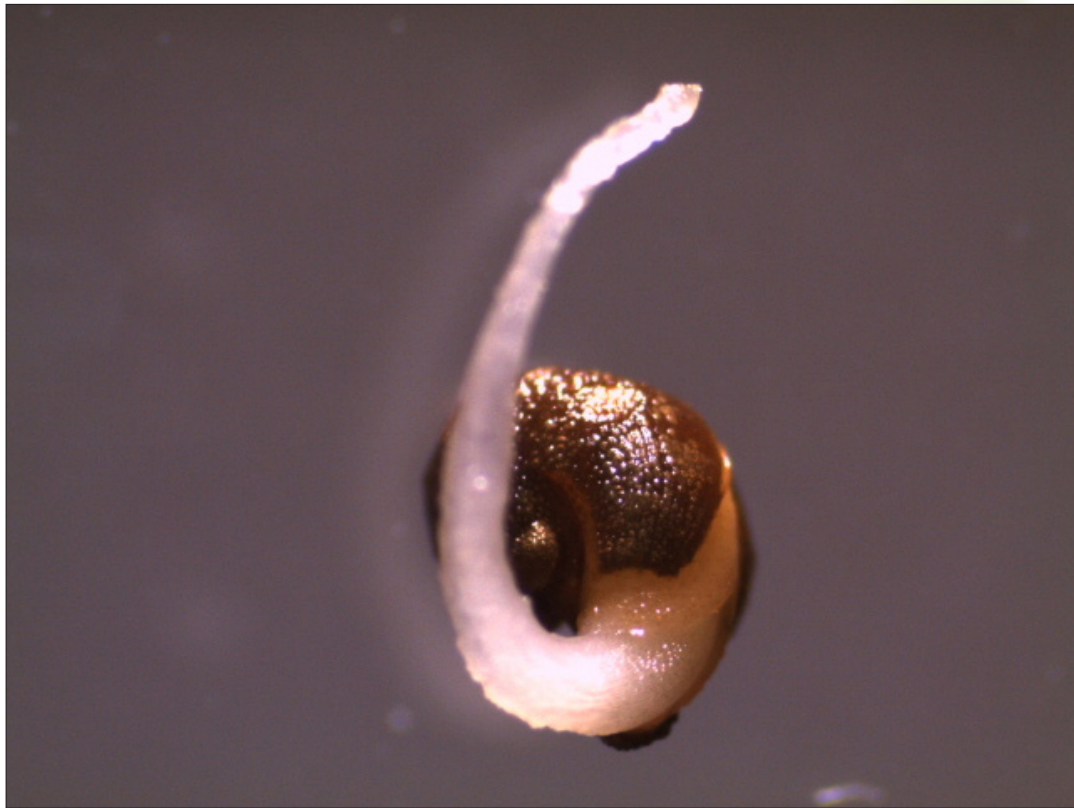
The seeds of *Ochradenus baccatus* exhibit physiological dormancy (Suleiman et al., 2011). Despite their small size (Figure 51), the TTC test is preferred for accurate results. After soaking the seeds in distilled water overnight, they should be cut longitudinally to expose the embryo. Seeds should be soaked in 1% TTC solution in Petri dishes covered with aluminum foil and incubated at room temperature (20°C) for 24 hours. Seeds should then be washed thoroughly with distilled water to remove excess stain and examined under the microscope (Figure 52). An image of germinated seedling is provided below for reference (Figure 53)



► Figure 51. Seeds of *Ochradenus baccatus*.



► Figure 52. Stained seed of *Ochradenus baccatus*.



► **Figure 53. Germinated seedling of *Ochradenus baccatus*.**

Panicum turgidum (Thamam, شمام)

Family: Poaceae



Preferred Method: TTC

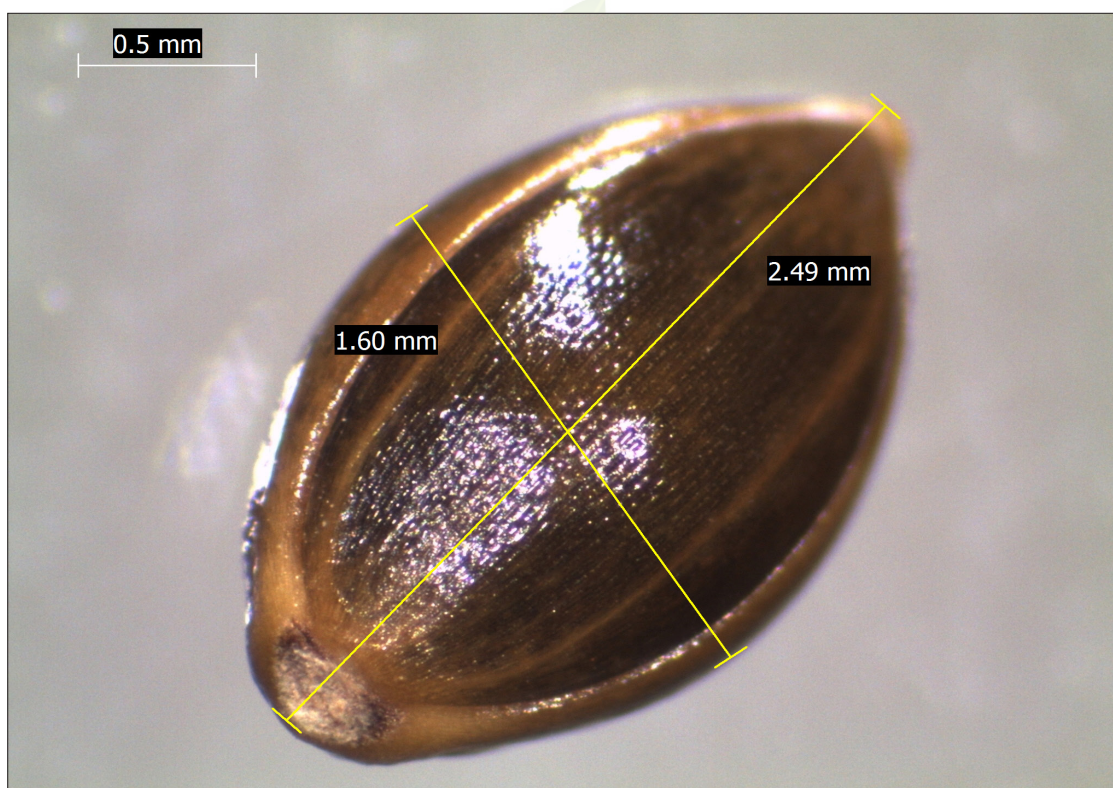
Previous studies suggest that, *Panicum turgidum* exhibits physiological dormancy (Al- Shamsi, 2009; El-Keblawy, 2013 b; Bhatt et al., 2020). The seed coat may also obstruct radicle protrusion (Batanouny et al., 2006), making the TTC test a better option for assessing viability. Pre-soaked seeds should be cut longitudinally and soaked in 1% TTC solution for 24 hours and then observed for staining to determine viability (Figures 54 to 59).



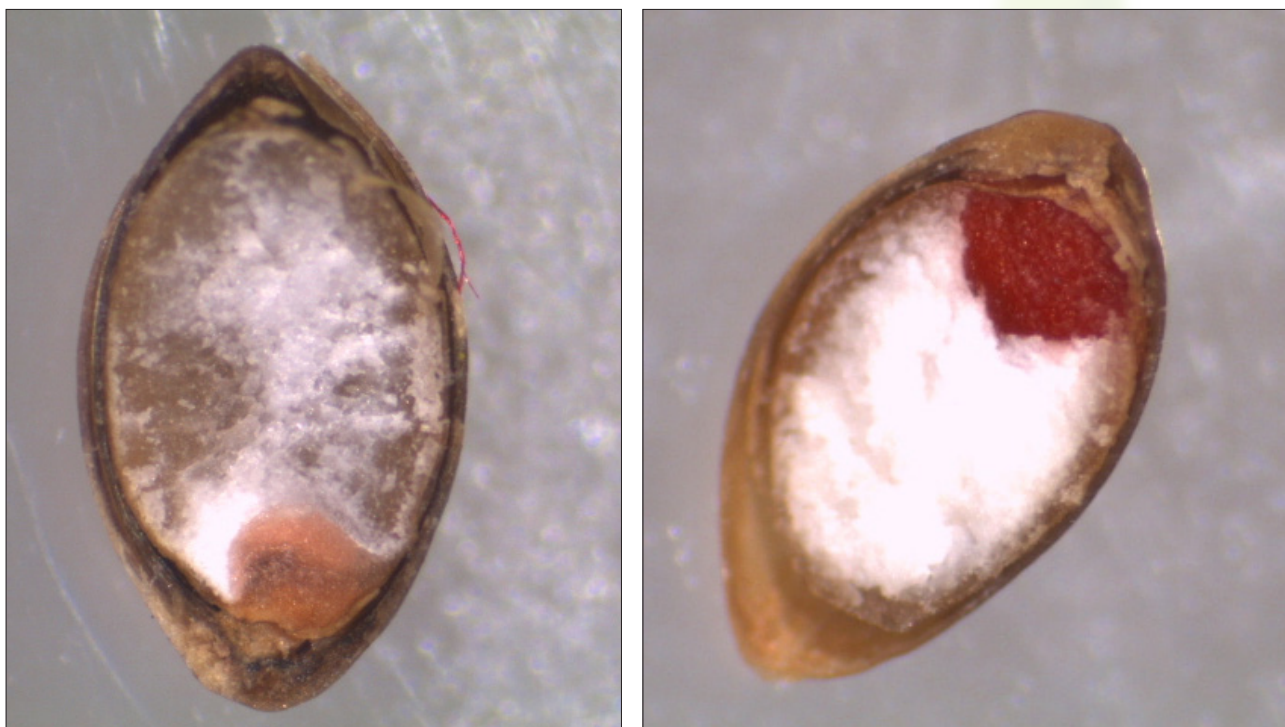
► **Figure 54. Seeds of *Panicum turgidum*.**



► Figure 55. Seed of *Panicum turgidum* enclosed in the husk.



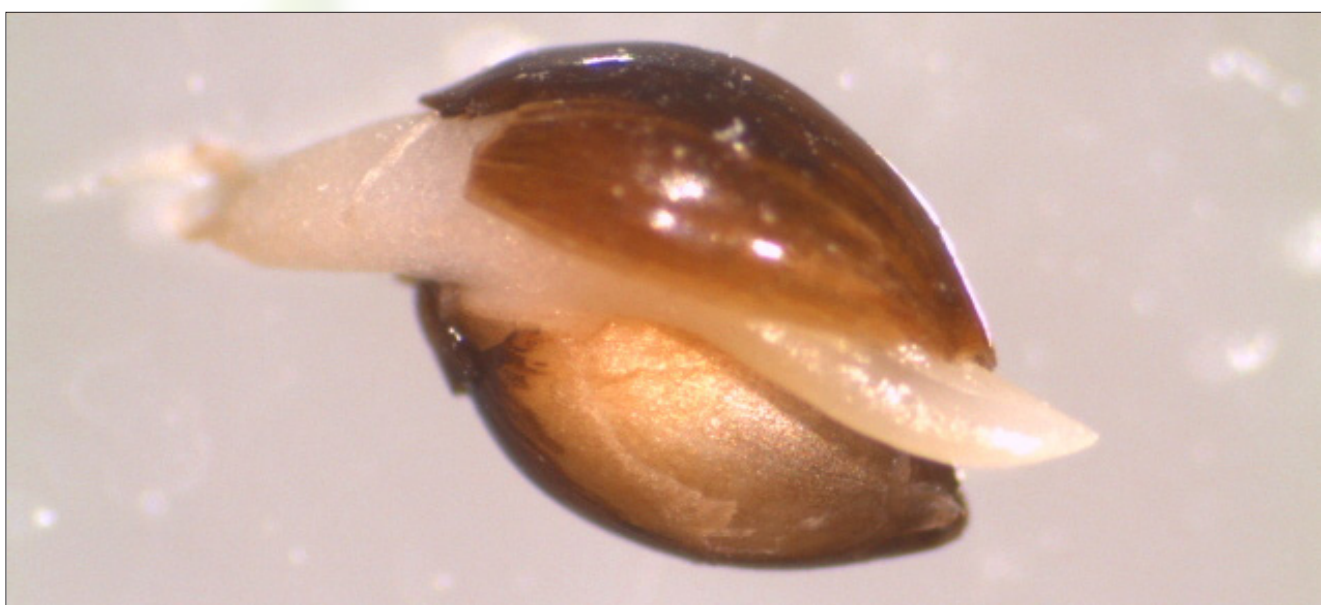
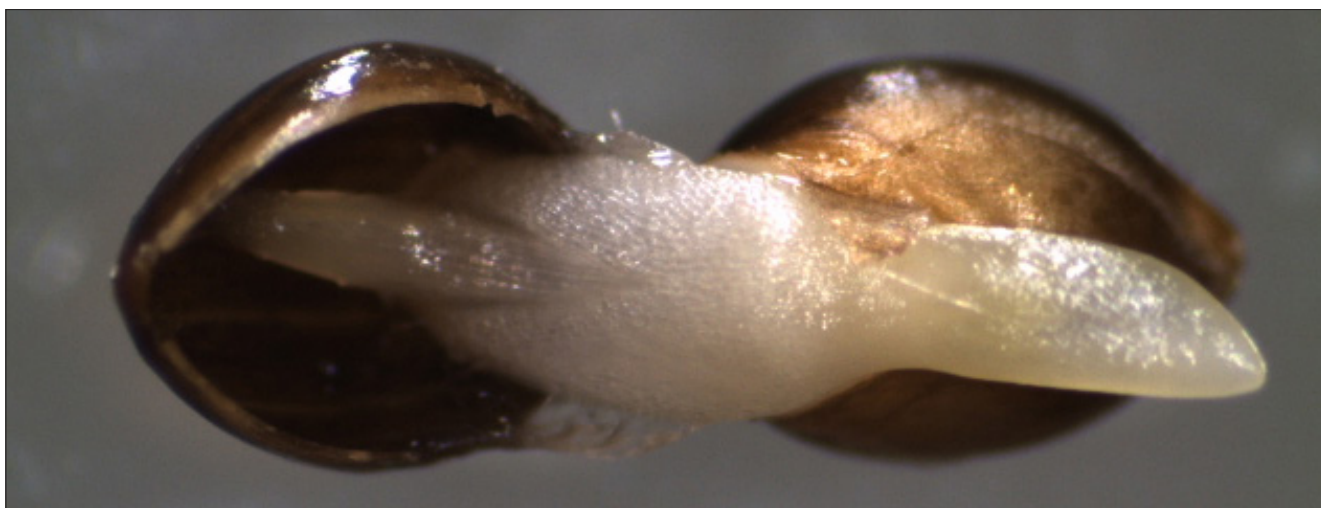
► Figure 56. De-husked seed of *Panicum turgidum*.



► Figure 57. Non-viable and viable seed of *Panicum turgidum*.



► Figure 58. Viable seed of *Panicum turgidum*.



► Figure 59. Germination of *Panicum turgidum*.

Peganum harmala (Harmal, الحرمل)

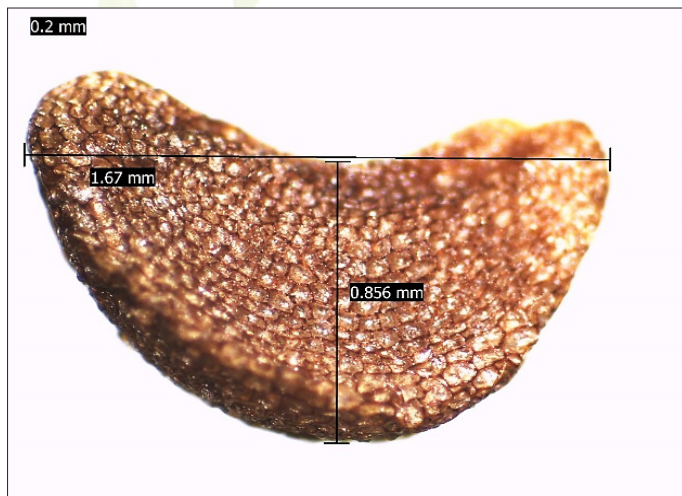
Family: Nitrariaceae



Preferred Method: Germination

As the seeds of *Peganum harmala* (Figure 60) germinate easily, their viability can be easily estimated using the germination method. The seeds should be soaked in water overnight and placed on the moistened filter paper/soil medium in Petri dishes. The germination should be recorded daily (Figure 61). Though the TTC test can also be conducted for this species (Figure 62), the germination test is the easiest, considering the size of the seed.

If viability has to be determined using the TTC method, the pre-soaking duration of the seeds should be reduced to 4-6 hours, as the seeds tend to germinate quickly (please refer to the section on Viability Assessment through TTC Method for detailed methodology).



► Figure 60. Seeds of *Peganum harmala*.



► Figure 61. Germinated seedlings of *Peganum harmala*.



► Figure 62. Stained seed of *Peganum harmala*.

Pennisetum divisum (Thumam, ثمام)

Family: Poaceae



Preferred Method: TTC

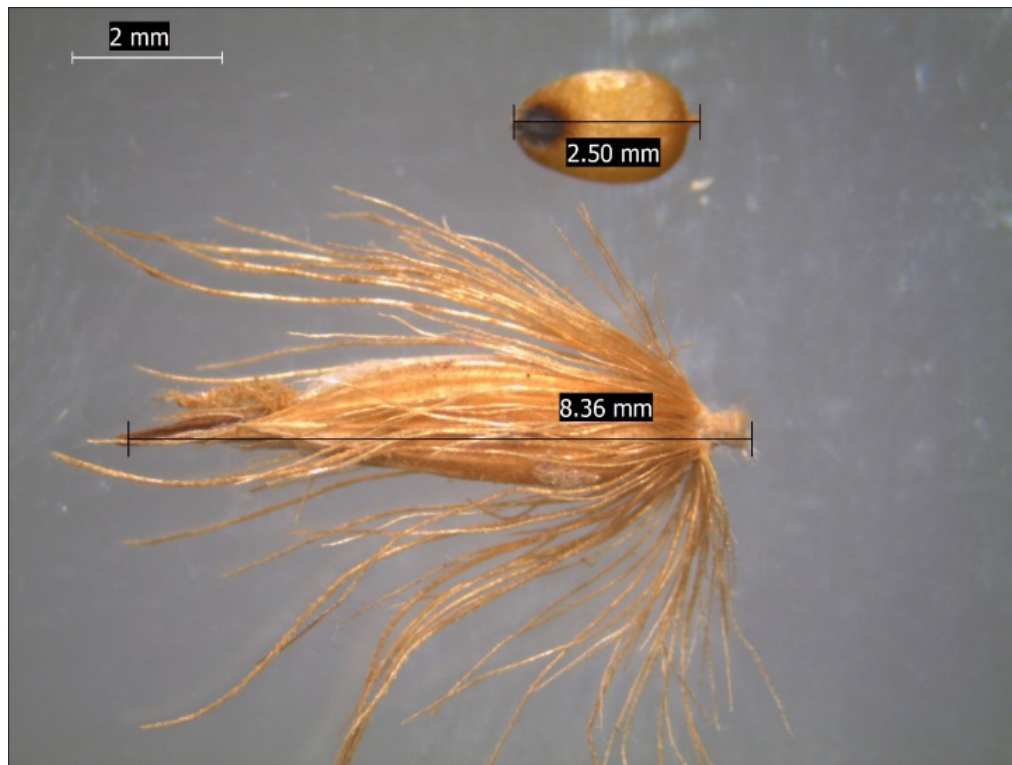
The inflorescence of *Pennisetum divisum* is a cylindrical spike with spikelets (Figures 63 and 64). Before estimating viability, the seed filling should be checked, and seeds should be gently separated from the spikelet by rubbing it between sandpapers without damaging the seeds. The seeds of *P. divisum* germinate easily, and checking the viability through germination is easier. However, Bhatt et al. (2020) noted the presence of physiological dormancy in this species. Hence, it is preferable to use the TTC test for more accurate viability results.

TTC Method: The pre-moistened (for 6 hours) seeds should be carefully cut longitudinally and soaked in 1% TTC for 24 hours (please refer to the section on Viability Assessment through TTC Method for detailed methodology). The staining should be examined under a microscope (Figure 65).

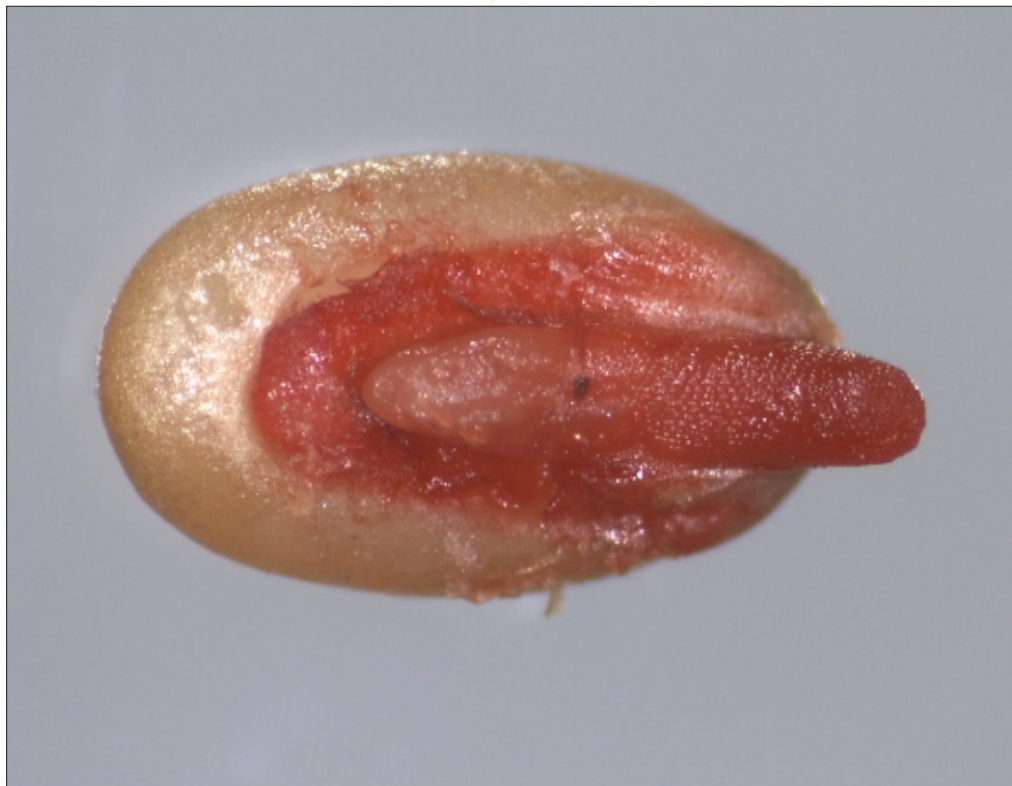
If you wish to check the viability through the germination method, the seeds should be soaked in water overnight and placed on the moistened filter paper in Petri dishes. The germination can be recorded daily (Figure 66).



► Figure 63. *Pennisetum divisum* spikelet and seeds.



► Figure 64. *Pennisetum divisum* seeds extracted from the spikelet.



► Figure 65. Stained seed of *Pennisetum divisum*.



► Figure 66. Germinated seed of *Pennisetum divisum*.

***Prosopis cineraria* (Ghaf, الغاف الرمادية)**

Family: Fabaceae

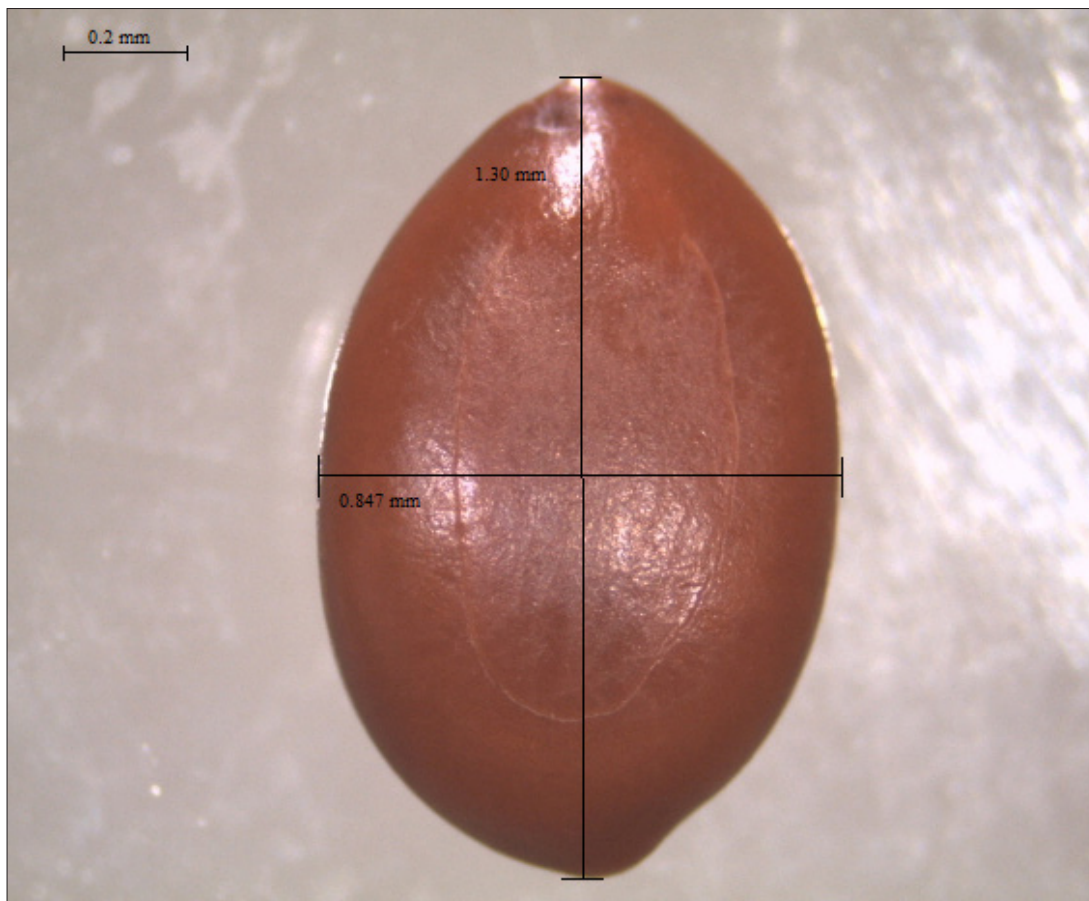


Preferred Method: TTC

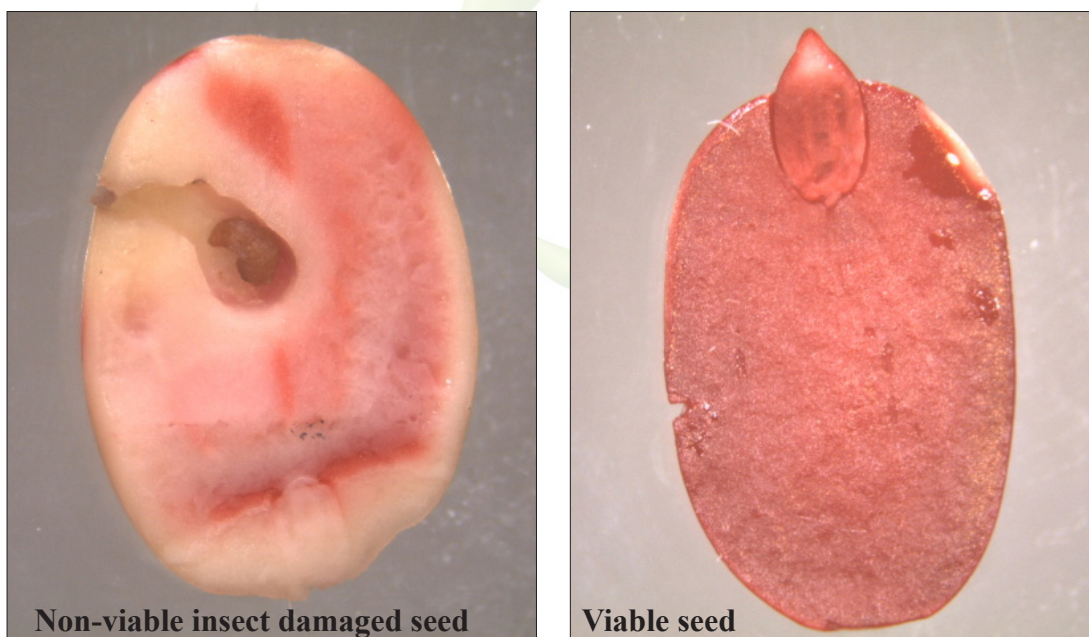
The seeds of *Prosopis cineraria* exhibit physical dormancy, which can be eliminated through careful scarification using sandpaper. The scarified seeds should be pre-soaked for 24 hours and cut longitudinally to expose the embryo. One half of the split seed should then be treated with 1% TTC solution for 24 hours (the other half should be discarded). After 24 hours of TTC treatment, the stained seeds should be counted and the viability can be calculated (Figures 67 to 69). If the viability should be checked with germination method, it is suggested to conduct the test immediately after summer (October-November) in soil medium. Viability through germination method will take 6-7 days to complete.



► **Figure 67. Seeds of *Prosopis cineraria*.**



► Figure 68. Seed of *Prosopis cineraria*.



► Figure 69. Unstained and stained viable embryo of *Prosopis cineraria*.

Rhanterium epapposum (Arfaj, العرفج)

Family: Asteraceae

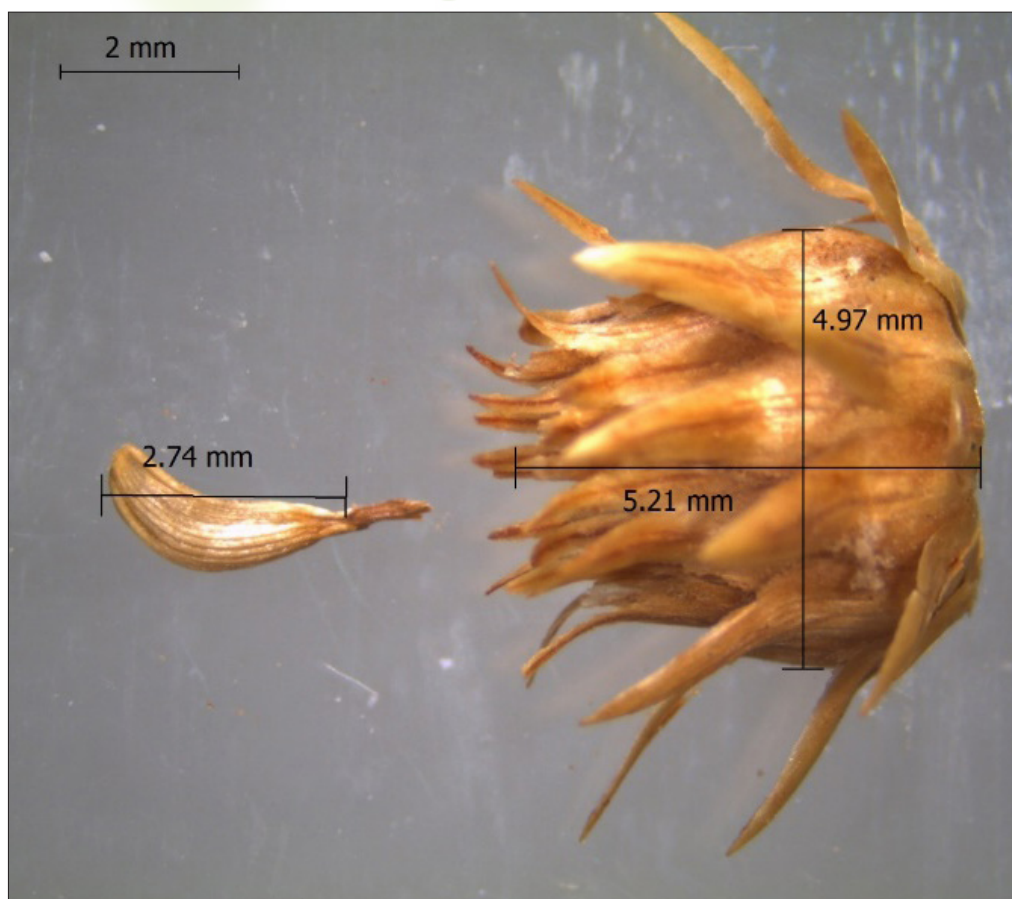


Preferred Method: Germination

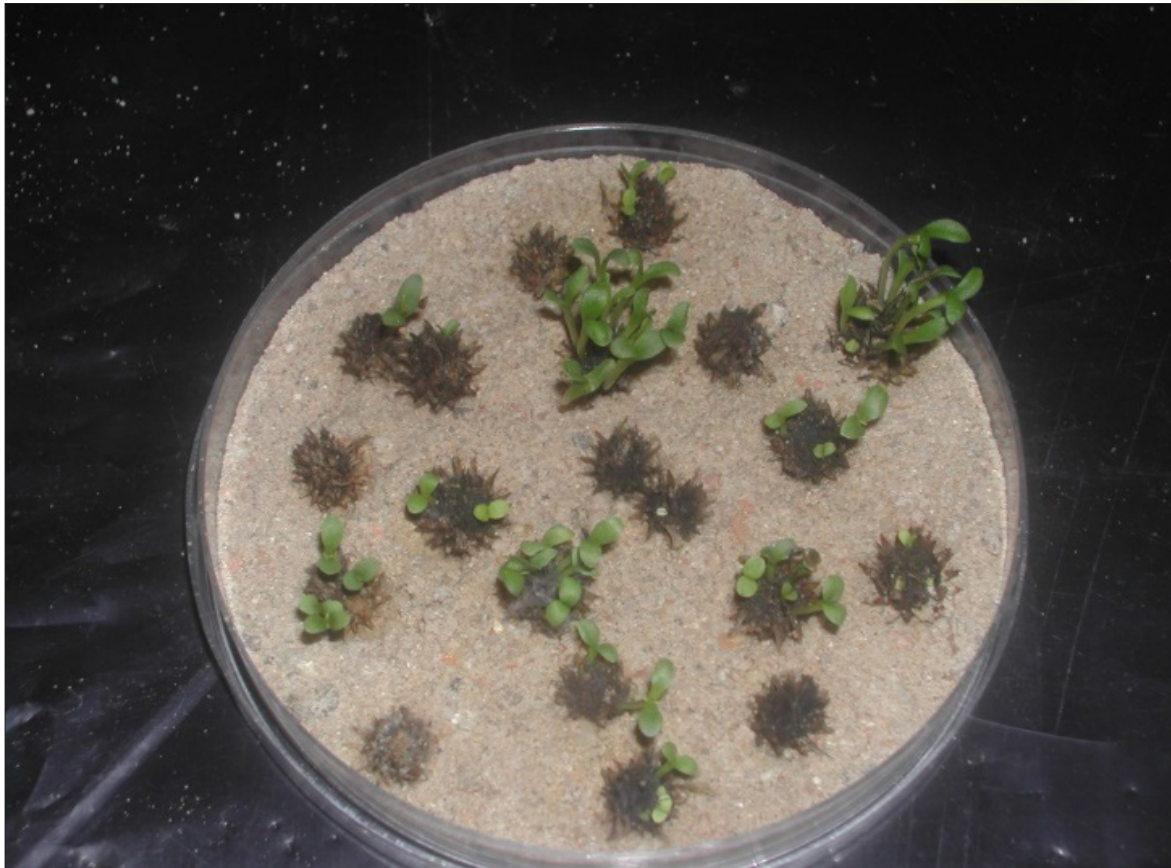
Capitulum (plural – capitula) of *Rhanterium epapposum* contains six to 8 seeds/achenes. Each capitulum is considered as a single unit for assessing viability through the germination method (Figures 70 and 71). Determining the seed viability of *R. epapposum* through the germination method may take longer due to physiological dormancy exhibited (Suleiman et al., 2009 b) at maturation. Therefore, treating the capitula with 750 ppm GA3 for 24 hours is recommended. GA3-treated capitula do not require further pre-soaking in water. The capitula should be thoroughly washed prior to the germination test and care should be taken to position the base of the capitulum in the soil in an upright angle. The pre-treated capitula can be sown in Petri dishes lined with desert soil, as the growing medium, and incubated at 20°C, 12-hour light cycles, and 40% RH in a growth chamber, to accelerate germination (Suleiman et al., 2009b), which might continue for 20 days.

It is important to determine the average seed filling of the capitulum, so that if the seed lots are used for germination in the field, the seed rate can be estimated based on seed filling and viability percentages.

TTC Method: As each capitulum contains multiple seeds, they should be extracted for conducting the TTC test. The seeds can be retrieved from the bottom portion of the outer layer of the capitulum using forceps. For accurate viability assessment, only one seed from each capitulum should be used. The seeds extracted from the capitula should be soaked in water for 24 hours, and the standard TTC procedure (1% TTC) can be followed. As mentioned earlier, the pre-moistened seeds should be cut longitudinally, and one half of each seed should be soaked in 1% TTC for 24 hours and washed thoroughly prior to observation under a microscope for staining (Figure 72).



► Figure 70. *Rhanterium epapposum* capitulum and seed extracted from capitulum.



► Figure 71. Germination test in *Rhanterium epapposum*.



► Figure 72. Stained seed of *Rhanterium epapposum*.

***Tamarix aucheriana* (Athal, Tarfa, أثل، طرفاء)**

Family: Tamaricaceae

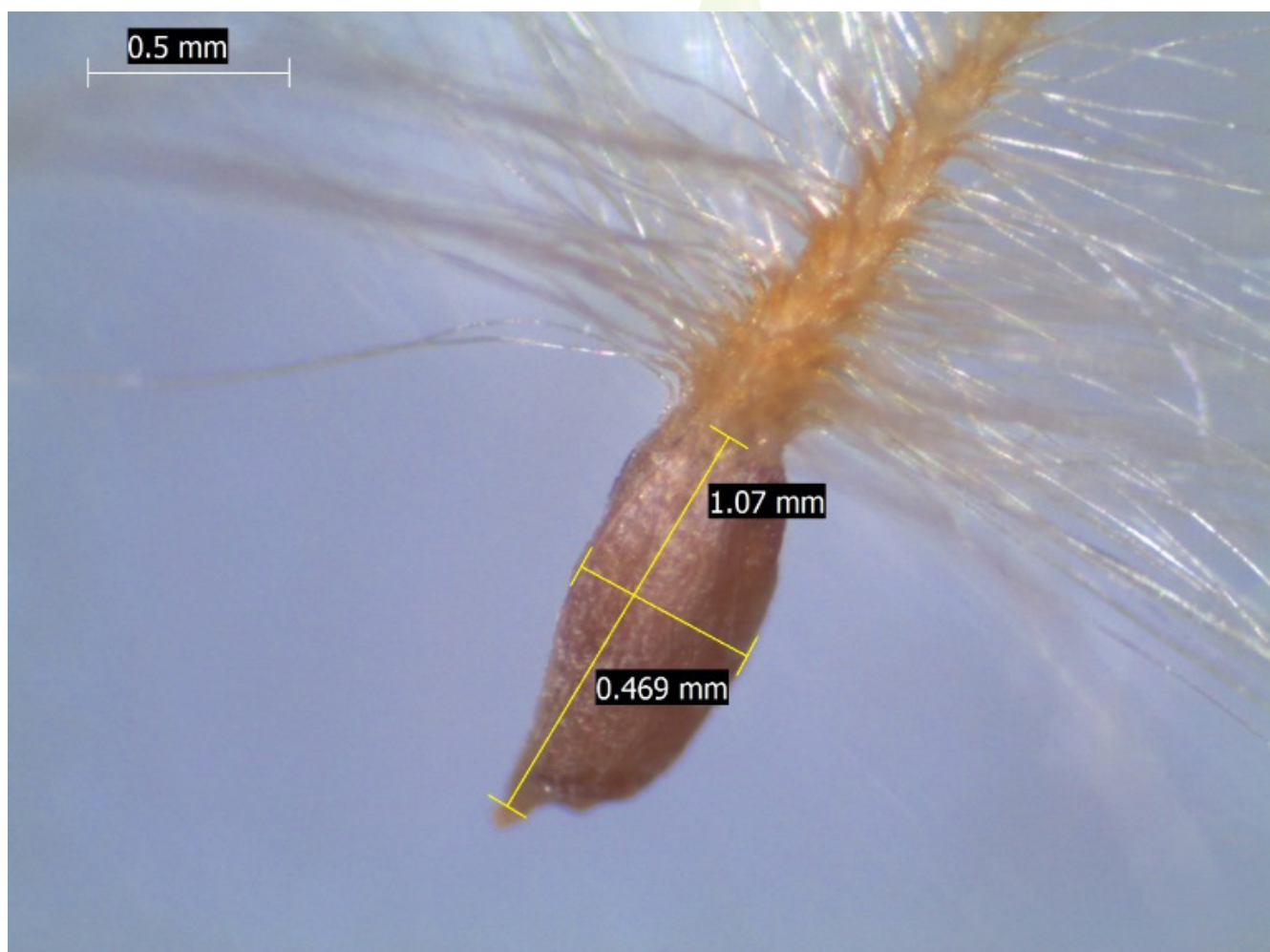


Preferred Method: Germination

Seeds of *Tamarix aucheriana* are enclosed in a three-valved capsule with unicellular hygroscopic hairs and are minute in size (Figures 73 and 74). They should be collected in December-January, as they lose viability rapidly after harvest. Due to their size, the seed viability of this species can be easily assessed using the germination method. Additionally, staining is challenging due to the seed color. The dry seeds should be sown on moistened filter paper in Petri dishes and incubated at 20°C with 12 hours of light and 40% humidity, either in laboratory conditions or a growth chamber. No pre-soaking is required for the germination test. An image of TTC stained seed is also provided below for reference (Figure 75).



► **Figure 73. Fruit of *Tamarix aucheriana*.**



► Figure 74. Seeds of *Tamarix aucheriana*.



► **Figure 75. Germinated seedling and TTC stained seed of *Tamarix aucheriana*.**

Ziziphus spina-christi (Sidr, سدر)

Family: Rhamnaceae



Preferred Method: TTC/Germination

Ziziphus spina-christi produces globose drupe fruits with an outer fleshy layer surrounding a single shell of hardened endocarp with seeds inside. After removing the fleshy part, the hard endocarp is cracked open with a measured force using a hammer. The endocarp typically contains two chambers separated by a divider, and inside each section, a seed can be found (usually). Care must be taken to apply the correct amount of force to avoid damaging the seeds (Figures 76 and 77). Due to their size (Figure 78), the TTC test is suitable for assessing seed viability. In each endocarp, there might be more than one seed. For accurate viability assessment, only one seed from each endocarp should be used. As detailed in the methodology section, the pre-soaked seeds should be split into two with a longitudinal cut and treated with 1% TTC for 24 hours. Staining should then be observed to calculate viability (Figure 79).

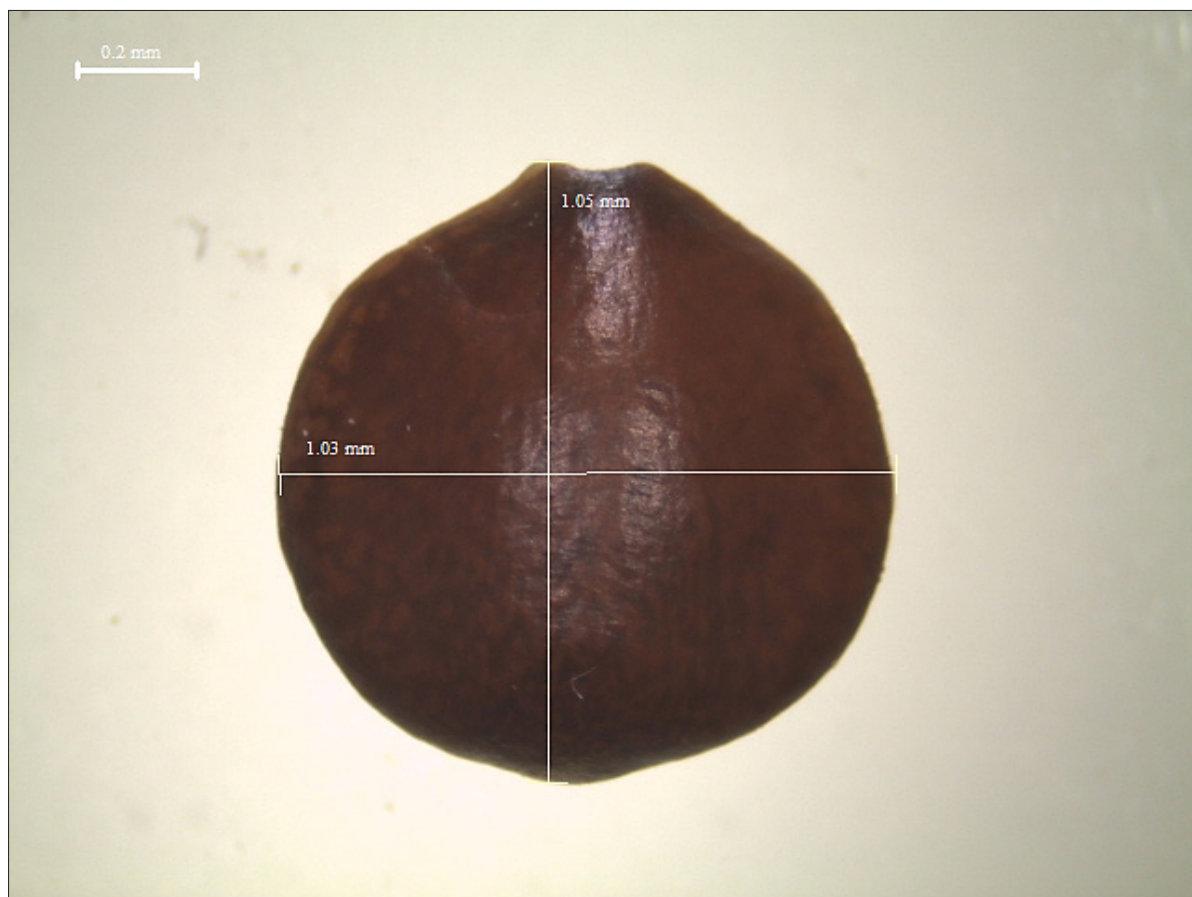
The viability of this species can also be assessed using the germination method. For this purpose, the endocarp should be pre-soaked in water for 24 hours. As the two chambers in the endocarp separate during germination, each endocarp should be sown separately in one jiffy pot or germination trays in agricultural soil medium to avoid counting errors. To assess the viability, 25 endocarps should be sown in separate units, with four replications. Care should be taken to record only one germination per endocarp (even if multiple germinations occur) to ensure accurate viability. The germination typically begins after 10 days and continues for additional two weeks. Hence, assessing the viability of *Z. spina-christi* through germination method is a time-consuming process.



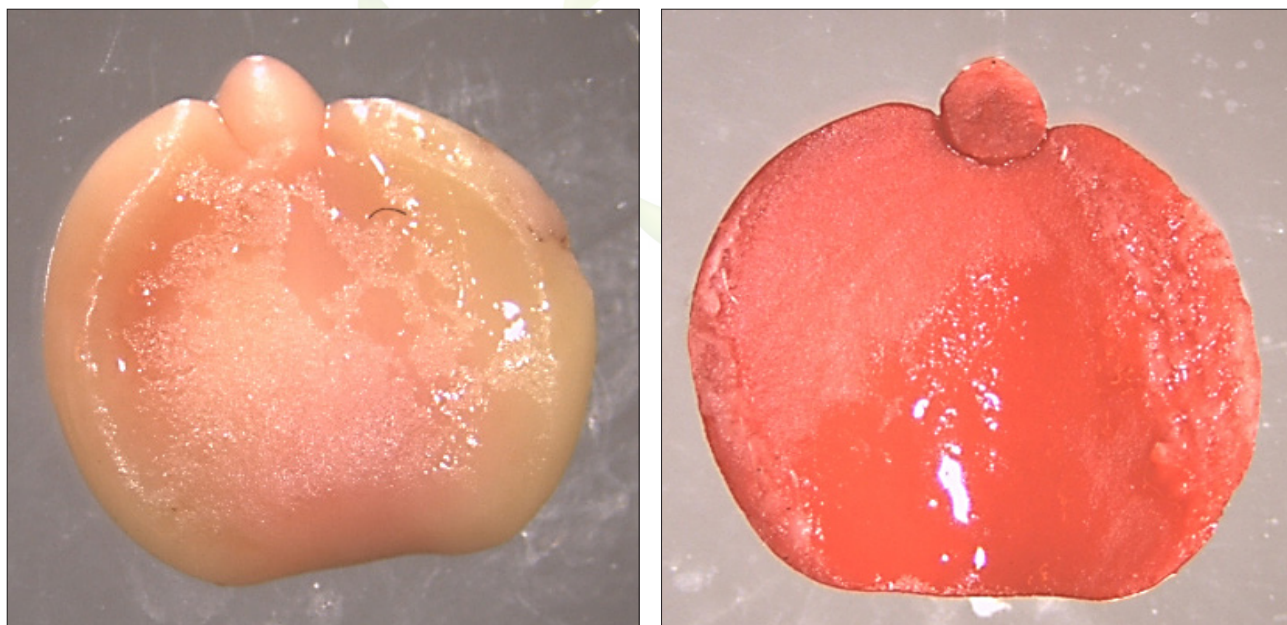
► Figure 76. Endocarp of *Ziziphus spina-christi*.



► **Figure 77. Extraction of *Ziziphus spina-christi* seed from the endocarp.**



► Figure 78. Seed of *Ziziphus spina-christi*.



► Figure 79. Unstained and stained viable seed of *Ziziphus spina-christi*.



► **Figure 80. Germination of *Ziziphus spina-christi*.**



► Figure 81. Germination and separation of seedlings in *Ziziphus spina-christi*.

Zygophyllum qatarense (Harm, هرم)

Family: Zygophyllaceae

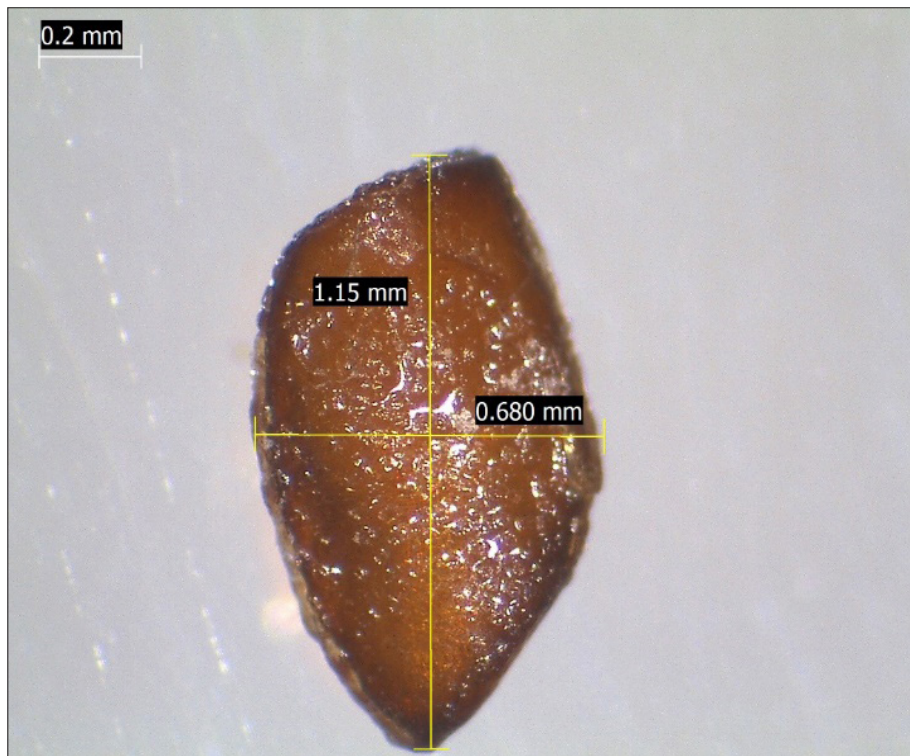


Preferred Method: TTC Test

The seeds of *Zygophyllum qatarense* manually extracted from the fruits (capsule form) should be used for testing the viability. Due to physiological dormancy in this species, it is better to assess the viability of seeds through the TTC method (Figures 80 and 81). Pre-moistened seeds should be longitudinally cut, and the cut portion (half seed) should be soaked in 1% TTC in Petri dishes (covered with aluminum foil) for 24 hours before observation, for staining under microscope (please refer to the section on Viability Assessment through TTC Method for detailed methodology). An image of germinated seedling is also provided below for reference (Figure 82).



► Figure 82. *Zygophyllum qatarense* seeds.



► **Figure 83. Dry and stained seed of *Zygophyllum qatarense*.**



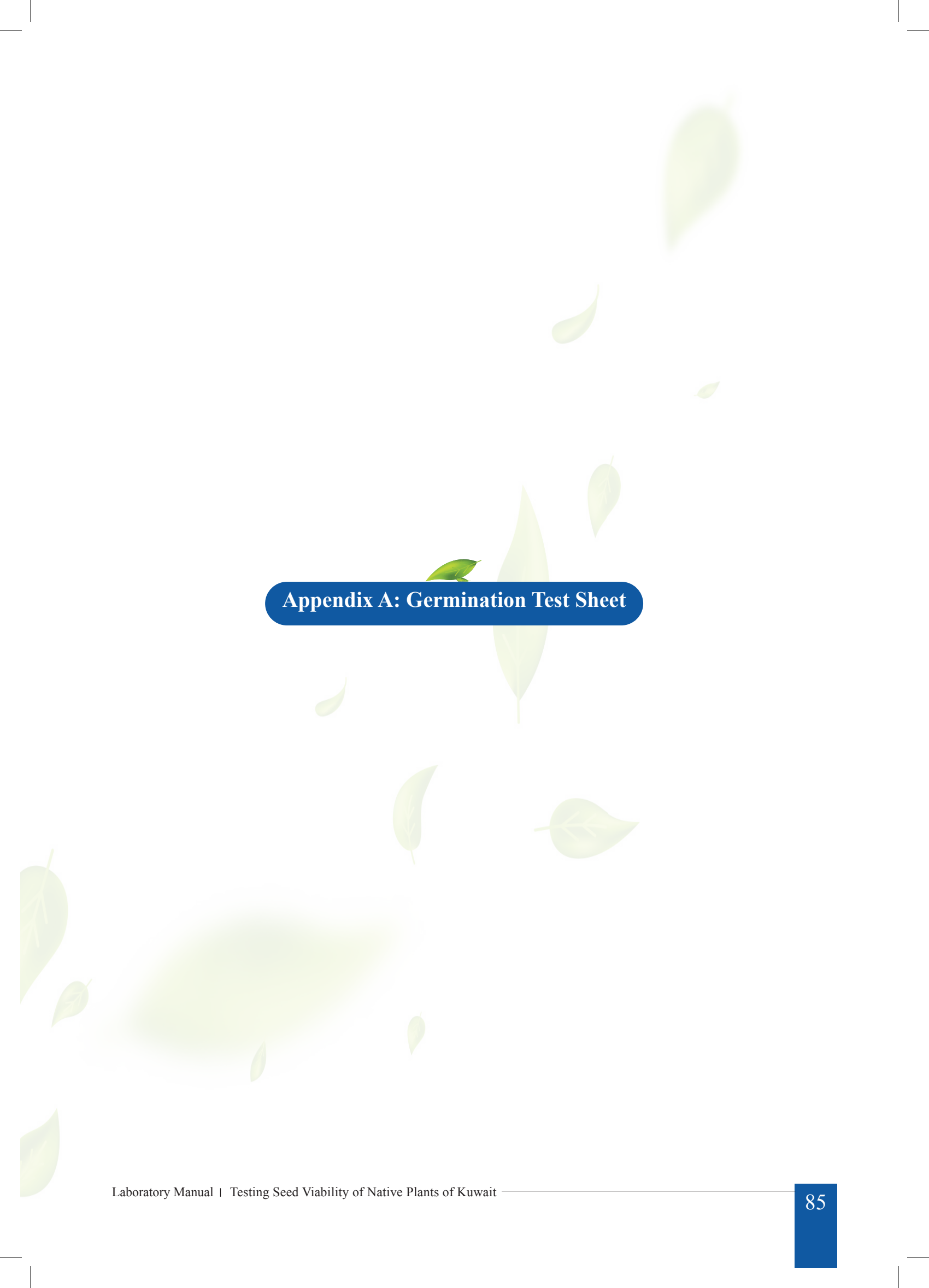
► **Figure 84. Germinated seedling of *Zygophyllum qatarense*.**

References

- Al-Shamsi, N. 2009. Germination ecology of two indigenous range grasses *Lasiurus scindicus* and *Panicum turgidum* . M.Sc Thesis. Al-Ain: UAE University.
- Batanouny, K., Zayed, K., & Kabil, H. (2006). Reproductive ecology of *Panicum turgidum* Forssk. *Taeckholmia*, 26(1), 63-88.
- Bhatt, A., Bhat, N. R., & Thomas, T. M. (2019). Germination behavior of *Seidlitzia rosmarinus* Boiss., a perennial halophyte of *Arabian deserts*. *Agriculture and Natural Resources*, 53(4), 348-354.
- Bhatt, A., Batista-Silva, W., Gallacher, D. J., & Pompelli, M. F. (2020). Germination of *Cenchrus ciliaris*, *Pennisetum divisum*, and *Panicum turgidum* is seasonally dependent. *Botany*, 98(8), 449-458.
- Chakravarty, A. K., & Kalkani, L. (2016). Study on variation in seed yielding components of *Cenchrus Ciliaris* Linn. *Annals of Arid Zone*, 5(1). Available at <https://epubs.icar.org.in/index.php/AAZ/article/view/63274>
- Clor, M. A., Al-Ani, T. A., & Charchafchy, F. (1976). Germinability and seedling vigor of *Haloxylon salicornicum* as affected by storage and seed size. *Rangeland Ecology & Management/Journal of Range Management Archives*, 29(1), 60-62.
- Dhief, A., Guasmi, F., Triki, T., Mohamed, N., & Aschi-Smiti, S. (2011). Natural genetic variation in *Calligonum Tunisian* genus analyzed by RAPD markers. *African Journal of Biotechnology*, 10(48), 9766-9778.
- Derbel, S., & Chaieb, M. (2007). Germination behaviour and seedling establishment of two desert shrubs, *Calligonum polygonoides* (Polygonaceae) and *Spartidium saharae* (Fabaceae), under experimental conditions. *Acta Botanica Gallica*, 154(4), 533-544.
- El-Keblawy, A. (2013 a). Effects of seed storage on germination of two succulent desert halophytes with little dormancy and transient seed bank. *Acta Ecologica Sinica*, 33(6), 338-343.

- El-Keblawy, A. (2013 b). Impacts of dormancy-regulating chemicals on innate and salinity-induced dormancy of four forage grasses native to Arabian deserts. *Grass and Forage Science*, 68(2), 288-296.
- ISTA. 2007. International Rules for Seed Testing. International Seed Testing Association, Bassersdorf, Switzerland.
- Mahajan, G., Mutti, N. K., Jha, P., Walsh, M., & Chauhan, B. S. (2018). Evaluation of dormancy breaking methods for enhanced germination in four biotypes of *Brassica tournefortii*. *Scientific Reports*, 8(1), 17103.
- Zaman, S., Padmesh, S., Bhat, N. R., & Tawfiq, H. (2009). Germination characteristics and storage behavior of *Tamarix aucheriana* (Decne.) seeds. *European Journal of Scientific Research*, 26(4), 532-538.
- Suleiman, M. K., Zaman, S., Al-Dossery, S., Thomas, R. R., & Jacob, S. (2009 a). Germination studies in *Horwoodia dicksoniae* Turrit. *International journal of environmental studies*, 66(2), 221-225.
- Suleiman, M. K., Bhat, N. R., Abdal, M. S., Zaman, S., Jacob, S., & Thomas, R. R. (2009 b). Germination studies in *Rhanterium epapposum* Oliv. *World Applied Science Journal*, 7(4), 468-471.
- Suleiman, M. K., Bhat, N. R., Jacob, S., & Thomas, R. R. (2011). Germination studies in *Ochradenus baccatus* Delile., *Peganum harmala* L. and *Gynandrisis sisyrinchium* Parl. *Research Journal of Seed Science*, 4(1): 58–63.
- Suleiman, M. K., Dixon, K., Commander, L., Nevill, P., Bhat, N. R., Islam, M. A., Jacob, S., & Thomas, R. R. (2018). Seed germinability and longevity influences regeneration of *Acacia gerrardii*. *Plant Ecology*, 219, 591-609.
- Zaman, S., Padmesh, S., & Tawfiq, H. (2009). Effect of Pre-germination Treatments on Seed Germination of *Helianthemum lippii* (L.) Dum.Cours. *Desert Plants*, 25, 18-21.





Appendix A: Germination Test Sheet

Laboratory Manual | Testing Seed Viability of Native Plants of Kuwait

Proj ect:	Sample ID:	Job ID:	Date:
--------------	---------------	---------	-------

TZ Result -	Genus -
	Species -
TZ Date -	

GERMINATION DATA

[illegible]

Conditions -

Temp:

RH:

$$\text{Germination (\%)} = G / X * 100$$

$$\text{Viability (\%)} = (\text{G} + \text{F} + \text{A}) / \text{X} * 100$$

G = number of seeds germinated

X = number of seeds sown (excluding empty and infested)

F = number of fresh seed

A = number of abnormal seedlings

