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The Assessment of the *Allium sativum* and *Tamarix aphylla* Comparative and Combined Antioxidant Potential

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Many diseases, including cancer and diabetes mellitus, are caused by reactive oxygen species (ROS). *Allium sativum* (Garlic) contains vitamins A, B, and C, as well as effective drugs like insulin, alliin, mineral salts, mucilages, allicin, and volatile oils. Garlic has antioxidant properties which showed a therapeutic effect on some cancer types. The overall goal of this study was to conduct pharmacological testing to assess the combined antioxidant abilities of *Allium sativum* (cloves) methanolic and *Tamarix aphylla* (leaves) extracts. The extract demonstrated garlic activities in a dose-dependent manner, with scavenging activity of 21, 32, and 39% at different concentrations of 100, 500, and 1000 µg/mL.

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The antioxidant activity of *Tamarix aphylla* methanolic extract was 29, 37, and 62% using the DPPH free radical scavenging assay at different concentrations of 100, 500, and 1000 µg/mL. However, combining extracts revealed the greatest scavenging activity at various concentrations of 100, 500, and 1000 µg/mL. By using the DPPH free radical scavenging assay, the combined methanolic extract of *Tamarix aphylla* and *Allium sativum* demonstrated substantial antioxidant activity of 45, 65, and 75% at concentrations of 100, 500, and 1000 µg/mL. According to our findings, the combined therapy of *Tamarix aphylla* and *Allium sativum* significantly inhibited DPPH free radicals. *Tamarix aphylla* and *Allium sativum* combined therapy may play an important role in the inhibition of free radicals that cause cancer.

Keywords: Allium sativum; Tamarix aphylla and antioxidant activity.

1. INTRODUCTION

Cancer is one of the life-threatening disease, and the discovery of new anti-cancer drugs is one of the most promising natural product research areas [1]. Garlic, botanically known as *Allium sativum*, is a gramineous herb that contains several compounds those are similar to onion compounds [2]. *Allium sativum* L. (garlic) is a widely consumed herb due to its therapeutic and aromatic properties [3]. Garlic contains vitamins A, B, and C, as well as effective drugs like insulin, alliin, mineral salts, mucilages, allicin, and volatile oils. Garlic has antioxidant properties and has been shown to be beneficial to the cardiovascular and immune systems due to its antimicrobial, antifungal, antibacterial, and antiviral properties [4]. Antioxidants are stable molecules that protect cells from the oxidation of unstable molecules (free radicals). Oxidative stress is characterized by an imbalance in the production and accumulation of reactive oxygen species (ROS). Many diseases, including cancer, diabetes, atherosclerosis, gynecological diseases, and especially endometriosis, are caused by reactive oxygen species (ROS) [5-9]. People are becoming more interested in herbal medications due to the fact that they have less or no side effects. Although the low risk of side effects, the prospect of a medication interaction cannot be ruled out. Herbal medications have been used throughout history, but there is a gap in the old traditional methods that complementary and alternative medicine incorporating new technologies is filling [10-13]. Consequently, we should keep searching for novel plant-based treatments that comprise huge information about nature.

It is necessary to search the vast reservoir of nature for new plant-based cancer therapies. *Tamarix aphylla*, a member of the Tamaricaceae family, is the mainly prominent *Tamarix* species, getting heights of up to 18 meters (60 feet). *Tamarix aphylla* has been found to contain flavonoid glycosides, carboxylic acid steroids, cardiac glycosides, terpenoids, and some selective secondary metabolites [14, 15]. It has a high potential for anti-diabetic, anti-bacterial, anti-inflammatory, and anti-fungal properties, as well as periodontal disease, anti-cholinesterase, and wound-healing properties [16]. The anti-diabetic, antibacterial, anti-inflammatory, and antifungal qualities of this plant has significant potential, and it also contains anti-cholinesterase, wound-healing, and periodontal disease-fighting characteristics. This plant contains numerous phenolic compounds with astringent effects [17]. Aside from its traditional uses, the plant has anti-diabetic, hypolipidemic, antifungal, antibacterial, cytotoxic, and antioxidant properties [11, 18, 19].

2. MATERIALS AND METHODS

2.1 Chemicals for Biological Activities

2, 2- diphenyl-1-picrylhydrazyl (DPPH) was purchased from Sigma Germany. The dried garlic fruit methanolic extract, aluminum foil and double beam spectrophotometer were used.

2.2 The Preparation of Methanolic Extract of *Allium sativum* and *Tamarix aphylla* Plant

The plants were collected in December 2021 from the District of Bannu in Pakistan. After the shade dried for 25 days, with the aid of a pestle and mortar, *Allium sativum* and *Tamarix aphylla* were grinded into a fine powder. The powders were immersed in 70% methanol in such a way that it was totally submerged in methanol, and then kept at room temperature for 72 hours with regular agitation, and the obtained mixture was filtered with help of Whatman No. 3 filter paper. After filtration, the obtained solution was kept at

room temperature for drying. After drying the gummy methanolic extract was lyophilized in a falcon tube. The lyophilized samples were kept for future use.

2.2 Antioxidant Assay

For the DPPH assay, the published protocol was followed with minor modifications [20]. 100µl from each of the samples including ascorbic acid solution of 100 µg/mL, 500 µg/mL, and 1000 µg/mL were taken and mixed it with 900 µL of DPPH. All these test tubes were incubated at 25°C for about 30 minutes in the dark because of their sensitivity to word light and checked the absorbance on double beam spectrophotometer at 517nm. By using the following equation the potential of the samples to scavenge the DPPH free radicals was calculated;

% DPPH free radicals scavenging effect = $(A_1-A_2/A_1)\times100$

Where A_1= the absorbance of DPPH (control) and A_2= the absorbance in the presence of samples.

3. RESULTS

3.1 Antioxidant Therapy of *Allium sativum*

The ability of DPPH to scavenge shows the following results with different concentrations of methanolic extract of the plant, which are 100 µg/mL, 500 µg/mL, and 1000 µg/mL, respectively. By increasing the extract concentration also increases i.e. 20% at minimum concentration while 40% and 60% with a maximum concentration of *Allium sativum* as shown in Fig 1. We noticed that the *Allium sativum* has less antioxidant activity than the standard solution in the DPPH free radicals assay.

3.2 Antioxidant Therapy of *Tamarix aphylla*

We compared the antioxidant activity of the extract of *Tamarix aphylla* with standard ascorbic acid using the DPPH free radicals assay. The ability of DPPH to scavenge shows the following result with different concentrations of plant extract, which are 100 µg/mL, 500 µg/mL, and 1000 µg/mL, respectively. By the increasing of the extract concentration also enhanced the scavenging properties i.e. 29% at minimum concentration while 37% and 62% with a maximum concentration of *Tamarix aphylla* as shown in Fig 2.

3.3 Antioxidant Therapy of Combined Plants such that *Allium sativum* and *Tamarix aphylla*

Furthermore, we examined the combined methanolic extracts of *Allium sativum* and *Tamarix aphylla* with standard ascorbic acid using DPPH free radicals assay. The capability of scavenging property of DPPH shows the following result with the different concentrations of methanolic extracts of the plants are 100 µg/mL, 500 µg/mL and 1000 µg/mL respectively. By increasing the order of activity of combined plants extraction also increases i.e. 60% at minimum concentration while 60% and 80% with a maximum concentration of *Tamarix aphylla* and *Allium sativum* as shown in Fig 3.

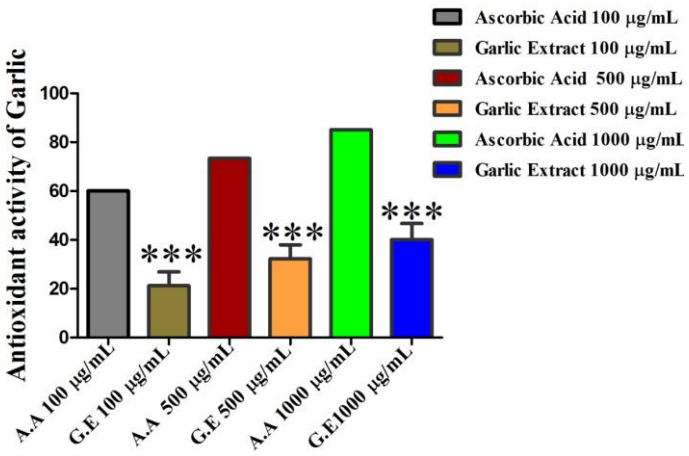

Fig. 1. Antioxidant therapy of *Allium sativum*

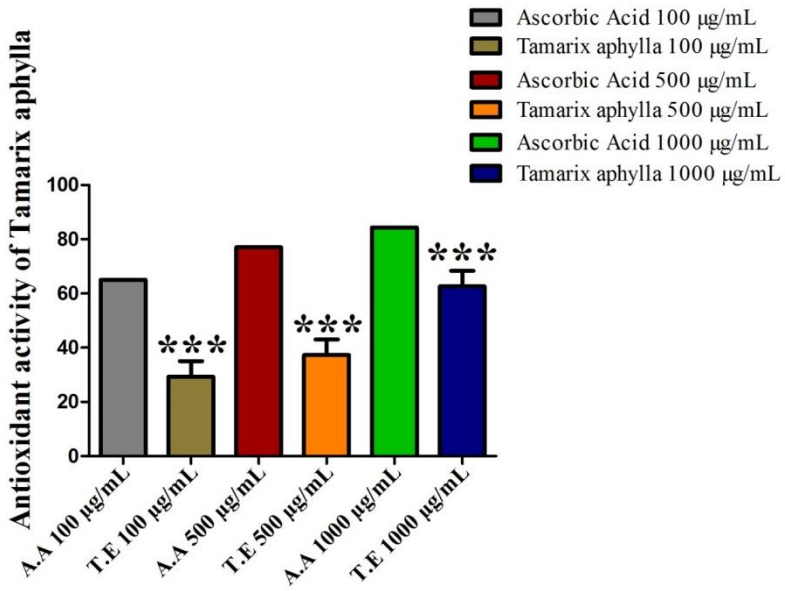

Fig. 2. Antioxidant activity of *Tamarix aphylla*

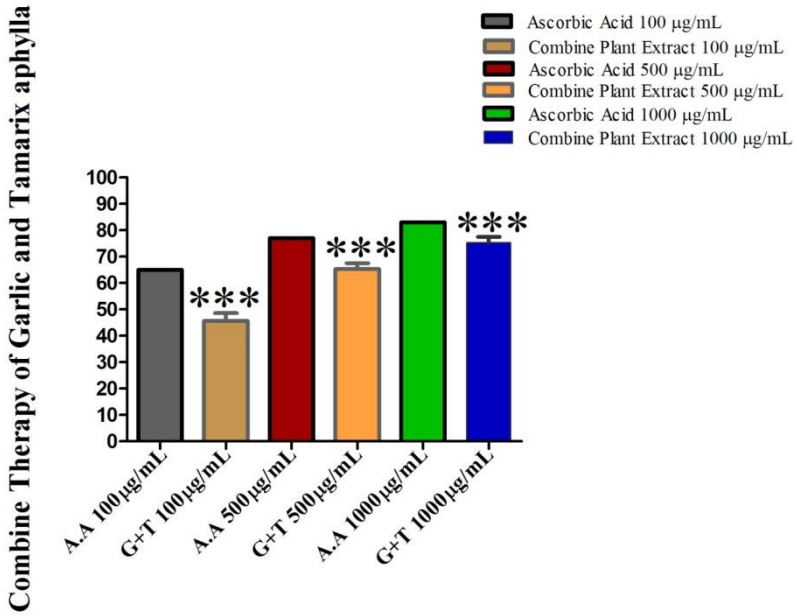

Fig. 3. Combined antioxidant activity of *Allium sativum* and *Tamarix aphylla*

4. DISCUSSION

Cancer is a group of life-treating diseases and findings of new anti-cancer drugs are one of the most emerging research areas of natural products [1]. Plants are well known for their medicinal expertise since ancient times. Plant-based medicine and traditional medicine are practiced nearly by half of the population in developing and underdeveloped nations. Antioxidants are crucial for human health since they boost immunity by being able to get rid of dangerous free radicals created by regular cellular metabolism and stressful environments. Spices and herbs are the most antioxidant-rich foods in the human diet, according to the Antioxidant Food Database, which was created by analyzing 3,100 different food products [21]. Garlic, an *Allium* vegetable, contains rich flavonoid and organo-sulfur compounds (OSCs) that have potent anti-cancer properties [22].

Novel bioactive metabolites from *A. sativum* has been recently identified as potent anti-cancer drug targets [23]. The most prevalent and widely dispersed class of secondary plant metabolites, polyphenols are thought to help reduce oxidative stress and inflammatory disorders [24]. It has been demonstrated that the phenolic chemicals in garlic's structure give it a high level of antioxidant action [25]. As demonstrated in Fig.1, the antioxidant activity of *A. sativum* was significantly high and correlated with the results of a previous study.

Due to the presence of active phytochemical substances like flavonoids or polyphenols, alcohol extract of *T. aphylla* leaves in Saudi Arabia has antioxidant, anti-inflammatory, and wound-healing properties [26]. Phenolic compounds play a crucial role in the prevention of lipid peroxidation in actual meat products, because of position and quantity of hydroxyl groups in phenolic compounds determine their capacity to scavenge free radicals, chelate iron, and have a lowering effect [27]. Furthermore, we examined the antioxidant activity of *T. aphylla* which was critical, as shown in Fig. 2. Next, we examined the antioxidant activity of both plant extracts mixture. The combined extract of Garlic and *T. aphylla* showed significant antioxidant results as shown in Figure 3. The plants can be considered as possible sources of anti-cancer compounds [20]. Further studies are required for the chemical characterization of the active compounds and more broad biological evaluations.

5. CONCLUSION

In conclusion, *Tamarix aphylla* extracts exhibited better antioxidant properties than garlic extracts. However, the methanolic mixture of both plants extracts significantly enhanced the antioxidant activity. Further study will focus on how they apply to meat and animal products.

CONSENT AND ETHICAL APPROVAL

It is not applicable.

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This study was carried out at the research laboratory of the Zoology Department, University of Science and Technology Bannu (28100), Kp, Pakistan.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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