

Comparison of Allelopathic Effect of *Zataria Multiflora* on the Germination and Growth Features of *Cymbopogen Olivieri* and *Stipa Arabica* Seedlings

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Abstract: Application of native and genetically modified species is one of the main approaches to revival and modification of ranches. But in the mean time, it should be noted that those species that are compatible to each other should be used in vegetation expansion projects. Shiraz oregano (*Zataria Multiflora*) is from among those plants which could cause allelopathic effects due to their various chemical compositions. Hence, due to profusion of this plant throughout the Khalil Beig ranch of Arsanjan, and the considerable amount of *Stipa Arabica* and *Cymbopogen Olivieri* in the adjacent areas to this ranch that are consumed by the livestock, it was decided to study the possibility of applying the aforesaid species for expansion of vegetation throughout the Khalil Beig ranch. To this end, an investigation was conducted in the greenhouse environment using the soil taken from the habitat of Shiraz oregano. Underground and aerial parts of this plant were collected and extracts of 25, 50, 75 and 100 percent as well as 50 and 100 % densities were obtained from aerial and underground parts, respectively. Also, a bittern was considered as the prototype (distilled water). Seeds of *S.arabica* and *C.olivieri* were cultivated in flower pots containing the soils of oregano habitat and were irrigated using the abovementioned bitters throughout the entire study. The investigations lasted for 5 weeks and the germination and growth rates of seedlings were recorded on a daily basis. In the end, characteristics of both species such as percentage of germination, length of stem, length of root, wet weight of stem, wet weight of root, dry weight of stem and dry weight of root affected by different density percentages of Shiraz oregano extract were analyzed through variance analysis. The Duncan test was applied for comparison of means. The results represented the preventive effect of compositions existing in Shiraz oregano on the studied features and the less vulnerability of *S.arabica* as compared with the *C.olivieri*.

Key words:

INTRODUCTION

Allelopathy is part of the science of chemical ecology and mostly discusses the preventive effects of allelochemicals existing in various plants and plant textures. Turk and Tawaha (2003) defined allelochemicals as secondary or bonus products of the main metabolite of a plant.

According to the International Association of Allelopathy, any process during which the secondary metabolites are generated by a plant as a result of which the growth of biological systems are affected, are considered allelopathic whether their effects are positive or negative. Muller *et al* (1968) attributed the cessation in growth of *Festuca Meghira* and *Erodun Cicarnum* to the presence of allelopathic chemicals in *Artemisia*. Mc Feron and Muller (1972) concluded that the chemicals washed off the mature leaves of *Adenoston Fasciculatum* due to precipitation prevent the germination of most seeds during their growth seasons. The resulting hay would decrease in the same soil. As findings suggest, hay generates allelopathic chemicals that are detrimental to hay. Friedman *et al* (1997) attributed the scarcity of sensitive one-year-old species to the adjacency of wormseeds and their allelopathic effects. Jefferson and Pennachio (2003) studied and approved the allelopathic effect of *Kenopodiase* plants based on the allelopathic impacts of extracts from the four species of *Chenopodiaceae* species on germination of *Atriplex bunburana*, *Enchylana tomentosa* and *Mareriana georgei* seeds.

Djurdjevic *et al.* (2004) assessed the preventive potential of *Alliu urnisim*. Kpoveissi *et al.* (2006) have indicated the preventive feature of *JUSTICIA ansellima* extract on different growth stages of *Vigna Unglicluta*. Hante (2003) concluded that *Atriplex Canescens* has preventive effect on germination of *Artemisia siebieri*. Although cultivation of bushes has commenced in arid and semi-arid ranches of Iran and continued ever –since, its cultivation has been deemed negative in wormwoods which could be attributed to its allelopathic effects. The

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results of field studies carried out by Samedayi and Baghertani (2005) on the allelopathic effects of a number of wormseed species on germination of seed and growth of *Avena indoviatica* seedling also suggests that the lengths of radicle, plumule and seedling would exponentially decrease as the concentration of the wormseed extract increases. Rezaee and Khajeddin realized the preventive effect of *Asgress* specie on germination of *Scariola orientalis* and *Agropyron elongatum*.

The Shiraz oregano is widespread throughout the Khalil Beig ranch but considering the dominance of this specie, the aforesaid ranch is not frequently used for livestock grazing. Taking into account the pharmaceutical features of this plant and its chemical compositions, it could be considered to have allelopathic potential.

On the other hand, there are widespread species of *C. olivieri* and *S.arabica* in ranches adjacent to habitat of Shiraz oregano which are of great significance in terms of grazing. Thus, to study the possibility of cultivating such species, it was necessary to evaluate the potential preventive effects of Shiraz oregano on germination and growth of the aforesaid species. Also, evaluation and comparison of both species was deemed necessary, so that the least affected seed could be proposed for cultivation within this region.

MATERIALS AND METHODS

In order to evaluate the preventive effect of *Z.multiflora* on burgeoning and growth features of *S.arabica* and *C. olivieri*, the aerial parts of Shiraz oregano including its fruit, stem, leaf and flower as well as the underground parts including its root were collected.

As the parts were dried out in open air, they were pulverized with grinders. Extractions of parts through distilled water were carried out as follows:

25g powder of aerial parts and 25g powder of root were separately mixed with water with a 1:3 proportion. The mix was then stirred for one hour with a shaker and then kept in the fridge for 24 hours at 4 °C. Then, they were again stirred for 2 hours and then centrifuge process was applied for 5 minutes at 2500 round per minute rate.

The extract was sieved through Wattman filters. Then, increasing the volume of extract, the required bitters were developed as follows: prototype bitter, aquatic extract bitter with 25% aerial part density, aquatic extract bitter with 50% aerial part density, aquatic extract bitter with 50% underground part density, aquatic extract bitter with 75% aerial part density, aquatic extract bitter with 100% aerial part density and aquatic extract bitter with 100% underground part density. Considering the test being conducted in greenhouse conditions, the soil from natural habitat of oregano was used in the flower pots of the seeds under study. The soil of Shiraz oregano habitat was sampled from depth of 0-2 cm with no disturbance using steel pots 20 cm in height.

In order to avoid mistakes regarding the burgeoned seeds in the pots that could belong to other species present in the soil, the pots were irrigated for one month prior to undergoing cultivation. Then, a number of 25 seeds of *S.arabica* and *C.olivieri* species approved in terms of quality were cultivated in each pot.

The number of recurrence for each bitter was 6 pots so that 84 flowerpots were applied in total. Having cultivated the seeds, pots of various extract densities were irrigated throughout the study.

The test lasted for 5 weeks and the number of germinated seedling were recorded on a daily basis. In the end, the seedlings were removed from the pots and the lengths of their respective stems and roots were measured.

Also, the wet and dry weights of stems and roots were measured by a scalar of 0.0001 g accuracy. As the data were collected, the data analysis was carried out using both the variance analysis and SPSS software. The diagrams were plotted in Excel sheets.

Results:

- **Burgeoning Percentage:**

As table 1 shows, the variance analysis of burgeoning percentage reflects the impact of density bitters. Figure 1 shows the average burgeoning percentage of *C.olivieri* specie in different densities of extract. As it is obvious, the highest percentage of burgeoning belongs to the prototype bitter and the least is relevant to the 50% extract of aerial parts, though there are no significant differences between 50% and 75% extracts of aerial parts. Figure 2 shows the effect of various densities of Shiraz oregano extract on germination of *S.arabica*. The variation trend of germination is the same as the former specie.

Table1: ANOVA result of effects of plant, density and interactions of Percent of Germination.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	2.33	2.33	0.08	ns
Error (A)	10	297.62	29.76		
Density (B)	6	22858.95	3809.83	74.24	**
Plant*Density	6	662.00	110.33	2.15	ns
Error (B)	60	3079.05	51.32		
Total	83	26899.95	324.10		
CV (%)	24.44				

** . Significant of 1% (P<0.01) ns: non.Significant (P>0.05)

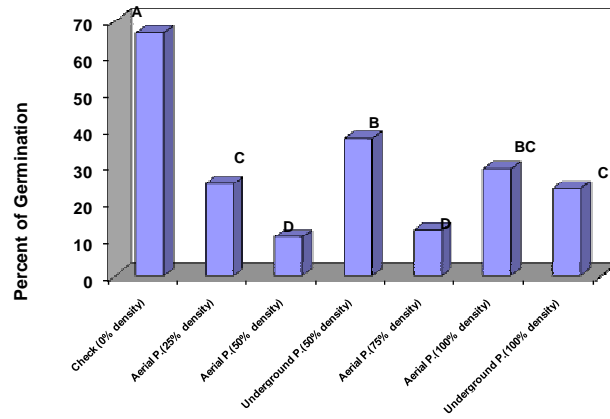


Fig. 1: Average germination percentage of *C. olivieri* in different concentrations of *Z. multiflora* extract.

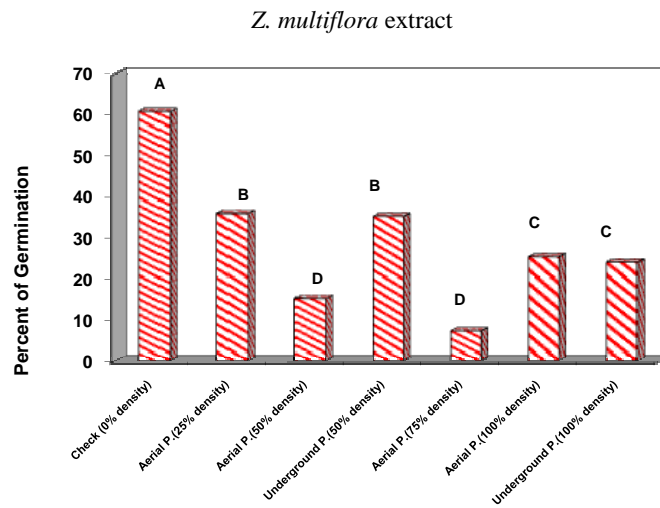


Fig. 2: Average germination percentage of *S. arabica* in different concentrations of *Z. multiflora* extract.

Length of Stem:

Table 2 shows a significant difference at 1% level between the length of stem among species, density of extracts and their mutual effects. Figure 3 shows that the length of stem in *S. arabica* is much longer than that of the *C. olivieri* when affected by various densities of extracts. Densities of *C. olivieri* and *S. arabica* respectively fall into two and three groups in terms of length of stems. The stem length of 25% densities, 50% underground parts and the prototype are placed in one group in case of *S. arabica* while stem length of 50% bitterns of aerial parts and 100% aerial and underground parts are placed in another group and the stem length of 75% densities falls into a separate group with the least length of stem.

Table 2: ANOVA result of effects of plant, density and interactions of Length of Stem.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	64468.98	64468.98	394.12	**
Error (A)	10	1635.77	163.58		
Density (B)	6	13570.47	2261.75	22.19	**
Plant*Density	6	8049.22	1341.54	13.16	**
Error (B)	60	6114.30	101.91		
Total	83	93838.75	1130.59		
CV (%)	24.06				

** Significant of 1% (P<0.01).

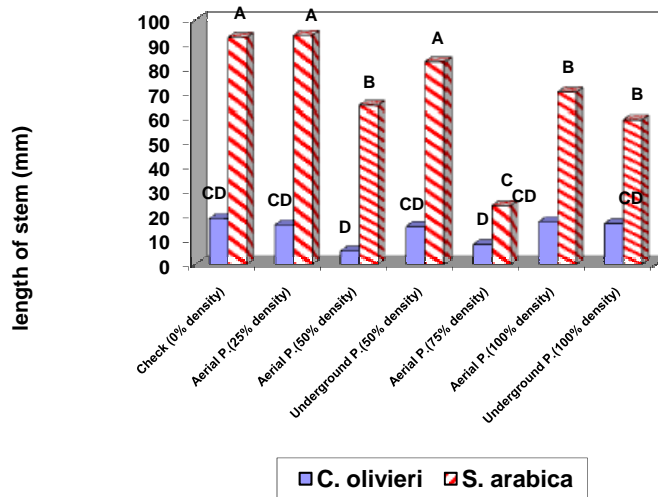


Fig. 3: Average stem length (mm) in interaction effects of species and concentrations of *Z. multiflora* extract.

Length of Root:

Results derived from variance analysis of root length represent a significant difference between species, extracts and their mutual effects at 1% level of density. Figure 4 depicts the longer root length of *S.arabica* compared to that of the *C.olivieri*. The largest length belongs to the 7th bittern and prototype.

There is no significant difference between the various densities affecting *C.olivieri* in terms of root length and they all fall into one group while it is not the same in case of *S.arabica* with densities falling into four distinct groups. The largest length belongs to 100% bittern of underground parts while the smallest belongs to 75% bittern.

Table 3: ANOVA result of effects of plant, density and interactions of Length of Root.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	3986.83	3986.83	255.63	**
Error (A)	10	155.96	15.60		
Density (B)	6	957.48	159.58	5.05	**
Plant*Density	6	605.43	100.90	3.19	**
Error (B)	60	1895.44	31.59		
Total	83	7601.13	91.58		
CV (%)	48.87				

** Significant of 1% (P<0.01).

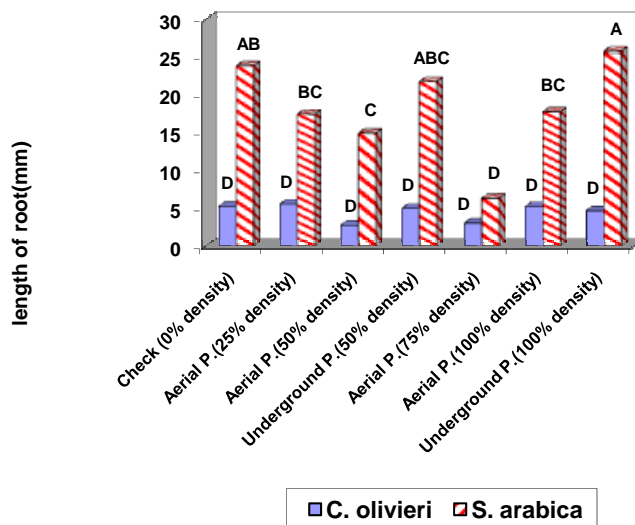


Fig. 4: Average root length (mm) in interaction effects of species and concentrations of *Z. multiflora* extract.

Wet Weight of Stem:

The variance analysis of this property shows the distinction of species, extract densities and their mutual effect on the wet weight of stem at 1% level.

As figure 5 shows, the S.arabica has higher wet weight than the C.olivieri specie. The latter species shows no distinction between the bitters in terms of wet weight of aerial parts as was the case for stem length, but the S.arabica shows significant differences between densities so that the prototype bitter and 25% density and 75% bitter have the highest and lowest wet weights of aerial parts, respectively.

Table 4: ANOVA result of effects of plant, density and interactions of Wet Weight of Stem.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	0.052851	0.052851	128.28	**
Error (A)	10	0.004120	0.000412		
Density (B)	6	0.023846	0.003974	14.33	**
Plant*Density	6	0.018293	0.003049	10.99	**
Error (B)	60	0.016646	0.000277		
Total	83	0.115756			
CV (%)	30.21				

** . Significant of 1% (P<0.01).

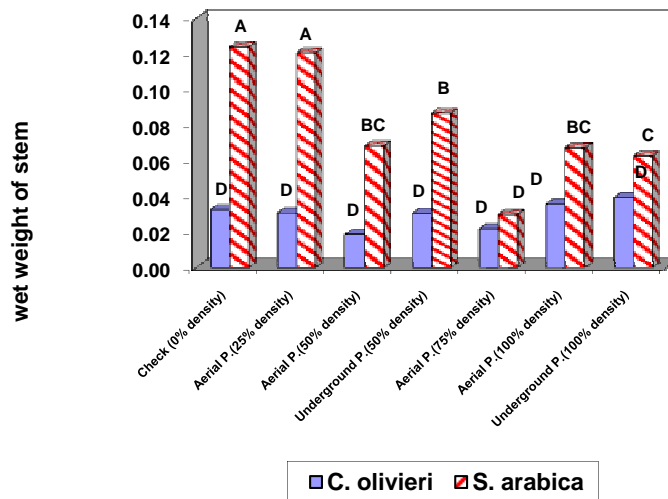


Fig. 5: Average stem fresh weight (gr) in interaction effects of species and concentrations of *Z. multiflora* extract.

Wet Weight of Root:

The variance analysis of this property shows the distinction of species, extract densities and their mutual effect on the wet weight of stem at 1% level.

Figure 6 shows that the S.arabica has higher wet weight of root as was the case for other properties. Also, the highest and lowest wet weights of root belong to 75% extract bitters and distilled water, respectively.

Table 5: ANOVA result of effects of plant, density and interactions of Wet Weight of Root.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	0.00406	0.00406	36.21	**
Error (A)	10	0.00112	0.00011		
Density (B)	6	0.00853	0.00142	12.18	**
Plant*Density	6	0.00220	0.00037	3.14	**
Error (B)	60	0.00700	0.00012		
Total	83	0.02291	0.00028		
CV (%)	85.50				

** . Significant of 1% (P<0.01).

Dry Weight of Stem:

Results of variance analysis of dry weight of stems are presented in table 6. Effects of specie type, densities of Shiraz oregano extract and their mutual effects have significant difference at 1% level.

Figure 7 shows the variation trend of dry weight of stem between the two species and various densities of Shiraz oregano extract. Variation trend of dry weight of stem is nearly similar to that of the wet weight of stem.

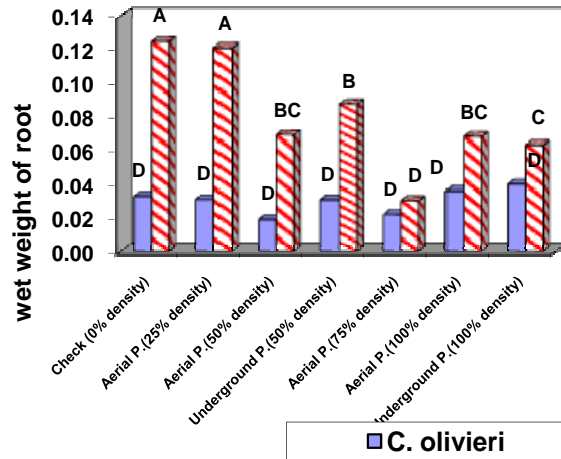


Fig. 6: Average root fresh weight (gr) in interaction effects of species and concentrations of *Z. multiflora* extract.

Table 6: ANOVA result of effects of plant, density and interactions of Dry Weight of Stem.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	0.000114	0.000114	42.27	**
Error (A)	10	0.000027	0.000003		
Density (B)	6	0.000256	0.000043	26.21	**
Plant*Density	6	0.000051	0.000008	5.21	**
Error (B)	60	0.000098	0.000002		
Total	83	0.000546	0.000007		
CV (%)	16.28				

** Significant of 1% (P<0.01).

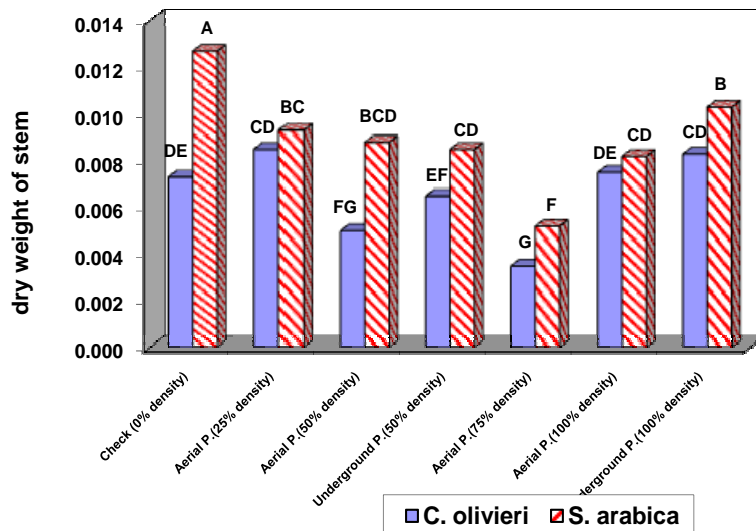


Fig. 7: Average stem dry weight (gr) in interaction effects of species and concentrations of *Z. multiflora* extract.

Dry Weight of Root:

Table 7 shows the effect due to type of specie, density bitern and their mutual effect on dry weight of root at 1% level. S.arabica once again shows higher values as was the case with other properties of the two species.

Table7: ANOVA result of effects of plant, density and interactions of Dry Weight of Root.

Source	df	SS	MS	Fvalue	Sig.
Plant. (A)	1	0.000792	0.000792	263.31	**
Error (A)	10	0.000030	0.000003		
Density (B)	6	0.000305	0.000051	24.27	**
Plant*Density	6	0.000115	0.000019	9.16	**
Error (B)	60	0.000126	0.000002		
Total	83	0.001368	0.000016		
CV (%)	21.62				

** Significant of 1% (P<0.01).

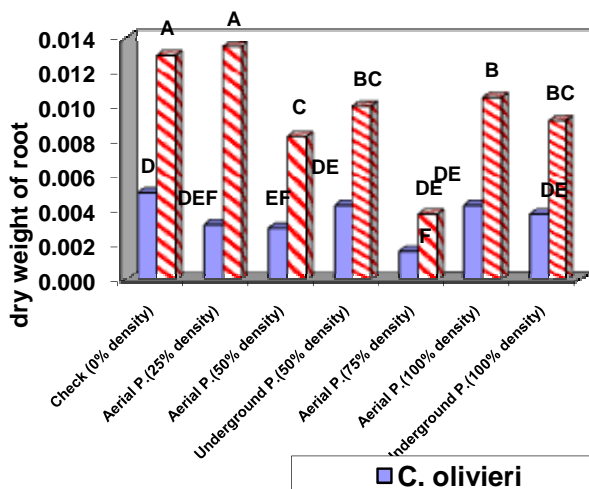


Fig. 8: Average root dry weight (gr) in interaction effects of species and concentrations of *Z. multiflora* extract.

Discussions and Conclusions:

The obtained results showed that the chemicals present in the extract of aerial parts of Shiraz oregano have preventive effects on properties of *C.olivieri* and *S.arabica* while the underground parts showed less impact especially at 50% density which correlated with findings of Turk *et al* (2003).They realized that the aquatic extract of *Brassica nigra* have significant effect on prevention of germination and growth of *Avena afatua* seedling. The descending rate of effect was as follows:

leaves>flowers>stem>root.

Sharma *et al.* (2000) also elicited that the effectiveness of extract derived from aerial parts has stronger preventive effects.

Also, the results of Mohsenzadeh *et al* (1997) studies suggest that the extract derived from leaves and flowers of wormseed has considerable impact on germination of *Agropyron* seedling, while the extract obtained from the root of wormseed had no significant effect. This might be closely related to their accumulation in the upper parts of plant. The results of current study and those mentioned above differ from that of Moeni's in which he studied the preventive effect of wormseed on nitrification.

His results showed the preventive effect of the aforesaid parts on nitrification as follows:

root>stem>leaf>flower

This contradiction could be due to the difference in the ultimate goal which are nitrification in case of the latter and germination and growth of seedlings in case of the former.

Although it was expected that the increase in density of extracts obtained from aerial and underground parts of Shiraz oregano would add up to the characteristics of *C.olivieri* and *S.arabica*, a yield point in the effect curve and a decrease in prevention rate could be observed to the extent that there is always a significant difference between the bitters of 75 and 100 percent extracts where 100% bitters yields better results.

Thus, the 75% density may be considered the upper bound of Shiraz oregano's impact on the other species after which a more moderate dip of curve is observed. In other words, densities of seed and sapling would become physiologically compatible. This could be justified according to Reiss's (1984) definition allelopathy. He described allelopathy as the either direct or indirect detrimental/useful effects of plants on each other due to generation of chemical substances in the environment. So the effects could either be negative or positive. It can

be declared that the increase in density of allelochemicals may lead to positive impacts due to their nature and THE chemical structure as compared to lesser densities.

Comparing the sensitivity of *C.olivieri* and *S.arabica*, the latter shows better conditions with less vulnerability compared to the former. So, the application of *S.arabica* is of higher priority than *C.olivieri* and cultivation of the latter is not recommended in this region.

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