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Research Article Isolation of Drought-Tolerant Endophyte Bacteria From Local Tomato Plants

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Abstract

Background and Objective: Exploration of drought-tolerant endophytic bacteria is significant to identify bacteria that can provide plant resistance to drought stress. This study aims to obtain the potential of endophytic bacteria to promote plant growth from tomato plants in dry land. **Materials and Methods:** Exploration of endophytic bacteria using polyethylene glycol 6000. Selected isolates were tested to increase the viability of tomato seeds using a Completely Randomized Design (CRD). **Results:** There were 123 isolates of endophytic bacteria isolated from the roots and stems of local tomato plants in a dry and rocky land, Muna Regency, Indonesia. Selection of drought, 55 (44.71%) isolates very sensitive to drought, 8 (6.50%) isolates tolerant to drought and 21 (17.02%) isolates very tolerant to drought. Dryness for the maximum polyethylene glycol concentration at osmotic pressure of -2.00 MPa. Inoculation of endophytic bacteria in local tomato seeds increased the viability and vigour of local tomato seeds compared to the absence of endophytic bacteria. Of the 21 isolates of drought-tolerant endophytic bacteria, there were 12 potential isolates in increasing the viability of local tomato seeds belonging to the *Pseudomonas* sp. and *Bacillus* sp. bacterial groups. **Conclusion:** *Pseudomonas* sp. and Bacillus sp. isolates identified the endophytic bacteria, which can be drought-tolerant and increase tomato seeds' viability.

Key words: Drought tolerant, dry land, endophytic bacteria, tomato, growth promoters, glycol, completely randomized design

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Drought stress is one of the agricultural problems that limit crop productivity in dryland¹. High temperature, lack of rainfall and lack of nutrients are considered as limit factors that greatly affect agricultural productivity in dryland². Drought stresses on plant growth can appear on a low to large scale with moderate to prolonged duration that limits yields³. Drought in plants will affect the water potential of plants, plant morphology and physiology. It also reduces nutrient diffusion and the flow of water-soluble nutrients, such as nitrates, sulphates, Ca, Mg and Si^{4,5}. In general, drought affects plant growth in both quantity and quality. And mitigation of drought risk needs to be achieved.

Muna Regency is one of the regencies in Southeast Sulawesi, Indonesia, geographically characterized by dry land and is dominated by rocks widely spread throughout the region. Nevertheless, Muna Regency has superior plants that are widely cultivated by the community, namely local tomatoes. Local tomatoes are crops cultivated by farmers for generations, especially in Muna Regency. These tomatoes can grow and develop in rocky land. They have different characteristics from other tomatoes, which are large and have a more savoury taste compared to other tomatoes. In Muna Regency, community needs for tomatoes have not been matched by adequate productivity. Tomato productivity in the Muna district is very low, reaching 2.2 tons ha⁻¹, while the average productivity of Southeast Sulawesi tomatoes is 5.4 tons ha^{-1 6}. Muna Regency has limited land for the development of tomato cultivation, most of this cultivation is carried out on land which is classified as rocky soil, dry land and low soil fertility.

Research about the use of microorganisms can help plants to cope with drought stress⁷. Endophytic bacteria live and are associated with plant tissue without causing a disease symptom in the plant⁸. The presence of endophytic bacteria in plant tissue plays a significant role in improving plant growth (plant growth promotion), producing growth promoters, tethering N, mobilizing F, playing a role in plant health and inducing plant resistance pathogenic disorders^{9,10}. Besides, endophytic bacteria play a role in helping plant growth gradually to adapt to environmental conditions that are gripped by drought by producing several compounds that can enhance plant growth so that it helps plants adapt to their environmental conditions⁸. Furthermore, Anyasi et al.¹⁰ state that endophytic bacteria can significantly increase plant growth by stimulating the growth of root biomass to provide ecological services to plant growth.

Exploration of endophytic bacteria from dry land on local tomato plants is expected to produce potential endophytic bacteria for agricultural development on dry land. Using selected and initialized microbial isolates is expected to promote the growth of dryland plants to achieve maximum yields despite adverse environmental conditions for plant growth.

This study aims to select endophytic bacteria that are drought tolerant and can increase the viability of seeds isolated from local tomato plants in dry and rocky areas of Muna Regency to develop biofertilizers in dry land.

MATERIALS AND METHODS

Study area: This research was conducted at the Agronomy Laboratory, Faculty of Agriculture, Halu Oleo University, Kendari City, Indonesia, from April, 2018-December, 2019. Samples of tomato plants were taken from Muna Regency, Indonesia, which were spread over seven sub-districts, namely Napabalano, Pasir Putih, Lohia, Sawerigadi, Kabawo, Kusambi and Maligano. Tomato plants used as a source of endophytic bacteria are healthy plants with the criteria of having healthy growth, large stem diameter and abundant yields. Tomato plants were pulled out to the roots using gloves, then were put into plastic samples and labelled containing data about the location and time of sampling. The tomato plant samples that have been obtained were placed in an icebox to be brought to the laboratory and stored at a temperature of around 40 until the isolation stage is carried out.

Isolation of endophytic bacteria of muna local tomato plants: Endophytic bacteria were isolated from the roots and stems of tomato plants. The roots and stems of the plant were washed and dried for 30 min. Then the roots and stems were weighed as much as 1 g and the surface was sterilized using 70% alcohol for 3 min. Subsequent sterilization using 4% NaOCI solution for 3 min and rinsed using sterile distilled water three times. The sterilized parts of the roots and stems and 0.1 mL of the last rinsed distilled water were applied to 5% TSA media to see the effectiveness of surface sterilization. Roots and stems scratched on 5% TSA media are then ground using a mortar and diluted to a dilution series of 10^{-10} . Dilution series of 10⁻⁸ and 10⁻¹⁰ were spread on TSA media in a petri dish with a 50µL scatter volume. The scattered suspension was incubated for two days, then observation of the growing colonies was carried out. Colonies showing morphological differences were further isolated. Isolation of bacterial colonies from culture medium was repeated (purification of isolates) to obtain a single bacterial colony or pure culture for further testing and stored in an Eppendorf tube containing 0.9 mL of a sterile glycerol 15% solution stored in a freezer under the temperature of -20.

Drought tolerant endophytic bacteria selection: The Nutrient Broth (NB) media was homogenized and sterilized for 15 min at 121°C. The media is added to PEG 6000 while it is still hot¹¹. The determined osmotic potential was -1.0, -1.25, -1.50, -1.75 and -2.0 MPa. A total of 1 mL suspension was grown on NB media with a population around 108 inoculated into 20 mL of NB media that had been added with Polyethylene Glycol (PEG) 6000 with variations in the concentration of 295.75-428.89 g L⁻¹. Furthermore, it was incubated at a shaking machine speed of 80 rpm at 38°C for 24 hrs. Besides, Optical Density (OD) measurements used a spectrophotometer (at a wavelength of 570 nm). Based on Optical Density values, it obtained a range of droughttolerant values, that was, very Sensitive OD = 0.3, Sensitive OD = 0.3-0.4, Tolerant OD = 0.4-0.5 and Very Tolerant $OD > = 0.5^{12}$.

Propagation of endophytic bacterial isolates and seed treatment: Pure culture of endophytic bacterial isolates was further propagated using TSA media for *Bacillus* sp. and King's B for the *Pseudomonas* sp. and incubated for 48 hrs. Furthermore, bacterial cell culture is harvested by suspension. Local tomato seeds are then put into endophytic bacterial suspension and shaker for 24 hrs at a speed of 120 rpm¹³.

Muna local tomato seed germination test: Local tomato seeds were inoculated with endophytic bacterial suspension and then germinated in a germination box size $20 \times 15 \times 10$ cm (length×width×height) using sterile husk charcoal media. For each treatment, 50 local tomato seeds were planted with three replications. Observations were made for 14 days for viability and vigour of local tomato seeds, which included:

Maximum growth potential (MGP): Maximum growth potential (MGP) is calculated based on the percentage of seeds showing growing symptoms which are calculated from the first day (on the 3rd day) to the last day (14th day), with the Eq.¹⁴:

 $MGP = \frac{\sum growth}{\sum plant \ seeds} 100\%$

Simultaneous growth (SG): Observations were made on strong-growing seedlings, which showed simultaneity in the test media. Growth equality is expressed in per cent. SG observation is carried out on the 14th day¹⁴:

$$SG = \frac{\sum best seed}{\sum planted seed} s100\%$$

Relative growth speed (RGS): Relative Growth Speed (RGS) illustrates seed viability, which is a comparison between the Growth Speed Value (GSV) and the maximum Growth Speed Value (GSVmax). The maximum GSV is obtained based on the assumption that at the first count (I), Normal Sprouts (NS) have reached 100%. The relative growth speed is calculated based on the accumulation of daily growth speed with the Eq.¹⁴:

$$RGS = \frac{GSV}{GSV \max}$$
$$GSV = \sum_{0}^{tn} \frac{N}{t}$$
$$GSV \max = \frac{100}{\sum \text{ count day}}$$

Where, tn is the last time observation, N is the Percentage (%) of normal sprouts at each time of observation and t is the time of observation.

Data analysis: The results of the observation data of drought-tolerant endophytic bacteria ability were analyzed descriptively. Meanwhile, the viability test for local tomato seeds was analyzed using variance analysis. If the analysis of variance shows a real effect, it will be continued with the Duncan Multiple Range Test (DMRT) at the 95% significance level.

RESULTS

The results of endophytic bacteria isolated from the local Muna tomato plant had been purified from contaminants that grew when bacteria were planted on plant media. Besides, the results of purification and selection of endophytic bacteria using TSA media obtained 115 isolates of endophytic bacteria. The number of endophytic bacterial isolates found in Muna's local tomato plant tissue is presented in the following Fig. 1.

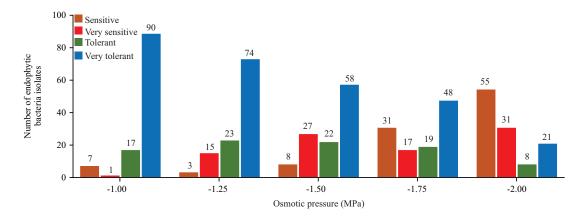


Fig. 1: Some endophytic bacterial isolates from Muna local tomatoes using PEG 6000

The results of tolerance level of endophytic bacteria from local mammals on NB media added with PEG 6000 obtained bacterial characteristics based on Optical Density (OD) values at osmotic pressure -1.00 MPa obtained one very sensitive bacterial isolates, seven isolates of sensitive bacteria, 17 bacterial isolates were tolerant and 90 bacterial isolates were very tolerant. At osmotic pressure -1.25 MPa, 15 sensitive bacterial isolates were obtained, three bacterial isolates were very sensitive, 23 bacterial isolates were tolerant and 74 bacterial isolates were very tolerant. For osmotic pressure, -1.50 MPa obtained eight isolates of highly sensitive bacteria, 27 isolates of sensitive bacteria, 22 isolates of tolerant bacteria and 58 isolates of highly tolerant bacteria. At osmotic pressure -1.75 MPa, 31 isolates of highly sensitive bacteria were obtained, 17 isolates of sensitive bacteria, 19 isolates of tolerant bacteria and 48 isolates of highly tolerant bacteria, whereas at osmotic pressure of -2.00 MPa found 31 isolates of bacteria sensitive to drought, 55 isolates of bacteria were very sensitive to drought, eight isolates of bacteria were tolerant to drought and 21 bacterial isolates were very tolerant to drought. Endophytic bacterial isolates from Muna local tomatoes can survive up to osmotic pressure of -2 MPa.

Drought stress drastically affected the number of endophytic bacterial isolates that were tolerant to drought. They decreased with increasing osmotic pressure. At osmotic pressure -1.00 MPa, the number of endophytic batteries was very drought tolerant, that were 90 bacterial isolates but at osmotic pressure -1.25, -1.50, -1.75 and -2.0 MPa the number of bacterial isolates endophytic which was very tolerant to drought decreased with the number of isolates at each osmotic pressure, that was 74, 58, 48 and 21, respectively.

Endophytic bacteria with the highest Optical Density value at osmotic pressure -2.00 MPa were found in isolate SWR I A05 with a value of 0.661 and followed by isolates LBR II B02

and LBR 1 A03, which was 0.659. The result of Table 1 shows that the presence of endophytic bacterial isolates at osmotic pressure of -2.00 MPa plant parts is more dominated at the root of the plant with 15 isolates than the parts of the plants that amounted to 6 bacterial isolates.

The results of the bacterial isolates which were isolated from tomato plants showed that the isolates that had the highest OD values at each osmotic pressure were LBR II B02 isolates with an osmotic potential value of -1.00, -1.25, -1.50, -1.75 and -2.00 MPa, i.e. 1.166, 1.143, 1.119, 0.988, 0. 659, respectively. So the lower the osmotic pressure, the more PEG 6000 was added. Therefore, the OD value produced by LBR II B02 isolates would decrease with decreasing osmotic pressure. At osmotic pressure -2.0 MPa had less water concentration than -1.00, -1.25, -1.50 and -1.75 MPa.

DMRT test results in Table 2 show that the highest potential for maximum growth of local Muna tomato seeds was found isolate SDM II A04, SDM II B05 and SDM II A03 with a value of 100% and the lowest value was found in the treatment without endophytic bacteria. Still, it was not significantly different from other treatments. DMRT test results on the highest level of growth of local Muna tomato seeds were found in SWR I A04 isolates that were significantly different from the treatment without endophytic bacteria. Still, they were not significantly different from other treatments. DMRT test results of the highest relative growth rate of local Muna tomato seeds were found in isolates SWR III B02, which were significantly different from isolates SDM I A02 and SDM II A03, SWR III B04, MO II 02, especially without the treatment of endophytic bacteria but they were not significantly different from other isolates. The treatment of local Muna tomatoes using endophytic bacterial isolates can increase the maximum growth potential, growth equality and relative growth speed of local Muna tomato seeds.

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Table 1: Endophytic bacteria from Muna local tomatoes were very tolerant of drought at a pressure of -2.00 MPa

	Osmotic pressure							
						Drought	Place of	Sample location
Isolate code	-1.00	-1.25	-1.50	-1.75	-2.00	tolerance	isolate	(district, village)
TB I B06	0.819	0.607	0.511	0.509	0.505	Very tolerant	Stem	Napabalano, Tobea
LBR II B02	1.166	1.143	1.119	0.988	0.659	Very tolerant	Stem	Lohia, Liangkabori
M0 II A02	0.819	0.696	0.660	0.635	0.577	Very tolerant	Root	Maligano, Maligano
SWR III B02	1.137	1.002	0.955	0.818	0.549	Very tolerant	Stem	Sawerigadi, Sawerigadi
BU II 06	1.059	0.532	0.520	0.515	0.510	Very tolerant	Root	Pasir Putih, Bumbu
LBR I A03	1.036	1.373	0.751	0.567	0.659	Very tolerant	Root	Lohia, Liangkabori
SWR I A02	1.226	1.074	1.053	0.929	0.528	Very tolerant	Root	Sawerigadi, Sawerigadi
KS III A08	0.880	0.732	0.768	0.727	0.655	Very tolerant	Root	Kabawo, Kasaka
SWR I A04	1.228	1.030	0.909	0.516	0.574	Very tolerant	Root	Sawerigadi, Sawerigadi
LI I B04	1.039	0.904	0.857	0.659	0.539	Very tolerant	Stem	Lohia, Liangkabori
SDM II A04	0.960	0.823	0.748	0.655	0.555	Very tolerant	Root	Kusambi, Sidamangura
M0 II A06	0.856	0.625	0.580	0.546	0.536	Very tolerant	Root	Maligano, Maligano
LAK II A02	1.666	1.163	0.587	0.530	0.565	Very tolerant	Root	Napabalano, Langkumapo
LI II A03	1.269	1.301	0.867	0.859	0.553	Very tolerant	Root	Lohia, Liangkabori
SWR III B04	1.059	1.200	0.980	0.873	0.615	Very tolerant	Stem	Sawerigadi, Sawerigadi
SDMIIB05	1.134	0.874	0.515	0.573	0.507	Very tolerant	Stem	Kusambi, Sidamangura
SWR II A04	1.259	1.294	0.986	0.866	0.555	Very tolerant	Root	Sawerigadi, Sawerigadi
SWR I A05	1.167	1.506	1.071	0.926	0.661	Very tolerant	Root	Sawerigadi, Sawerigadi
SDM II A03	1.432	1.287	1.032	0.854	0.560	Very tolerant	Root	Kusambi, Sidamangura
SWR II A02	1.293	1.606	0.931	0.833	0.534	Very tolerant	Root	Sawerigadi, Sawerigadi
SDM I A02	1.289	0.994	0.738	0.618	0.564	Very tolerant	Root	Kusambi, Sidamangura

Table 2: Effect of seed treatment with drought-tolerant endophytic bacteria

Endophytic bacterial isolates	Isolate code	MGP	SG	RGS
No endophytic bacteria	Control	80.67 ^b	81.33 ^b	71.97°
Pseudomonas sp.	TB I B06	97.33ª	94.67ª	93.79 ^{abc}
Pseudomonas sp.	LBR II B002	96.00ª	90.67 ^{ab}	94.71 ^{abc}
<i>Bacillus</i> sp.	M0 II 02	94.67ª	93.33ª	82.73 ^{cde}
<i>Pseudomonas</i> sp.	SWR III B02	96.00ª	96.00ª	99.34ª
<i>Bacillus</i> sp.	BU II B6	98.67ª	96.00ª	96.24 ^{ab}
<i>Pseudomonas</i> sp.	LBR I A003	82.67 ^b	90.67 ^{ab}	91.30 ^{abc}
<i>Pseudomonas</i> sp.	SWR I A02	96.00ª	94.67ª	97.90 ^{ab}
<i>Pseudomonas</i> sp.	K5 III A08	98.67ª	94.67ª	87.59 ^{a-d}
<i>Pseudomonas</i> sp.	SWR I A04	97.33ª	97.33ª	98.33 ^{ab}
Bacillus sp.	L11B004	97.33ª	89.33ªb	93.86 ^{abc}
<i>Pseudomonas</i> sp.	SDM II A04	100.00 ^a	86.67 ^{ab}	93.14 ^{abc}
<i>Bacillus</i> sp.	M0 II 06	94.67ª	92.00 ^{ab}	96.24 ^{ab}
<i>Pseudomonas</i> sp	LAK II A2	97.33ª	94.67ª	98.75ª
<i>Pseudomonas</i> sp.	L12 A03	97.33ª	90.67 ^{ab}	95.97 ^{ab}
<i>Pseudomonas</i> sp.	SWR III B04	97.33ª	94.67ª	97.61 ^{de}
<i>Pseudomonas</i> sp.	SDMIIB05	100.00ª	88.00 ^{ab}	94.32 ^{abc}
<i>Pseudomonas</i> sp.	SWR II A04	97.33ª	94.67 ^{ab}	92.09 ^{abc}
<i>Bacillus</i> sp.	SWR I A05	96.00ª	81.33 ^b	87.34 ^{a-d}
<i>Pseudomonas</i> sp.	SDM II A03	100.00 ^a	86.67 ^b	86.31 ^{bcd}
<i>Pseudomonas</i> sp.	SWR II A02	98.67ª	89.33 ^{ab}	95.25 ^{ab}
<i>Pseudomonas</i> sp.	SDM I A02	97.33ª	81.33 ^b	79.12 ^e

Numbers followed by different superscripted letters in the same column are significantly different in the UJBD test level $\alpha = 0.05$, MGP: Maximum growth potential, SG: Simultaneous growth and RGS: Relative growth speed

DISCUSSION

Muna Regency is one of the regencies in Southeast Sulawesi characterized by dry land and dominated by rocks. Communities in this area often face the problem of drought, which affects the cultivation of crops. Abiotic stresses due to drought, salinity, cold and heat are the main environmental factors that affect plant growth and they are generally serious threats to agricultural activities. Drought is a stress condition where the plants suffer from water shortages and plants will depend on the availability of groundwater used to carry out their metabolic activities¹⁵. Inoculation of plants by utilizing

microorganisms can increase plants' drought tolerance that grows in arid regions¹⁶. Plants that grow under adverse environments, such as those on dry land plants, will undergo limited water and nutritional deficiencies but have several mechanisms to overcome the bad factors that occur. one of them is by microorganisms help. In this study, there were 115 endophytic bacteria isolated from the local Muna tomato plants that grew in the dry land of the Muna district. Out of 115 endophytic bacterial isolates, 21 (17.21%) of bacterial isolates can tolerate maximum dryness (osmotic pressure of -2.00 MPa). Previous studies have reported that selection of in vitro tolerant bacterial dryness is an alternative to obtaining drought-tolerant microbes such as Forchetti et al.¹⁷ they argued that the selection of endophytic bacterial drought tolerance in vitro could be made by providing a drought simulation using Polyethylene Glycol (PEG). This compound can reduce the osmotic potential of the solution through the ethylene oxide sub-unit matrix activity, which can bind water molecules with hydrogen bonds. PEG solution in vitro media is expected to create a potential osmotic equivalent to soil conditions in the field capacity and critical humidity point so that bacteria responded similarly when they were stressed in the field¹¹.

According to Schloter et al.¹⁸ bacteria that have an Optical Density (OD) value, greater than 0.5 at certain osmotic pressure are categorized as bacteria that are very tolerant of drought. The observations showed that the number of endophytic bacteria that are very tolerant to drought decreases with increasing osmotic pressure, i.e. 90, 74, 58, 48 and 21 endophytic bacterial isolates at osmotic pressure -1.00, -1.25, -1, 50, -1.75 and -2.00 MPa, respectively. The choice of osmotic pressure level is determined by referring to Michel and Kauffman that the pressure created through the addition of water-soluble PEG compounds causes the culture media to become thicker due to a decrease in the potential pressure of the water so that the water in the media is more difficult to use by bacteria¹¹. Several researchers have carried out the use of PEG in selecting drought-tolerant bacteria before getting bacteria that are very tolerant to drought such as based on Sandhya et al.19 who report that from 81 isolates of Pseudomonas fluorescent bacteria isolated from sunflower, only 26 bacterial isolates which can tolerate maximum drought levels at a pressure of 70.73 Mpa. Rehman and Nautiyal used PEG to select Rhizobium sp. NBRI2505 sesbania to get bacteria that have the tolerance to drought²⁰. Likewise, research conducted by Vardharajula et al.²¹ tested the ability of *Bacillus* sp. to drought tolerance. Furthermore, Marulanda et al.22 said that B. megaterium microorganisms have a high tolerance to water deficits caused by osmotic

pressure (PEG), this is because *B. megaterium* under water deficit pressure can induce adjustment of cell osmotic potential, an indication of cellular adaptation this osmotic is a mechanism by which microbial cells can deal with drought stress²³.

Some endophytic bacterial isolates isolated from local Muna tomatoes exhibit tolerance and are very tolerant of drought at maximum osmotic pressure (-2.00 MPa). This indicates that endophytic bacterial isolates can be promoted as bacteria that can induce crop resistance in a dry land. The results of a study conducted by Cherif et al.²⁴ showed that endophytic bacteria significantly increased the growth and biomass of plants exposed to drought. Moreover, Theocharis et al.25 reported that endophytic bacteria could induce resistance to abiotic stress in plant species. Likewise, research conducted by Redman et al.20 that rice plants showed drought tolerance after inoculation with endophytic bacteria and contributed to an increase in the rate of growth, yield and plant biomass so that the utilization of endophytic bacteria was useful for increasing agricultural production to marginal land.

The results showed that the treatment of seeds with endophytic bacterial isolates increased the maximum growth potential, growth synchronization and the speed of growth of local Muna tomato seed compared to seed without the use of endophytic bacteria. Based on the test results of 21 endophytic bacterial isolates, it is obtained 12 isolates that can increase the viability and vigour of local tomato seeds. The results show that the inoculation of endophytic bacterial Paraburkholderia phytofirmans in tomato plant seeds significantly increased seed growth and simultaneous growth compared to without endophytic bacteria isolates²⁵. Khan et al.²⁶ reported that PGPEB Sphingomonas sp. LK11 can significantly increase the growth of Solanum pimpinellifolium despite experiencing salinity stress. Furthermore, Leda et al.27 reported that the bacterium Azospirillum brasilense Sp7 applied to the seeds of corn and tomato plants could increase the growth of plant seeds. Thus, endophytic bacteria isolated from local Muna tomatoes in Muna dry land have potential as drought-tolerant inoculums and can improve seed viability, so that testing of endophytic bacteria on a field scale needs to be carried out to be applied in dryland agriculture.

CONCLUSION

Endophytic bacterial isolates that are re-isolated from local tomatoes in Muna dry land have potential as droughttolerant potential inoculums and can improve the viability and vigour of tomato seeds so that they are very promising for developing biofertilizers in dry soil to rehabilitate degraded ecosystems in the future. Drought tolerant endophytic bacterial selection reveals all endophytic bacterial isolates tested have different potential in providing resistance to pressure and drought. There were 21 (17.02%) endophytic bacterial isolates that were very tolerant of drought at maximum osmotic pressure (-2.00 MPa). There were 21 drought tolerant endophytic bacteria isolates, 12 potential isolates in enhancing the viability of local tomato seeds, including the Pseudomonas sp. and Bacillus sp. isolates. Further studies on environmental factors that influence, growth and survival of endophytic bacteria are needed to identify the appropriate and promising types of bacteria as potential bacteria that can be utilized in dryland agriculture through field trials.

SIGNIFICANCE STATEMENT

This found 21 isolates of endophytic bacteria that were very drought tolerant at maximum osmotic pressure from local tomatoes. The endophytic bacteria isolate the potential as a potential inoculum for drought tolerance and can increase the viability and vigour of tomato seeds so that it is very promising for the development of biofertilizers in the dry land. The use of drought-tolerant endophytic bacteria isolates is expected to enrich the theory about the use of endophytic bacteria in agricultural development in dry land so that it can achieve maximum results even though environmental conditions are less favourable for plants growth.

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REFERENCES

1. Halo, B.A., R.A. Al-Yahyai and A.M. Al-Sadi, 2020. An endophytic talaromyces omanensis enhances reproductive, physiological and anatomical characteristics of drought-stressed tomato. J. Plant Physiol., Vol. 249. 10.1016/j.jplph. 2020.153163.

- 2. Li, G.X., B.C. Xu, L.N. Yin, S.W. Wang and S.Q. Zhang *et al.*, 2020. Dryland agricultural environment and sustainable productivity. Plant Biotechnol. Rep., 14: 169-176.
- Morales, F., M. Ancín, D. Fakhet, J. González-Torralba and A.L. Gámez *et al.*, 2020. Photosynthetic metabolism under stressful growth conditions as a bases for crop breeding and yield improvement. Plants, Vol. 9. 10.3390/ plants9010088.
- 4. Fathi, A. and D.B. Tari, 2016. Effect of drought stress and its mechanism in plants. Int. J. Life Sci., 10: 1-6.
- Anjum, S.A., X.Y. Xie, L.C. Wang, M.F. Saleem, C. Man and W. Lei, 2011. Morphological, physiological and biochemical responses of plants to drought stress. Afr. J. Agric. Res., 6: 2026-2032.
- Pasolon, Y.B. and U. Rianse, 2011. Potensial and use of agricultural resources of Southeast Sulawesi. Res. Trop. Agric., 4: 109-112.
- Vurukonda, S.S.K.P., S. Vardharajula, M. Shrivastava and A. Skz, 2016. Enhancement of drought stress tolerance in crops by plant growth promoting rhizobacteria. Microbiol. Res., 184: 13-24.
- Hardoim, P.R., L.S. van Overbeek, G. Berg, A.M. Pirttilä and S. Compant *et al.*, 2015. The hidden world within plants: Ecological and evolutionary considerations for defining functioning of microbial endophytes. Microbiol. Mol. Biol. Rev., 79: 293-320.
- 9. Anyasi, R.O. and H.I. Atagana, 2019. Endophyte: Understanding the microbes and its applications. Pak. J. Biol. Sci., 22: 154-167.
- Anyasi, R.O., H.I. Atagana and R. Sutherland, 2019. Comparative study of the colonization of *Chromolaena* and tobacco plants by *Bacteria safensis* CS4 using different methods of inoculation. Pak. J. Biol. Sci., 22: 309-317.
- 11. Michel, B.E. and M.R. Kaufmann, 1973. The osmotic potential of polyethylene glycol 6000. Plant Physiol., 51: 914-916.
- 12. Getahun, A., D. Muleta, F. Assefa and S. Kiros, 2020. Plant growth-promoting rhizobacteria isolated from degraded habitat enhance drought tolerance of acacia (*Acacia abyssinica* Hochst. ex Benth.) seedlings. Int. J. Microbiol., Vol. 2020. 10.1155/2020/8897998.
- Sutariati, G.A.K., N.M. Rahni, A. Madiki, L. Mudi, I.M. Guyasa and Zahrima, 2020. Characterization of endophyticrhizobacteria from areca nut rhizosphere to dissolve phosphates, nitrogen fixation of IAA hormone synthesis. Pak. J. Biol. Sci., 23: 240-247.
- Eke, P., A. Kumar, K.P. Sahu, L.N. Wakam and N. Sheoran *et al.*, 2019. Endophytic bacteria of desert cactus (*Euphorbia trigonas* Mill) confer drought tolerance and induce growth promotion in tomato (*Solanum lycopersicum* L.). Microbiol. Res., Vol. 228. 10.1016/j.micres.2019.126302.

- 15. Zandalinas, S.I., R. Mittler, D. Balfagón, V. Arbona and A. Gómez Cadenas, 2018. Plant adaptations to the combination of drought and high temperatures. Physiol. Plant., 162: 2-12.
- Thakur, M., D. Mittal, P.K. Khosla, V. Saini, R.V. Saini and A.K. Saini, 2021. Rhizobacteria associated with *Spilanthes acmella* Murr. Confer drought-tolerance and plant growth promotion. Biointerface Res. Appl. Chem., 11: 13155-13170.
- Forchetti, G., O. Masciarelli, M.J. Izaguirre, S. Alemano, D. Alvarez and G. Abdala, 2010. Endophytic bacteria improve seedling growth of sunflower under water stress, produce salicylic acid and inhibit growth of pathogenic fungi. Curr. Microbiol., 61: 485-493.
- Schloter, M., W. Wiehe, B. Assmus, H. Steindl, H. Becke, G. Höflich and A. Hartmann, 1997. Root colonization of different plants by plant-growth-promoting *Rhizobium leguminosarum* bv. trifolii R39 studied with monospecific polyclonal antisera. Appl. Environ. Microbiol., 63: 2038-2046.
- Sandhya, V., Z.S. Ali, M. Grover, G. Reddy and B. Venkateswarlu, 2009. Alleviation of drought stress effects in sunflower seedlings by the exopolysaccharides producing *Pseudomonas putida* strain GAP-P45. Biol. Fertil. Soils, 46: 17-26.
- 20. Rehman, A. and C.S. Nautiyal, 2002. Effect of drought on the growth and survival of the stress-tolerant bacterium *Rhizobium* sp. NBRI2505 sesbania and its drought-sensitive transposon tn 5 mutant. Curr. Microbiol., 45: 368-377.
- Vardharajula, S., S.Z. Ali, M. Grover, G. Reddy and V. Bandi, 2011. Drought-tolerant plant growth promoting *Bacillus* spp.: Effect on growth, osmolytes and antioxidant status of maize under drought stress. J. Plant Interact., Vol. 6. 10.1080/17429145.2010.535178.

- 22. Marulanda, A., J.M. Barea and R. Azcon, 2009. Stimulation of plant growth and drought tolerance by native microorganisms (AM fungi and bacteria) from dry environments: Mechanisms related to bacterial effectiveness. J. Plant Growth Regul., 28: 115-124.
- 23. Cherif, H., R. Marasco, E. Rolli, R. Ferjani and M. Fusi *et al.*, 2015. Oasis desert farming selects environment-specific date palm root endophytic communities and cultivable bacteria that promote resistance to drought. Environ. Microbiol. Rep., 7: 668-678.
- Theocharis, A., S. Bordiec, O. Fernandez, S. Paquis and S. Dhondt-Cordelier *et al.*, 2012. Burkholderia phytofirmans PsJN primes *Vitis vinifera* L. and confers a better tolerance to low nonfreezing temperatures. Mol. Plant-Microbe Interact., 25: 241-249.
- Mitter, B., N. Pfaffenbichler, R. Flavell, S. Compant and L. Antonielli *et al.*, 2017. A new approach to modify plant microbiomes and traits by introducing beneficial bacteria at flowering into progeny seeds. Front. Microbiol., Vol. 8. 10.3389/fmicb.2017.00011.
- Khan, A.L., M. Waqas, S. Asaf, M. Kamran and R. Shahzad *et al.*, 2017. Plant growth-promoting endophyte *Sphingomonassp.* LK11 alleviates salinity stress in *Solanum pimpinellifolium*. Environ. Exp. Bot., 133: 58-69.
- Lade, S.B., C. Román, A.I. Cueto-Ginzo, L. Serrano, E. Sin, M.A. Achón and V. Medina, 2018. Host-specific proteomic and growth analysis of maize and tomato seedlings inoculated with *Azospirillum brasilense* Sp7. Plant Physiol. Biochem., 129: 381-393.