Arab International Journal of Environmental Sciences (AIJES)
ISSN 2812-5398 (online) – ISSN 2812-5401 (print)
Volume 2, Issue 1, Article Number 4 (2023)
http://doi.org/ 10.21608/aijes.2023.1478

Effect of genetic diversity on adaptation of endangered Sinai Thyme in Saint Katherine Protectorate, Egypt

Asmaa A. Zaghloul[1*], Om Mohamed A. Khafagi[1], Mohamed M. Moursy[2], Hany A. Marghany[2]

[1] Department of Botany, Faculty of Science (Girls branch), Al-Azhar University, Cairo, Egypt

[2] Department of Botany, Faculty of Science (Boys branch), Al-Azhar University, Cairo, Egypt

ARTICLE HISTORY
Received: February 18, 2023
Accepted: July 1, 2023

INDEXING: Arab International Journal of Environment Science – (AIJES), published by General Association for Studies, Research and Technology is covered by Egyptian Knowledge Bank
https://aijes.journals.ekb.eg/

***CORRESPONDING AUTHOR**
Asmaa A. Zaghloul Department of Botany, Faculty of Science (Girls branch), Al-Azhar University, Cairo, Egypt. Email: asmaazaghloul2828.2@gmail.com

CITE THIS ARTICLE
Zaghloul, A.A., Khafagi, A.O., Moursy, M.M., Marghany, H.A. (2023). Effect of genetic diversity on adaptation of endangered Sinai Thyme in Saint Katherine Protectorate, Egypt. Arab International Journal of Environmental Sciences, 2(1): 4. http://doi.org/10.21608/aijes.2023.1478

ABSTRACT Saint Katherine Protectorate (SKP) is located in South Sinai, Egypt. It harbors many endemic and endangered medicinal plants that are overexploited and facing a serious threat of extinction. From these important medicinal plant species, we select Sinai Thyme, *Thymus decussatus* (a near-endemic and endangered plant) to investigate the genetic diversity level among some populations using both random amplified polymorphic DNA (RAPD) and inter-simple sequence repeat (ISSR) molecular markers. RAPD and ISSR represent a level of polymorphism of 86.70%. Assessing genetic polymorphisms among the *Thymus* populations at varying microhabitats can help in their genetic improvement and conservation programs.

Key words: Sinai Thyme, genetic diversity, Saint Katherine Protectorate.

INTRODUCTION

The Sinai Peninsula forms a land bridge between Africa and Asia and its flora and fauna have been influenced by both continental masses; and contains the Saint Katherine Protectorate which covers the mountainous region of Southern Sinai. St. Katherine Protectorate is characterized by the highest mountains in Egypt, a dense wadi system and an arid climate, it was declared as a protected area in 1996 due to its biological and cultural interest (**SKP-MP, 2003**). St. Katherine Protectorate is located between 33º 30’ to 34º 30’ E, and 28° 50’ to 29° 50’N (**Zaghloul *et al*., 2022**) Saint Katherine Protectorate (SKP) located at Southern Sinai is considered one of the largest protected areas in Egypt which included the highest mountains rich with about 44 % of endemic plant species in Egypt. In Southern Sinai, Mountains flora varies from the other areas because of the higher variability in ecosystems, climatic conditions, water environment, and elevations (**Omar, 2014; Soliman *et al*., 2022**). Saint Katherine Protectorate harbors many rare endangered and native plants that are overexploited and facing severe threat of extinction. *Thymus* genus, belonging to family Lamiaceae, integrates 928 species distributed all over the world

and among them about 215 species were mainly distributed in Mediterranean area (**Minarchenko *et al*., 2019**). Owing to its aromatic nature, thyme plant has good antimicrobial, antiparasitic, antispasmodic and antioxidant properties (**Iftikhar *et al*., 2023**). Therefore, their medicinal properties made it one of the most essential and common medicinal herbs (**Iftikhar *et al*., 2023**). Three species known as *T. capitatus*, *T. bovei*, and *T. decussatus* have been reported for *Thymus* genus in Egypt.

Figure 1: Photo of endangered Sinai Thyme (*Thymus decussatus* Benth.) in Saint Katherine Protectorate, Egypt

Effect of genetic variation on plant adaptation

Knowledge of genetic variation among *T. decussatus* populations is critical for their survival and also for better use of their genetic resources. Sinai thyme is distributed in scattered and patches microhabitats **(Khedr, 2021),** where it was restricted to mountains heads at altitudes above 1500 m (**Soliman *et al*., 2022)** and it was growing well in the bases of cliffs and wades. Thyme will be regularly a significant species for corrupted ecosystems particularly arid environments for longer grazing pressure (**Thompson and Gilbert, 2013**).

Genetic polymorphism refers to the degree of genetic variation that can occurs at different scales including populations, subpopulation or even among neighboring individuals and it plays an important role in adaptation of a plant species to environmental stimuli (**Caliebe *et al*., 2022**). Ecogeographical patterns affect the distribution of genetic diversity in a species. Association of genetic diversity with variation in ecogeographical patterns has been detected in several species of aromatic plants (**Youssef and Mahgoub, 2015**).

Determination genetic variation in plant species

DNA markers are used in determining genetic variability in many plant species. Random amplified polymorphic DNA (RAPD), inter simple sequence repeats (ISSRs), simple sequence repeats (SSRs), and amplified fragment length polymorphism (AFLP) are the most widely used DNA markers (**Seehalak *et al.*, 2006**). Guanine-Cytosine content is the percentage of nitrogenous bases in a DNA content has been reported to be a factor deciding a primer's efficiency, since GC content is correlated with annealing temperature and is related to the generation of more DNA fragments (**Cervantes-Salguero, 2022).**

Genetic variability of aromatic and medicinal plant populations has been analyzed using either RAPD or ISSR or both (**Akçali Giachino, 2020**). The genetic diversity among the different accessions or populations of thyme plant using either RAPD or ISSR or both molecular markers was evaluated in different studies. Genetic variation within and among eight populations of the endemic Tunisian *Thymus algeriensis* plant species collected from variant geographical habitats was assessed using seven RAPD primers (**Caliebe *et al.*, 2022**). Their results recorded presence of high genetic variation within and among the studied populations.

RAPD markers were also used in genetic identification of five accessions of Palestinian *Thymus* for their ability to produce polymorphisms (**Solyman and Alkowni, 2014**). Moreover, genetic diversity of ten *Thymus kotschyanus* **populations** obtained from various ecosystems in Iran was analyzed using RAPD markers (**Khoshsokhan *et al.*, 2014**).Their findings showed high level of genetic diversity among the ten studied populations of *T. kotschyanus*. Even, twenty ISSR primers were applied for evaluation of the genetic relationships among fourteen *Thymus* accessions belonging to three *T. daenensis*, *T. kotschyanus* and *T. vulgaris* species (**Yousefi *et al.*, 2015**). Their findings showed high level of genetic diversity among populations.

The establishment of efficient conservation strategies for growing plants (**Salgotra and Chauhan, 2023**).Thus, great efforts should be carried out on preserving the rare endangered species as well as the endemic species with restricted geographical distribution (**Zhu *et al.*, 2023**). From these near-endemic and endangered plant species, *T. decussates* plant species growing in Saint Katherine Protectorate was selected in this study to assess the degree of genetic variation among their populations using both of RAPD and ISSR molecular markers. RAPD and ISSR markers have been successfully applied in assessing the extent of genetic diversity among eight *T. decussatus* populations collected in Saint Katherine Protectorate, Southern Sinai, Egypt from eight microhabitats (**Mahgoub *et al.*, 2020**). **Mahgoub *et al*. (2020)** suggested all primers with variable percentages of polymorphism with an average of 86.21 % were found to be polymorphic. **Mahgoub**

***et al*. (2020)** were entirely compatible with results on five Palestinian Thymus species, *T. kotschyanus*, *T. caespititius*. In addition, numerous research studies on Thymus species from several countries showed that the rate of polymorphism ranges from 62% to 92% (**Saboh *et al*., 2019**). Nevertheless, the polymorphism level obtained in **Mahgoub *et al*. (2020)** is rather lower than the recorded average polymorphism (94.31%), which examined genetic polymorphism in 13 Thymus accessions collected from different habitats in Iran using 20 RAPD primers **(Yousefi *et al*., 2015).**

Mahgoub *et al*. (2020) showed that the number of DNA fragments produced per primer ranged from seven to ten which are close to the result obtained in their analysis on populations of Thymus vulgaris. High level of polymorphism recorded in our study reflects the great degree of genetic variation among Thymus populations collected from various microhabitats in Saint Katherine Protectorate (SKP). Such heterogeneity may be due to the high degree of gene exchange between the studied populations (**Khoshsokhan *et al*., 2014**).

Numerous studies have documented that the RAPD method is an effective tool for thyme taxa genome. The analysis of a large number of DNA fragments can be considered as a good representation of the *Thymus* taxa genome and the ability to distinguish all the samples studied by RAPD banding profiles indicates that this technique may provide a faster and cheaper way for the identification of the *Thymus* taxa (**Mahgoub *et al.* 2020**). **Mahgoub *et al.* (2020)** showed high polymorphic among the Thyme populations surveyed (87.18 %) by ISSR markers. The polymorphism has an average of 88.9% in *Thymus daenensis* (**Rahimmalek *et al*., 2009).**

In another study also it was recorded that the number of polymorphic bands ranged from 75% to 100%, with an average of 96.76% between fourteen *Thymus* accessions belonging to three species (*T. daenensis*, *T. kotschyanus*, and *T. vulgaris*) (**Sarfaraz *et al*., 2020**)

ISSR markers can be a highly insightful, efficient, and reliable method for genetic diversity trials (**Iqbal *et al*., 2023**). The efficacy of the molecular marker techniques depends primarily on the degree of polymorphism produced by the primers used. To characterized the degree of genetic variation among species of *T. decussatus* populations (**Mahgoub *et al*., 2020**).

RAPD and ISSR markers were appeared genetic variation among the populations of Thymus decussates with an average of 86.70% (**Mahgoub *et al*., 2020**). RAPD and ISSR markers were used in similar studies to assess the genetic variation among *Nepeta septemcrenat*, *Thymus decussates* and *Phlomis aurea* plant species collected from Saint Katherine Protectorate, South Sinai, Egypt (**EL-Sadek *et al*., 2017**)**.** The primers of ISSR produced more polymorphic bands than primers of RAPD, hence ISSRs were found to be more efficient than RAPD in

estimating genetic diversity (**EL-Sadek *et al*., 2017**).. Similar results also have been obtained for other plants including Salvia (**Javan *et al*., 2012**)

(**Mahgoub *et al*., 2020**) support the fact that there are SKP region-specific variations in relation to soil and climatic adaptation cycle. Also **Catană *et al*. (2013)** support this fact in their study on endemic plant species (*Draba dorneri Heuff* and *Commiphora wightii*), in their study on three medicinal plant species belongs to the Lamiaceae family (*Nepeta septemcrenata*, *Ballota undulate* and *Teucrium polium*) which grows naturally in Saint Catherine Mountain at three different altitudes. All of these result indicated that there are relationship between geographical distributions and the genetic diversity.

There are high genetic variation occurs between *T. decussatus* populations as recorded in **Mahgoub *et al*. (2020)** contrasts with mountain plants. For endemic plants from Tibet and Central Asian desert plants and several other studies of outcrossing endemic species, it was found that there was high genetic variability in colonies than in habitats (**Chen *et al*., 2009**). Differences in polymorphism among restricted species may be linked to the degree to which their populations, in accordance with occur in heterogeneous habitats (**Wang and Zhang, 2022**). Despite the short geographical distances which separate the current *T. decussatus* populations, high genetic variation was observed between the populations studied using both RAPD and ISSR markers (**Mahgoub et al., 2020).**

Also, the extent and distribution of genetic diversity is significantly influenced by the mating mechanism and reproduction mode. Higher homozygosity results in low levels of genetic variation in self-compatible species, while species with primarily outcrossing mating systems experience higher rates of genetic variation (**Pluhár *et al*., 2012**).

CONCLUSION

Both RAPD and ISSR analyses showed the same percentage polymorphism, as 86.21 percent for RAPD markers and 87.18 percent for ISSR markers It can be concluded that amongst the Thymus populations rising in the Saint Katherine region there is a high genetic diversity. Specific geographical and ecological conditions permit certain potential genetic modifications or DNA changes such as translocation, deletion, point mutation and so on. The molecular analysis using markers RAPD and ISSR showed that both markers were able to differentiate between the different populations of species as *T. decassatus* and can thus be used to research the degree of genetic variation between species as *T. decussatus*.

Reference

Akçali Giachino, R. R. (2020). Investigation of the genetic variation of anise (*Pimpinella anisum* L.) using RAPD and ISSR markers. Genetic Resources and Crop Evolution, 67(3): 763-780.

Caliebe, A., Tekola-Ayele, F., Darst, B. F., Wang, X., Song, Y. E., Gui, J., Sebro, R. A., Balding, D. J., Saad, M., Dubé, M., IGES ELSI Committee. (2022). Including diverse and admixed populations in genetic epidemiology research. Genetic epidemiology, 46(7): 347-371.

Catană, R., Mitoi, M., Ion, R. (2013). The RAPD techniques used to assess the genetic diversity in *Draba dorneri*, a critically endangered plant species. Advances in Bioscience and Biotechnology, 4, 164-69.

Cervantes-Salguero, K., Biaggne, A., Youngsman, J. M., Ward B. M., Kim, Y. C., Li, L., Hall, J. A., Knowlton, W. B., Graugnard, E., and Kuang, W. (2022). Strategies for controlling the spatial orientation of single molecules tethered on DNA origami templates physisorbed on glass substrates: Intercalation and stretching." International journal of molecular sciences, 23(14): 7690.

Chen, F. J., Wang, A. L., Chen, K. M., Wan, D. S., Liu, J. Q. (2009). Genetic diversity and population structure of the endangered and medically important *Rheum tanguticum* (Polygonaceae) revealed by SSR markers. Biochemical Systematics and Ecology, 37, 613–21.

EL-Sadek, L. M., El-Badan, G., Shabana, H. A. (2017). Genetic diversity of the endemic species *Phlomis aurea* Decne. in southern Sinai, Egypt. Egyptian Journal of Botany, 57(7th International Conf.), 147-159.

Iftikhar, T., Majeed, H., Zahra, S. S., Waheed, M., Niaz, M., Bano, N. (2023). Thyme. In Essentials of Medicinal and Aromatic Crops. Cham: Springer International Publishing, 399-429.

Iqbal, J., Altaf, M. T., Jan, M. F., Raza, W., Liaqat, W., ul Haq, I., Mehmood, A. (2023). Exploring genetic diversity in cotton genotypes using EST-SSR and ISSR markers: A comparative study. Sarhad Journal of Agriculture, 39(4): 800-814.

Javan ZS, Rahmani F, Heidar R. (2012). Assessment of genetic variation of genus Salvia by RAPD and ISSR markers. Australian J Crop Science, 6(6): 1068-73.

Khedr, A. H. A. (2021). Microhabitats supporting endemic plants in Sinai, Egypt. In Management and Development of Agricultural and Natural Resources in Egypt's Desert. Cham: Springer International Publishing, 369-381.

Khoshsokhan, F., Babalar, M., Fatahi, M., Poormeidani, A. (2014). Assessment of genetic diversity of some wild populations of *Thymus Kotschyanus* using RAPD molecular markers. Cercetări

Agronomice în Moldova, 3(159): 71–81.

Khoshsokhan, F., Babalar, M., Fatahi, M., Poormeidani, A. (2014). Assessment of genetic diversity of some wild populations of Thymus Kotschyanus using RAPD molecular markers. Cercetări Agronomice în Moldova, 3(159): 71–81.

Mahgoub, H. A., Moursy, M. M., Zaghloul, A. A., Khafagi, O. M. A. (2020). Molecular Characterization of Endangered Sinai Thyme (*Thymus Decussatus* Benth.) Growing on Gebel Mousa, South Sinai, Egypt. Al-Azhar Bulletin of Science, 31(2-C): 35-51

Minarchenko, V. M., Lysiuk, R. M., Kovalska, N. P. (2019). Medicinal plant resources: textbook. Kyiv: Palyvoda A. V, 240.

Omar K. A. (2014). Ecological and climatic attribute analysis for Egyptian *Hypericum sinaicum*. American Journal of Life Sciences, 2(6): 369–81.

Pluhár, Z., Kocsis, M., Kuczmog, A., Csete, S., Simkó, H., Sárosi, S., Molnár, P., Horváth, G. (2012). Essential oil composition and preliminary molecular study of four *Hungarian Thymus* species. Acta Biologica Hungarica, 63(1): 81–96.

Rahimmalek, M., Bahreininejad, B., Khorrami, M., Tabatabaei, B. (2009). Genetic variability and geographic differentiation in *Thymus daenensis* subsp. *daenensis*, an endangered medicinal plant, as revealed by inter simple sequence repeat (ISSR). Markers. Biochemical Genetics. 47(11–12): 831–42.

Saboh, M., Ameen, T., Mahfoud, H. (2019). Genetic diversity in some wild *Thymus* species distributed in Latakia (Syria) as detected by RAPD markers. Tishreen University Journal for Research and Scientific Studies- Biological Sciences Series, 14(3): 77-89.

Salgotra, R. K., Chauhan, B. S. (2023). Genetic diversity, conservation, and utilization of plant genetic resources. Genes, 14(1): 174.

Sarfaraz, D., Rahimmalek, M., Saeidi, G., Sabzalian, M. R. (2020). Genetic relations among and within wild and cultivated Thymus species based on morphological and molecular markers. 3 Biotech, 10(7): 289.

Seehalak, W., Tomooka, N., Waranyuwat, A., Thipyapong, P., Laosuwan, P., Kaga, A., Vaughan, D. A. (2006). Genetic diversity of the Vignagermplasm from Thailand and neighbouring regions revealed by AFLP analysis. Genetic Resources and Crop Evolution. 53(5): 1043-59.

SKP MP (2003). Saint Katherine protectorate management plan. Reference editioned. EGYPT:

nature conservation sector-Egyptian Environmental Affairs Agency (EEAA).

Soliman, A., Sheta, B. M., Bahnasway, M., Orabi, G. M. (2022). Avifaunal updated survey in St. Catherine protectorate, Egypt. Journal of Medical and Life Science, 4(1): 9-17.

Solyman, E., Alkowni, R. (2014). RAPD for assessment of thymes genetic diversity in Palestine. Palestine Technical University Research Journal, 2(2): 01–08.

Thompson, K., Gilbert, F. (2013). The effects of grazing on the endangered Sinai Thyme (*Thymus decussatus*) in a hyper-arid environment. Journal of arid environments, 99, 14-22.

Wang, Q., Zhang, H. X. (2022). Population genetic structure and biodiversity conservation of a relict and medicinal subshrub Capparis spinosa in arid Central Asia. Diversity, 14(2), 146.

Yousefi, Y., Najaphy, A., Zebarjadi, A., Safari, H. (2015). Molecular characterization of *Thymus* species using ISSR markers. The Journal of Animal & Plant Sciences, 25(4): 1087–94.

Yousefi, Y., Najaphy, A., Zebarjadi, A., Safari, H. (2015). Molecular characterization of *Thymus* species using ISSR markers. The Journal of Animal & Plant Sciences, 25(4): 1087–94.

Youssef, M. A., Mahgoub, H. A. (2015). Phytochemical and molecular analysis of some medicinal plants of Labiatae family growing at different altitudes on Saint Katherine Mountain, South Sinai, Egypt. Egyptian Journal of Genetics And Cytology, 44: 331-56.

Zaghloul, A. A., Khafagi, O. M. A., Moursy, M. M., & Marghany, H. A. (2022). Vegetation Characteristics and Environmental Gradients of Musa Mountain in Saint Katherine Protectorate, South Sinai, Egypt. The Arab International Journal of Environmental Sciences, 1(1), 1-14.

Zhu, Y., Xu, X., Xi, Z., Liu, J. (2023). Conservation priorities for endangered trees facing multiple threats around the world. Conservation Biology, 37(6): e1414