

# **USER MANUAL**



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#### 1.0 Introduction

The purpose of the Sinar GermPro is to measure the ENZYMIC activity in the germs or embryos of grain kernels and seeds.

This manual details the actions required to determine the germination capacity of samples, using a Sinar GermPro.

In the Sinar GermPro, a solution containing Tetrazolium salt is used as a reagent. This liquid is sensitive to Hydrogen Creating enzymes. The hydrogen produced by the action of these enzymes will reduce Tetrazolium to a light red colour (RED FORMALAZIN), when a germ capable of germination is in contact with the solution. If the seed is incapable of germination, no colour is produced. Please note, if pale stains are present with a slight brown colour this may indicate heat damage and the 4mIGE test may give a low result for the first 2 days as the vigour of the seed may be reduced as it deteriorates.

It is important that when the reaction takes place, a vacuum is created to accelerate the process, hence the hand pump supplied with each GermPro. As a result, tests take only a few minutes instead of several hours normally required by conventional methods.

## 2.0 Reagent

For testing the germinative capacity of malt, 235-triphenyl-tetrazolium-chloride is required. You will need to use a 0.5 to 1% solution for testing purposes - i.e. 0.5 gm/1.0 gm to 100 ml of distilled water. When preparing the solution, warm the distilled water (without heating). This will make the preparation of the solution easier. Once prepared, store the solution in a dark bottle in the absence of light. Shelf life is 2 to 3 months when stored correctly.

### 3.0 GermPro Setup.

- 3.1 Connect the GermPro to a suitable power supply using the power cord supplied.
- 3.2 Switch on using the switch on rear panel, the word "HI" will appear on the display.
- To set the test temperature, press the "**TEMP**" key. The current temperature setting is displayed. Use the "UP" and "**DOWN**" keys to select the required temperature within the range of ambient to +60°C. The temperature indexes in 1° intervals. Typically, a temperature +40°C is required for repeatable, accurate results.

Press the "**SET**" key to store the selected temperature. The display will flash and the unit will beep to indicate the value has been stored. It is not necessary to repeat this procedure again, unless a change of temperature is required.

Use a digital temperature probe to confirm the solution temperature. Pour the solution into the glass flask up to the max fill line. Place the flask in the heating chamber within the instrument. Allow the fluid to warm for approx 20mins. Place the temperature probe in the solution. Increase or decrease the instrument set temperature as described above to achieve the required solution temperature.

3.4 To set the test time period, press the "**TIME**" key. The current time setting is displayed.

Use the "**UP**" and "**DOWN**" keys to select the required time. The time indexes in 10 second intervals.

Press the "**SET**" key to store the selected time. The display will flash and the unit will beep to indicate the value has been stored. It is not necessary to repeat this procedure again, unless a change of time is required.

- 3.5 Remove the vessel stopper, (it has a domed top) with grain slides.
  - Unscrew and remove the fluted ring securing the incubation chamber.
  - Withdrawer the glass incubation chamber and stand to one side.
- 3.6 Pour the required Tetrazolium solution into the incubation chamber. Fill to the **black fill line** marked on the side of the vessel.
  - Put the incubation chamber back into the GermPro.
  - Screw down the fluted ring securing the incubation chamber.
- 3.7 Replace the stopper with slides.
- 3.8 A pre-heat period is required for the incubation chamber to reach equilibrium. This will vary with the selected temperature. Allow 30 minutes for a set temperature of 40°C.

### 4.0 To commence a test.

- 4.1 Ensure the time period and temperature settings are correct.
- 4.2 Prepare the grain sample to be tested. Select a small quantity of well-blended grain. Using a sharp blade carefully split the individual grains lengthways to expose the germ embryo.
- 4.3 When 100 grains have been split, discard half the split grains. Place the remaining grains into the incubation slides provided.
- 4.4 Locate the slides onto the hook on the underside of the stopper.
- 4.5 Lower the slides into the chamber and secure the stopper.
- 4.6 Using the hand vacuum pump, press the handle several times to evacuate to desired vacuum. Normally 20-30 cm/Hg.
- 4.7 Press the "RUN/STOP" key. The GermPro will automatically use the stored elapse time. The count down time is displayed in minutes and seconds. On completion the unit beeps and the word "END" is displayed.
- 4.8 To interrupt a test, press the "**RUN/STOP**" key again. The unit beeps and the word "**END**" is displayed.
- 4.9 Release the vacuum by squeezing the release lever on the hand pump and remove the stopper with slides for visual inspection.
- The level of liquid with the GermPro should be checked and topped up if necessary. Change the Tetrazolium solution on a regular basis to ensure that it remains clean and effective. A dirty solution will become tired and less active resulting in poor results.

The GermPro remains on in standby mode until the next test is to be performed.

Repeat stages 4.1- 4.9

#### 5.0 Determination

- 5.1 Determine the germination percentage by close observation of the germs in the half grains. This should be done within 10 minutes of taking the sample out of the GermPro (leaving the sample too long may invalidate the test). It is important to remove the sample holder cover and closely examine each half-grain with tweezers and use a strong magnifying glass if pre-germination is suspected.
- 5.2 The germination percentage is determined by counting the number of dead, pre-germinated and heat damaged grains and subtracting it from 100.
- 5.3 Live Grains are acceptable when the embryo is stained pink, report as **% Germ / Capacity.** Ensure that cutting damage is not misinterpreted as being a fail.

#### 6.0 General use and maintenance

- 6.1 Take care not to overfill the incubation chamber.
- 6.2 It is recommended that the Tetrazolium solution be changed on a regular basis. To clean the chamber rinse with distilled water.
- 6.3 Leave the GermPro switched on between tests. The keeps the dry block heater permanently at the required test temperature.
- 6.4 Periodically lubricate the orange stopper "O" ring with silicone grease.