

A Review of Secondary Metabolites and Inhibitory Effect of Medicinal Plants on some Plant Pathogens

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Article History: Received: 25 December 2023/Accepted in revised form: 22 January 2024 © 2012 Iranian Society of Medicinal Plants. All rights reserved

ABSTRACT

Effective compounds in medicinal plants include alkaloids, glycosides, volatile oils (essential oils) and other effective compounds (bitter compounds, flavones, flavonoids, mucilages, vitamins, tannins, salicylic acid). The use of these metabolites is considered as one of the new methods of controlling, preventing or treating plant diseases. Plant extracts and essential oils have antifungal and antibacterial effects and so far no microbial resistance has been reported against them. In this article, the antifungal properties of different concentrations of extracts and essential oils was reviewed, analyzed and discussed. The authors searched the main related keywords of medicinal plants in main biological data centers e.g. Science Direct, Pubmed and Google Scholar and then the articles classified and only the valid full papers about inhibitory effects of medicinal plants on plant pathogens were discussed. The importance of medicinal plants in controlling plant pathogenic agents is due to the presence of compounds and their effective substances under the title of secondary metabolites such as terpenes, phenolic compounds, and nitrogenous (alkaloids, morphine, codeine and nicotine) compounds can inhibit the growth of plant pathogens. Different studies have proven the role of the active ingredients of medicinal plants in controlling of plant pathogens and several extracts and oil essential of medicinal plants with effective active ingredients were used in controlling various plant pathogens. According to the results of different researches, the use of extracts and oil essential of medicinal plant to inhibit the growth of plant pathogens and plant diseases can be recommended. Considering the abundance and diversity of medicinal plants in Iran, further studies on the effects of the active ingredients of native medicinal plants in controlling a wider range of plant pathogenic agents are suggested in order to obtain plant-based pesticides.

Key words: Alkaloid, Disease, Essential oil, Fungi, Medicinal plants, Phenolic compounds

INTRODUCTION

Medicinal plants have had a special place in the traditional agricultural system of Iran for a long time, and the use of these plants as medicine for the prevention and treatment of diseases has attracted the attention of traditional medicine specialists since ancient times. Medicinal plants with rich sources of secondary metabolites provide the basic active ingredients of many drugs. Although the biosynthesis of secondary metabolites is genetically controlled, but their production is strongly influenced by environmental factors [20]. The tendency to use medicinal plants is increasing day by day, because many synthetic drugs and chemicals used in modern medicine have many side effects and their effects are temporary. In addition to the growing importance of medicinal plants in the world, which are quickly replacing many chemical drugs, the export of these plants can also be a great source of foreign exchange income for the country. Iran, with its historical background and potential geographical, climatic and plant species diversity, can respond to the needs of human society in the field of medicinal plants. The increasing importance, position and role of medicinal plants in sustainable management, especially in the macro dimensions of economic, environmental, health (medicinal self-sufficiency),

employment, food security and genetic resources in the national and global arena, is such that it can be done today. Deepening, revitalization and its role, especially in drug supply, were taken into consideration as one of the indicators of development in the country [24].

Plant extracts, as environmentally friendly compounds and substitutes for chemical poisons, can play a significant role in the management of plant diseases [10]. In recent years, the use of secondary plant compounds has flourished. These compounds are used to protect plants from diseases before and after harvest [8]. Medicinal plants have a special place in the traditional agricultural systems of Iran. The low side effects of these drugs and their different effective compounds have caused them to be of special importance and characteristics despite the presence of drugs with chemical origin of medicinal plants. Medicinal plants are rich reservoirs of effective substances and secondary metabolites. Plant essential oils, which include a wide range of secondary metabolites, have broad antimicrobial activities against bacteria, fungi and viruses. Therefore, they have many applications in the fields of pharmaceuticals, medicine and food industries. It has been proven that the use of these compounds in the agricultural sector, in addition to increasing yield and improving the quality of manufactured products, will improve the level of community health and reduce adverse environmental effects. Therefore, it can be said that the biological and pharmacological activities of these compounds have led to a greater desire for scientific knowledge of these substances and have multiplied the need for studies related to them [16]. Therefore, the importance of medicinal plants in controlling plant pathogenic agents is due to the presence of compounds and their effective substances under the title of secondary metabolites, and [26] stated that the effective compounds in medicinal plants can be divided into four main groups: alkaloids, glycosides, volatile oils, essential oils) and other effective compounds. Other effective compounds are compounds such as bitter compounds, flavones, flavonoids, mucilages, vitamins, tannins, salicylic acid. Each of these mentioned natural compounds have unique properties that distinguish them from chemical compounds. Many of these compounds have been used to prevent or treat human, animal and plant diseases. In a research on Borago officinails L., the percentage of mucilage was 7.167% [21], the amount of phenol was 7.72% [22], and the amount of seed oil of this plant was estimated to be 29.38% [23].

STATEMENT OF THE PROBLEM AND RESEARCH METHOD

The emergence of the phenomenon of resistance to all kinds of synthetic chemical pesticides, the poisoning caused by the use of chemical pesticides to animals, aquatic animals and beneficial insects, as well as the negative effects of toxic residues, have created many problems for human health and the environment [1], [11]. In recent years, the use of herbal products in preventing pathogenic agents such as viruses, bacteria, fungi and parasites has been widely considered. Some fungi are among the pathogenic agents that can cause problems in humans, animals and plants. Plant extracts are among the substances that have been used as antifungal products [40]. Today, the biological control of plant pathogens with the aim of reducing the dangerous effects of chemical pesticides, including the threat to human health, environmental pollution, eliminating non-target organisms and the emergence of resistant pathogens, is a priority. The degradability of plant extracts in nature and their low toxicity for humans and other mammals and their less harmful effects on the environment have turned these compounds into alternatives or supplements to chemical poisons for the protection of agricultural and storage products. Due to the sensitivities in the use of synthetic chemical compounds (due to the threat to human health and the environment), as well as the increasing demand for organic products (produced without the use of chemical inputs) and also due to the creation of new and resistant strains of pathogens in Compared to the used fungicides, today, efforts are being made to use some natural materials and products without harmful side effects, such as plant extracts and essential oils, as an alternative to chemical fungicides in controlling or reducing Diseases and storage waste of garden products should be used. Using chemicals is one of the most effective, fastest and most accessible ways to control plant diseases. But due to their environmental pollution, as well as endangering human health and creating resistance in pathogenic agents, researchers are looking for alternative methods to control plant diseases. Currently, the use of extracts and essential oils is considered as one of the new methods of controlling plant diseases. These materials are of natural origin, which do not cause environmental pollution, and so far no microbial resistance has been reported against plant extracts and essential oils. These compounds have a high potential to be used in the integrated management of diseases, and considering the problems in the use of fungicides, studying and researching new, safe and low-cost methods for controlling and managing plant diseases is a necessity. Plant extracts and essential oils have been used against various plant pathogens and successful results have been presented. In this regard, the use of the antimicrobial potential of plant metabolites has received attention in recent years [15] and this review was conducted under the title: A review of Secondary Metabolites and Inhibitory Effects of Medicinal Plants on some Plant Pathogens, and based on this review research, at first, the metabolites and compounds in medicinal plants was expressed, then the effects of plant extracts and essential oils on the control of plant pathogens were discussed. For this purpose the main related keywords of medicinal plants in main biological data centers e.g. Science Direct, Pubmed and Google Scholar were searched. Then the articles classified and only the valid full papers about inhibitory effects of medicinal plants on plant pathogens were discussed.

SECONDARY PLANT COMPOUNDS

Secondary metabolites are divided into three distinct chemical groups: terpenes, phenolic compounds, and nitrogenous compounds. Their abundance can be seen in (Diagram 1). This diagram shows secondary metabolites in plants that are known so far. As can be seen, terpenoids are the most abundant (31%), followed by alkaloids (28%) and flavonoids (13%), respectively [25].

Terpenoids

Terpenoids are the most structurally diverse natural plant compounds (Figure 1). These compounds are very important from a commercial point of view due to their wide application in industrial products such as seasonings, medicines, perfumes and insecticides and antimicrobial agents. Because of their unique properties, terpenoids are used as biological materials in industry to produce heavy duty tires, shock absorbers, and latex products such as surgical gloves [17]. Other examples of plant terpenoids with significant economic value include: Menthol, a monoterpenoid extracted from mint and used in the flavoring industry. Abscisic acid, a diterpenoid isolated from conifer rosin, used in lacquer, varnishes and soap industry. Artemisinin and Taxol, which are used as antimalarial and anticancer drugs [5]. The term terpene refers to hydrocarbon molecules, while terpenoids are terpene molecules that have undergone further changes. Terpenoids are the most abundant and diverse natural compounds known in plants. These precious compounds have many roles in the plant itself, including the protection of plants against herbivores, the attraction of pollinators, the coexistence of plants with other organisms, and the competition of plants with other plants. Taxol (Figure 2) is a very important cyclic diterpenoid that was extracted from the yew tree (Taxus brevifolia) at Year 1962. The molecular structure of this compound was determined at Year 1971 by X-ray crystallography. Today, this compound is used as an important anticancer drug in chemotherapy. The anticancer mechanism of this compound is that by binding to the subunits of microtubules, it prevents their polymerization, therefore, during cell division, it causes the chromosomes to not separate properly and by disrupting cell division, it prevents cell growth [18].



Fig. 1 Percentage of secondary metabolites in plants



Phenolic compounds

Phenolic compounds are one of the most important categories of compounds whose immune system stimulating effects have been proven [12]. Phenolic compounds are one of the most common groups of plant metabolites. Plant phenolic compounds can act as antioxidants, structural polymers, absorbers, UV screens, and defense reaction chemicals. Also, these phytochemicals have many benefits for human health. From a physiological point of view, phenolic compounds have antioxidant, anti-inflammatory, anti-aging and anti-cancer activities [41]. This group of effective substances have phenol in their structure. The chemical structure of phenols is shown in Figure (3). Some of them are only soluble in organic solvents, some are soluble in water, and some of them are insoluble. These compounds are found in plants in free and mostly ester or glycosidic forms. Among the most important phenolic compounds, we can mention flavonoids and coumarins.



Fig. 3 Chemical structure of phenol

Flavonoid

The term flavonoid in a broad sense includes all plant pigments. About 4,000 types of flavonoids have been identified in higher plants, especially in plants, of which more than 90 types of flavonoids are found in citrus fruits and more than 30 types in the composite family. So far, no flavonoids have been reported in algae. Among the phenolic compounds, flavonoids have all the actions of secondary metabolites in plants. Flavonoids are phenylpropanoid derivatives that have a 15-carbon structure. In most flavonoids, ring A is metadihydroxyl or metatrihydroxyl, while ring B has one, two or three hydroxyl groups. This difference is caused by the origin of the biosynthesis of two rings. The A ring is derived from three condensed acetic acid molecules and the B ring is derived from sugars in the shikimic pathway [19]. Flavonoids cause coloration in flowers, fruits and sometimes leaves. They are also effective in plant pollination and fertility due to their ability to attract insects. Flavonoids increase the resistance to pathogenic agents in plants and are also strong absorbers of ultraviolet rays (250-340 nm). Plants such as *Achillea millefolium, Crataegus monogyna, Ginkgo biloba, Thymus vulgaris* L, *Hypericum perforatum, Chamaemelum nobile, Punica granatum, Camellia sinensis, Crataegus persica, Laurus nobilis, Passiflora*, citrus, *Glycyrrhiza glabra* are among the valuable sources of flavonoids. One of the most important flavonoids is anthocyanin (Figure 4).



Fig. 4 Anthocyanin chemical structure

Coumarin

Some plant phenols participate in the phenomenon of allelopathy as allelochemical compounds, they affect the germination, growth and reproduction of other plants, which often results in the reduction of crops. On the other hand, phenolic compounds such as coumarin can be used to control pests, diseases, and weeds. Coumarins are a group of phenolic compounds that are considered as phenylpropanoid lactones, and simple coumarin (Figure 5) is considered the simplest member of this family [13]. All coumarins are benzo-alpha-pyrone derivatives [6] and these compounds have phytotoxic, antifungal, antibacterial, insecticidal and anthelmintic effects [26]. One of the most important coumarins is amblyferon (Figure 6), which is used in enzyme assays [38].



Fig. 5 Chemical structure of Amblyferon



Fig. 6 Chemical structure of coumarin

Alkaloids

Alkaloids are compounds that have at least one nitrogen atom in the heterocyclic ring. Alkaloids are the best known natural compounds containing nitrogen. These compounds have a bitter taste and have a toxic effect on the cell membrane system, especially nerve cells. They act as an insecticide and are very important in the pharmaceutical industry. This class of secondary metabolites have a mainly defensive role and are sometimes proposed as sources for nitrogen storage in the plant [36]. Among the most important alkaloids, we can mention morphine (Figure 7), codeine (Figure 8) and nicotine (Figure 9).



Fig. 9 Chemical structure of nicotine

Highly addictive compounds such as cocaine, morphine, nicotine, caffeine and THC or Tetra hydro cannabinol (marijuana) are in this group. Indole alkaloids are secondary metabolites with a wide range that have a wide variety of medicinal activities. Indole alkaloids and their derivatives have been widely used in clinical trials, including vinblastine, vincristine, camptothecin, vindoline, reserpine and indoline. Extensive studies have shown the diverse biological activity of indole alkaloids from antibacterial activity to anti-inflammatory activity and antitumor activity. In addition, indole alkaloids from the marine environment are a promising and active group of biomolecules that cover biological, cytotoxic, antiviral, antiparasitic and anti-inflammatory activities [35].

THE CONTROL OF PLANT PATHOGENS BY PLANT EXTRACTS AND ESSENTIOL OILS

The effects of plant extracts on the control of plant pathogens

The growth, development and performance of plants are affected by harmful soil pathogens such as Fusarium solani. But the use of chemical fungicides to control of this pathogen cannot be a suitable method, because the use of pesticides threatens human health and causes environmental pollution. While the control of plant pathogens by medicinal plant extracts including ginger rhizome extract (GRE) can be effective and useful. In the surveys conducted about, ginger rhizome extract (GRE) to control F. solani showed that the mycelium structure of F. solani was destroyed by the extract of this plant and the growth of mentioned soil pathogen was completely inhibited. It also stopped cellular respiration, the activity of enzymes that destroy the cell wall and reduced the amount of fusaric acid and thus reduced the infection severity caused by F. solani. The analysis of ginger extract by UPLC-MS/MS device showed that phenolic acids had the highest and tannins had the lowest amount. The mechanism of action of controlling this pathogen was carried out by the main phenolic and flavonoid compounds such as 4-hydroxybenzaldehyde and quercetin present in ginger extract, and the result of this experiment introduced these two compounds as natural fungicides in controlling of F. solani. [39]. In a similar study [1] on the antifungal effect of plant extracts of Mentha piperita L. on plant pathogenic fungi including Rhizoctonia solani, Fusarium oxysporum, Phytophthora drechsleri and Bipolaris sorokiniana by two methods, paper disc and mixing with culture medium, showed that water is the best solvent for extracting inhibitory substances from Mentha piperita L. and extracts extracted with methanol and acetone had little effect on the mycelium growth of Bipolaris sorokiniana, and ethanolic and chloroform extracts had no Fungistatic effect. In examining the concentration gradient of the extract, F. oxysporum and B. sorokiniana were affected at a concentration of two milligrams per paper disk, while the mycelium growth of two other fungi was affected at a concentration of four milligrams per paper disk. In the method of mixing with culture medium, the aqueous extract at a concentration of 500 ppm, the growth of P. drechsleri and at a concentration of 1000 ppm the growth of B. sorokiniana completely prevented. However, in the case of the other two fungi, even at a concentration of 2000 ppm, the growth did not stop completely. In the another study for controlling of *F.oxysporum* f. sp. lycopersici race 3, by the extract of medicinal plants; Adhatoda vasica, Eucalyptus globulus, Lantana camara, Nerium oleander and Ocimum basilicum, in laboratory conditions using water and specific organic solvents, revealed that the growth of F. oxysporum stopped by use of cold distilled water extracts of O. basilicum and E. globulus with highly inhibiting effective. The control of the pathogen growth by butanol and ethanol extracts of the five mentioned plants better than aqueous extracts. The concentrations of 1.5 and 0.2% (v/v) of butanol extract of O. basilicum inhibited completely the growth of F. oxysporum f. sp. lycopersici race 3. Hydrolytic enzymes such as β glucosidase, pectin lyase and protease inhibited by butanol extract (0.2%) of the mentioned plants. Thus the results of this investigation was showed that the ecological balance does not disturb with using plant extracts of O. basilicum, to control F. oxysporum f. sp. Lycopersici and is safe environmentally and is economical, as well [14]. Other investigations [28] on the antifungal properties of different concentrations of plant extracts showed that the plants Nepeta cataria, Nicotiana tabacum, Thymus yulgaris, Foeniculum vulgare and Hyssopus officinalis have a good inhibitory effect on Rhizoctonia solani. The highest inhibitory effect was related to the extracted extract with methanol solvent. The minimum inhibitory concentration of the methanolic extract of Nepeta cataria, Nicotiana tabacum, Thymus vulgaris, Foeniculum vulgare and Hyssopus officinalis, on the examined pathogenic fungus was 1.5, 1.5, 2, 3, 3 and 2.5 mg/ milliliters. Therefore, it seems that the natural compounds of these plants, especially Nepeta cataria, Nicotiana tabacum and Thymus vulgaris, can be used to control the disease caused by this fungus. Of course, the results of another research showed that in the case of standard Aspergillus flavus (PTCC 5006), nystatin and aqueous extracts of Anethum graveolens, T. vulgaris and Coriandrum sativum had equal amounts and finally Rosa damascena had the most antifungal effects. Regarding the isolated strain of this fungus from the environment, nystatin, aqueous extracts of T. vulgaris, C. sativum and then aqueous extracts of A. graveolens and finally R. damascena had the most antifungal effects. In the case of standard Aspergillus fumigatus (PTCC 5009), the most effective antifungal compounds investigated included nystatin, aqueous extracts of Anethum graveolens, Thymus vulgaris, Rosa damascena, and C.sativum, respectively. Regarding the isolated strain of this mushroom, the most effective antifungal compounds included nystatin, aqueous extracts of A. graveolens, T. vulgaris, C. sativum and finally Rosa damascena. The results of this research showed that in all cases the extracts reduced the growth of the colonies of fungi, and this effect increased with the increase in the concentration of the aqueous extracts of A. graveolens, T. vulgaris, C. sativum and R. damascena. By proving the effectiveness of the aqueous extracts of A. graveolens, T. vulgaris, C.sativum and R. damascena flowers on the growth of two species of Aspergillus, including A. flavus and A. fumigatus, it can be hoped that by purifying the effective substance of the mentioned plants and conducting further research, the industrial production of a combination with acceptable antifungal effects and low side effects for the treatment of fungal infections will be achieved [40]. In a laboratory study [9], the antifungal effects of two plant extracts, Ferulago angulata and Zataria multiflora, were studied on F. oxysporum. Concentrations of 0.4, 0.8 and 1.2% of the mentioned extracts were prepared in PDA culture medium and zero value was considered as a control. The effect of the aqueous extract of dried aerial parts separately in preventing the growth of fungal mycelium in PDA

food medium inside the petri dish was investigated. For this purpose, tablets with a diameter of 6 mm from the 7-day-old mold of *F. oxysporum* were placed in the center of the pan containing PDA culture medium along with plant extract. The results showed that the used extracts have an inhibitory effect on the growth of this mushroom, and the effect of *Z. multiflora* leaf extract with a concentration of 2.1% and *F. angulata* flower extract with a concentration of 0.8% was higher than other concentrations. Therefore, the extracts of medicinal plants can be considered as antifungal compounds and biological control agents. Of course, the secondary metabolites in the extracts of medicinal plants are responsible for this antifungal actions.

The role of essential oils in controlling plant pathogens

One of the important compounds of medicinal plants are essential oils, which have many biological effects, and the use of these natural compounds in the control of pests and plant diseases is one of the ways to reduce environmental risks. In an experiment, the inhibitory effect of different concentrations of the essential oil of the medicinal plant Mentha pulegium L. belonging to the mint family (one of the important, widely used and economic medicinal plants of Iran), against the growth of plant pathogenic fungi F. equiseti, F. cerealis, F. graminearum, F.oxysporum f.sp. lentis, F. proliferatum, A. niger, Botrytis cinerea and Sclerotinia sclerotiorum were investigated. The results of this research showed that the essential oil of Mentha pulegium L. has a significant effect in inhibiting the growth of pathogenic fungi, and with increasing concentration, the inhibitory effect increased significantly, so that the most inhibitory effect against fungal growth were observed in concentrations of 2.5, 2, 1.5, 1, 0.8 and 0.6 ml of essential oil per liter of culture medium [31]. Also, in order to inhibit the pathogenic agents of edible mushroom, including the bacteria *Pseudomonas tolaasii* and the fungus Trichoderma harzianum, the essential oil of Cuminum cyminum, Z. multiflora and two ethanolic extracts of Dorema ammoniacum and F. angulata were compared to a chimeric recombinant peptide (Lactoferricin-Lactoferrampin) which is one of the most important antimicrobial components of camel milk. The results of antibacterial evaluations showed that the chimeric peptide, at a concentration of 20 µg/ml, had the highest performance with an average inhibitory halo diameter of 2.32 mm among the examined compounds, followed by *Cuminum cyminum* essential oil and *Dorema ammoniacum* extract, respectively, with seven and Six millimeters had the most antibacterial effect. The results of the antifungal effects of the compounds tested in a period of seven days showed that the essential oils of C. cyminum and Z. multiflora plants completely prevented the growth of T. harzianum, and then the extracts of D.ammoniacum and F. angulata plants with an average colony growth of 26 and 60.66 mm had the most antifungal effect. The lowest antifungal effect was obtained in the case of the chimeric recombinant peptide. According to the results of this research, the chimeric peptide had more antibacterial effects compared to essential oils and plant extracts, and essential oils and extracts had more antifungal ability than the tested peptide [33]. The use of essential oils of medicinal plants in the control of plant diseases has been considered by many researchers around the world. F.oxysporum species causes many diseases in different plants. In order to investigate the combined and separate effect of *Foeniculum vulgare* and *Salvia rosmarinus* medicinal plant essential oil on the inhibition of mentioned fungus, an experiment was conducted in the form of a completely randomized design with five replications. In this experiment, the method of mixing essential oil with culture medium was used. The essential oil of S. rosmarinus was more inhibitory than the essential oil of F. vulgare. In addition, the combined use of essential oils caused more inhibition of the growth of the fungus than their separate use and was able to stop its growth completely. The inhibition percentage of F. vulgare and S. rosmarinus essential oil at the highest concentration was 2.49% and 7.68%, respectively. By combining the essential oils of these two plants, the inhibition percentage reached 100% at the highest concentration. Combining the essence of several plants with each other increases its inhibitory effect. Therefore, by using low concentrations of essential oil, the percentage of inhibition of the pathogenic agent can be increased [30]. Therefore, according to the results of this research, the use of oil essential medicinal plants to inhibit the growth of plant pathogenic agents is recommended.

CONCLUSION

Plant pathogens such as fungi, bacteria, parasitic nematodes, etc. annually cause severe damage to agricultural products in Iran. Usually, chemical pesticides are widely used to control pests and plant diseases throughout the country, which cause environmental pollution, incurring large costs Emergence of resistance to all kinds of

synthetic pesticides, especially antibiotics, poisoning caused by the consumption of chemical poisons to animals, aquatic animals, and beneficial insects, as well as the adverse effects of toxic residues, have caused many problems for human health and the environment. One of the new methods in combating plant diseases is the use of extracts and essential oils of medicinal plants. Considering the increasing damage caused by plant diseases and due to the side effects of using pesticides use of natural materials and compounds of plant origin for control of plant pathogenic agents as an alternative to dangerous pesticides, is of particular importance. The role and importance of secondary compounds and essential oils in the control of various types of plant diseases, including fungal, bacterial, viral and nematode diseases, is very clear because they are an effective and sustainable control methods for a number of soil-borne and seed-borne pathogens. Many countries have started the integrated control of important plant diseases by using new technology to prepare and formulate non-chemical poisons, including plant-based pesticides. Out of a total of 1,200 species of medicinal plants in Iran, 475 species are in Kohgiluyeh and Boyerahmad province, while the number of species of medicinal plants in Europe is 800, which means that this province can have a good potential for the use of medicinal plants in various fields, including the control of plant pathogens. Considering the abundance and diversity of medicinal plants in Iran (especially in Kohgiluyeh and Boyerahmad province), further studies on the effects of the active ingredients of native medicinal plants in controlling of plant pathogenic agents in order to obtain plant-based pesticides, is recommended.

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