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ALLELOPATHIC POTENTIAL OF COTTONWOOD (*POPULUS DELTOIDES* BARTR. EX MARSH.) LEAF EXTRACTS ON SOME RICE (*ORYZA SATIVA* L.) CULTIVARS

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Abstract

An experiment was conducted to declare the allelopathic potential of *Populus deltoides* leaf extracts on four local *Oryza sativa* L. cultivars. The Laboratory study consisted of seed germination, radicle, and plumule growth phases. The experiment was performed under five leaves extract concentrations (0%, 25%, 50%, 75%, and 100%). The results revealed that seed germination indices were significantly reduced and inhibited by increasing leaf extract concentration. The development of the plumule and radicle showed a significant reduction in concentration changes. The current study demonstrated that there was a strong allelopathic effect of cottonwood on rice cultivars. It is important to consider that the *P. deltoides* was not recommended as a suitable tree for agroforestry with rice until further research has been conducted on field experiments under the shelter of cottonwood, to identify the allelochemicals in other parts of the cottonwood tree and the soil litter content in the stands.

Introduction

Allelopathy is defined as the natural inhibitory interaction between plants and the environment by producing and releasing some specific chemical compounds into the surroundings. The allelopathic compounds are produced through secondary metabolism in plants that can influence other plant species in agricultural systems, natural systems, or both (agroforestry systems). This phenomenon is a biological force that reduces plant performance by harmful effects such as resource competition, affecting the

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germination and growth disorder of neighboring plant species through chemical interference. Allelopathic compounds that are called allelochemicals can generate different inhibition on the growth of the other plants and induce chemical interactions between organisms or plant species to affect their development, health, behavior, or even population biology (WANG et al. 2020). Therefore, a quantitative analysis is needed to understand the roles of allelopathy as a biological phenomenon in agroforestry systems, study the allelopathic interactions of trees species on agricultural plants, help the past decades' knowledge gaps and guide future research (ZHANG et al. 2021, BANUELAS et al. 2019).

Agroforestry systems involve the combination of cultivated woody tree species with annual crops in the same spatial and temporal arrangements. This combination leads to economic profits, a sustainable environmental land-use system, and multi-layer cultivation. Nevertheless, interactions are inevitable. One of them is allelopathic interactions between the release of allelochemicals from dead and falling leaves of trees and inhibition of seed germination and growth. Allelochemicals could be found in the various parts of a plant, released from the litter, and affect the development of neighbor crop plants in agroforestry systems. Therefore, It must be determined that the allelopathic compatibility of crops with trees before incorporating them into an agroforestry system because released allelochemicals by some trees could affect the establishment and development of crops and reduce economic and environmental efficiency. Otherwise, allelopathic interference effects will recognize as the primary reasons for low productivity in agroforestry systems (JOHN et al. 2006, AMOO et al. 2008).

Tree species (especially multi-purpose ones) as an integral part of agroforestry programs could conserve the soil and increase agricultural soil productivity and quality by helping to add the organic matter into it, improving the water holding capacity of it, biodiversity of microbes, nutrients concentrations, declining pests population and conservation of the soil. However, several tree species have negative effects on the performance of crops through mono-culture plantation and allelopathy. The roots, leaves, and bark of these species release allelochemicals into the soil and caused negative interaction depending on the concentration of allelochemicals. These interactions inhibited the growth of neighbor plants (AMOO et al. 2008, GARIMA and DEVI 2017).

Furthermore, There have been many studies that reported about the suppressive effects of the tree species' leaves extracts on the germination of other agricultural species, such as allelopathic effects of *Senna siamea*, *Albizia lebbeck*, *Azadirachta indica*, *Cedrela odorata*, *Leucaena leucocephala*, *Gliricidia sepium*, *Eucalyptus grandis*, *Terminalia superba* and *Tec-*

tona grandis trees species on seeds of *Zea mays* (Maize), *Vigna unguiculata* (Cowpea), *Lycopersicon esculentum* (Tomatoes), and *Hibiscus esculentus* (Okra) (ABUGRE et al. 2011). *Azardiracta indica*, *Vitellaria paradoxa*, and *Parkia biglobosa* trees on germination and growth of *Vigna unguiculata* (cowpea) (ALEEM et al. 2014). *Pinus halepensis* and *Quercus coccifera* as two agroforestry trees on the germination of *Triticum aestivum* (wheat), *Hordeum vulgare* (barley), *Lens culinaris* (lentil), *Cicer arietinum* (chickpea), and *Vicia faba* (faba bean) (ALRABABAH et al. 2009). Also Allelopathic potential of *Tetrapleura tetraptera* leaf extracts on *Lycopersicon esculentum*, *Abelmoschus esculentum*, *Amaranthus spinosus*, *Capsicum annum*, and *Solanum melongena* (AMOO et al. 2008). The aqueous leaf extracts of *Pinus sylvestris*, *Broussonetia papyrifera*, and *Pinus tabulaeformis* could promote *Amygdalus pedunculata* seed germination and seedling growth (WANG et al. 2021).

On the other hand, there are represents that showed the allelopathy in ecosystem-level of forests (BLANCO 2007). Many kinds of the literature indicated the trees' foliage leachates had the allelopathic potential on the plants of understory that leaf extracts could influence seedling growth in the natural condition (TEIXEIRA DA SILVA et al. 2015).

The allelopathic capacity of forest trees has been reported previously such as allelopathic effects of *Albizia julibrissin* (ABEDI and ABDI 2021), *Melia azedarach* (MAJEED et al. 2017), *Zanthoxylum armatum*, *Ougeinia oojeinensis* and *Boehmeria rugulosa* (VASISHTH et al. 2020).

Since agroforestry is a leading alternative for food security and forest conservation, it seems that identifying the local trees and crops with the minimum allelopathic interaction is necessary for the best agroforestry system management. Scientific studies could be used to identify the best crops cultivars and the best accompanying tree species to improve the agroforestry system's productivity and move from monoculture to multicultural plantation. A successful agroforestry system management depends on identifying local tree crops with a minimum accumulation of toxins in the soil (THAPALIYAL et al. 2008, ALRABABA et al. 2009).

In recent years, the studies focused on the chemical aspects of agricultural plants and forest trees plantation relationship (LU et al. 2017). Evaluation of allelopathic effects of leaf and bark extracts of *Dalbergia sissoo* on wheat germination under the different concentrations by SIYAR et al. (2018) observed that the mean germination time significantly increased. The final results suggested the negative effects of allelopathy of this tree species on wheat. In addition, GARIMA et al. (2017) introduced the *P. deltoids* as a multipurpose tree species but had unfavorable effects on crops through allelopathy. They explored the allelopathy effects of *P. del-*

toides leaf extracts on wheat (*Triticum Aestivum*) germination and radicle and plumule length in laboratory conditions and determined that increase in the concentration of leaf extract had more inhibition effects. LU et al. (2017) introduced the poplar as an allelopathic species and reported the impact of monoculture plantation of poplar on the rhizosphere microbial community. SHER et al. (2011) noted the allelopathic potential of *Populus euphratica* against *Sorghum vulgare*, *Setaria italica,* and *Triticum aestivum*. The poplar leaves have toxic and reduce the germination, plumule, and radical growth, fresh and dry weight of all studied crops in laboratory experiments. AMOO et al. (2008) studied the allelopathic potentials of *Tetrapleura tetraptera* leaves extract as a multi-purpose tree species on *Lycopersicon esculentum*, *Abelmoschus esculentum*, *Amaranthus spinosus*, *Capsicum annum,* and *Solanum melongena* as five agriculture crops and reported the significant preventive effects on crops seedling growth. SINGH et al. (2001) studied the allelopathic interface of *P. deltoides* in seven winter seasons crops including *Triticum aestivum* (wheat), *Lens culinaris* (lentil), *Phaseolus mungo* (black gram), *Avena sativa* (oat), *Trifolium alexandrinum* (clover), *Brassica juncea* (Indian mustard) and *Helianthus annuus* (sunflower). Their conclusion showed a significant reduction (10–30%) in germination, height, and biomass of crops. SHARMA et al. (2000) investigated the allelopathic effects of *Populus deltoides* on wheat in the laboratory condition and showed that germination and growth of wheat were suppressed by extraction added to the soil.

In the north of Iran, *P. deltoides* is an important fast-growing and frequently cultivated deciduous tree species for timber, fuelwood, and fodder. That is planted within the rice field borders.

Rice (*Oryza sativa* L.) is a common crop in Guilan province and is ranked the second most widely used crop in the world after maize, which has a significant impact on global food in all of the world (RAHAMAN et al. 2021). So, its high-yield planting is crucial. Nevertheless, there was no study on the allelopathy interaction of these two species. There is little knowledge available in the field of the trees' farming effects on crops in the agro-ecosystem in Iran. Therefore, this study was performed to assess the allelopathy potential effects of cottonwood (*Populus deltuides*) under different leaf extracts concentration on four local variants of rice that originated from Guilan province in the north of Iran. No study has determined these effects before. We examined the possibility of the allelopathic effects of poplar leaves extracts on rice germination, plumule, and radicle growth. The present laboratory investigation was to take the first step for revealing the species inhibitory effects of *Populus deltoides* on the germination of rice cultivars in north Iran and the most promising species to set up field exper-

iments and finally establish agroforestry systems. The present laboratory investigation was to take the first step toward revealing the species inhibitory effects of *Populus deltoides* on the germination of rice cultivars in the north of Iran. *P. deltoides* plantations are the most beneficial species to set up field experiments, and eventually establish agroforestry systems.

Materials and Methods

Plant materials

All experimental steps were performed in the laboratory at Ahar Faculty of Agriculture and Natural Resources at the University of Tabriz. Laboratory study consists of two phases, seed germination and radicle, and plumule growth phases by the following instructions.The experimental seeds were four local rice Varieties including Shiroodi, Gharib, Alikazemi, and Hashemi. All seeds were collected from the fields of Guilan province, North of Iran. The region belonged to a humid climate zone.

Preparation of aqueous leaves extracts

Fresh leaves were collected directly from the fully mature cottonwood trees at the different parts of the trees canopy (from lower, middle, and top parts) and air-dried. The leaves were powdered and screened with a one-millimeter sieve to remove visible extra residues. The 1% leaf aqueous extracts were prepared by soaking air-dried leaves powder in 100 ml of distilled water and were shaken on the shaker for one hour. The leaves' aqueous extracts were stored in a refrigerator at 4ºC for 24 hours, and these processes were repeated for two days. The Leaf aqueous extracts were filtered through a Whatman filter of 50 uniform sizes and were prepared in 100% (100 ml of aqueous leaf extract without distilled water), 75% (75 ml. Leaf extract with 25 ml; distilled water), 50% (50 ml Leaf extract with 50 ml distilled water) and 25% (25 ml; leaf extract with 75 ml distilled water).

Seed germination phase

A hundred healthy local seeds in each rice cultivar as experimental treatments including T1 (Shiroodi), T2 (Gharib), T3 (Alikazemi), and T4 (Hashemi) were counted and placed in sterilized Petri dishes (9 cm diameter) on double layers of filter paper. Each treatment was replicated four times for experimental replication. The germination experiment was performed at 25º C temperature. Samples were irrigated by leaf extractions

daily (100%, 75%. 50%, 25%, and 0% (as control, consisting of distilled water) (BOKHARY 1978). The germination was complete within six days (When the seed's germination completely stopped). This experiment was laid out in a completely randomized design with four treatments in four replications (GARIMA et al. 2017, NANDAL and DHILLON 2007).

Germination percentage was determined by counting the number of germinated seeds for six days on a daily observation basis.

Radicle and plumule growth phase

Ten seeds from the previous phase were transferred to the large culture vessels. They were kept at room temperature for 10 days and irrigation continued using each extract. Then, radicle and plumules lengths were measured and recorded. Seedlings of each replication were dried oven-dried at 70ºC until weight loss accrued and weighted to determine the dry biomass weight (CHATURVEDI and JHA 1992). The experimental design at this phase also included a completely randomized design with four treatments in four replications.

The germination index was determined by counting the number of germinated seeds (GARIMA et al. 2017).

Statistical analysis and indices calculations

Statistical analysis was performed by Tukey test among growth indices (including the germination rate, germination percentage, germination inhibition, plumule length, radicle length, total length, fresh weight, dry weight, tissue moisture content, weight vigor index, length vigor index, seed vigor index, plumule vigor index) and concentrations (S0 (Control), S1 (25%), S2 (50%), S3 (75%) and S4 (100%) in Rcommander (Rcmdr) package (FOX and BOUCHET-VALAT 2020) of R software at $P \leq 0.05$ (SIYAR et al. 2018):

$$\text{GR (germination rate)} = \Sigma \text{ (seeds germinating per day)}$$
$$\text{GP (germination percentage)} = (\text{total germinated seeds} / \text{total seeds}) \cdot 100$$
$$\text{GI (germination inhibition)} = [(\text{treatment GP} - \text{control GP})/\text{control GP}] \cdot 100$$
$$\text{WVI (weight vigor index)} = (\text{GP}/100) \cdot \text{TW (total weight)}$$
$$\text{LVI (length vigor index)} = (\text{GP}/100) \cdot \text{TL}$$
$$\text{SVI (seed vigor index)} = (\text{RL} + \text{PL}) \cdot \text{GP}$$

where:
PL – plumule length
RL – radicle length
TL – total length
FW – fresh weight – shoot and root fresh weights
DW – dry weight – shoot and root dry weights
TMC – tissue moisture content
PVI – plumule vigor index.

Result

Seed germination rate (GR) and percentage (GP) reduced significantly in increasing leaf extract concentration compared with control concentration (S0) in all treatments (Figure 1, *a*, *b* and *c*; Table Anx 1). In addition, this increase was significant between treatments (Table 1). Therefore, the seed germination was inhibited significantly in the highest concentrations. The most inhibition was in the highest leaf extract concentration (S4 = 100%).

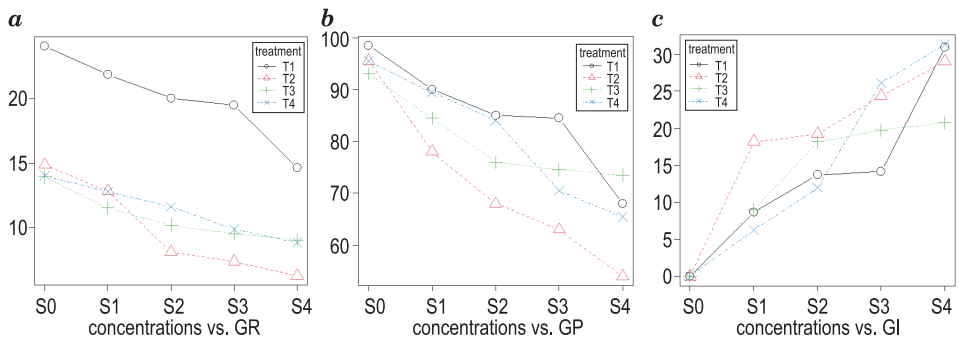

Fig. 1. Seed germination rate (GR, *a*), germination percentage (GP, *b*), and germination inhibition (GI, *c*) in different leaf extract concentrations

Table 1

ANOVA-Tukey test between treatments

	Specification	Sum of squares	df	Mean square	F	Sig.
GR	between groups	1327.427	3	442.476	56.854	.000*
	within groups	591.484	76	7.783	–	–
	total	1918.911	79	–	–	–
GP	between groups	1924.200	3	641.400	4.689	0.005*
	within groups	10395.600	76	136.784	–	–
	total	12319.800	79	–	–	–
GI	between groups	291.022	3	97.007	0.704	0.553ns
	within groups	10473.609	76	137.811	–	–
	total	10764.631	79	–	–	–
PL	between groups	12.497	3	4.166	6.094	0.001*
	within groups	51.952	76	0.684	–	–
	total	64.449	79	–	–	–
RL	between groups	31.805	3	10.602	4.590	.005*
	within groups	175.559	76	2.310	–	–
	total	207.364	79	–	–	–
TL	between groups	33.415	3	11.138	2.963	0.037*
	within groups	285.651	76	3.759	–	–
	total	319.066	79	–	–	–

Cont. Table 1

FW	between groups	0.011	3	0.004	3.134	0.030*
	within groups	0.085	76	0.001	–	–
	total	0.096	79	–	–	–
DW	between groups	0.017	3	0.006	48.517	.000*
	within groups	0.009	76	0.000	–	–
	total	0.026	79	–	–	–
SVI	between groups	124593.065	3	41531.022	5.266	0.002*
	within groups	599420.991	76	7887.118	–	–
	total	724014.055	79	–	–	–
LVI	between groups	46.564	3	15.521	3.382	0.022*
	within groups	348.827	76	4.590	–	–
	total	395.391	79	–	–	–
WVI	between groups	0.013	3	0.004	1.451	0.235ns
	within groups	0.233	76	0.003	–	–
	total	0.246	79	–	–	–
PVI	between groups	4.656E7	3	1.552E7	3.382	0.022*
	within groups	3.488E8	76	4589826.250	–	–
	total	3.954E8	79	–	–	–
TMC	between groups	1618.877	3	539.626	17.362	.000*
	within groups	2362.182	76	31.081	–	–
	total	3981.058	79	–	–	–

* Significant difference in $p \leq 0.05$; ns no significant difference

Note: GR – germination rate; GP – germination percentage; GI – germination inhibition; PL – plumule length; RL – radicle length; TL – total length; FW – fresh weight; DW – dry weight; TMC – tissue moisture content; WVI – weight vigor index; LVI – length vigor index; SVI – seed vigor index; PVI – plumule vigor index

Development of plumule, radicle, and total lengths showed a significant decrease with concentration changes irregularly. However, increasing in concentration of leaf extracts generally reduced the length of growth of plumule and radicle compared to the control level (S0 = Distilled water). The relevant effects appeared on the seedlings' survival by delay in radical growth (Figure 2; *a*, *b*, and *c*).

Therefore, the seedlings' growth decreased significantly with an increase in the concentration of extracts in all rice cultivars (Table 1, Table 2).

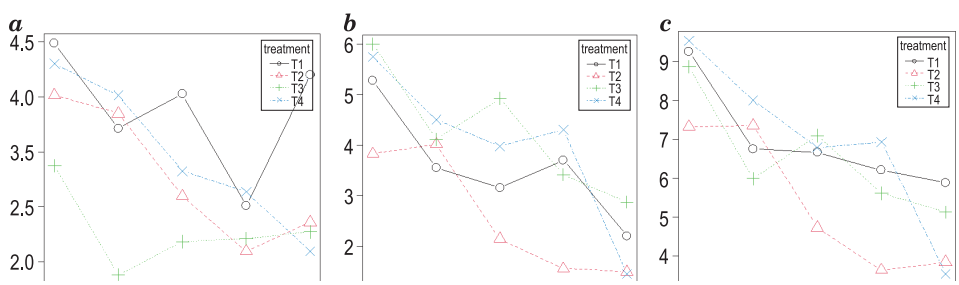

Fig. 2. Plumule length (PL, *a*), radicle length (RL, *b*), and total length (TL, *c*) in the different leaf extract concentrations

Table 2

Pearson correlations test

Specification		GR	FW	DW	PL	RL	TL	GP	GI	SVI	LVI	WVI	PVI	TMC
GR	correlation	1	0.127 ^{ns}	-0.530*	0.487*	0.348*	0.499*	0.733*	-0.523*	0.645*	0.608*	0.541*	0.608*	0.551*
	Sig	–	0.260	.000	.000	.002	.000	.000	.000	.000	.000	.000	.000	.000
FW	correlation	–	1	0.224*	0.512*	0.288*	0.463*	0.406*	-0.433*	0.584*	0.514*	0.805*	0.514*	0.510*
	Sig	–	–	0.045	.000	.010	.000	.000	.000	.000	.000	.000	.000	.000
DW	correlation	–	–	1	-0.162 ^{ns}	-0.262*	-0.283*	-0.303*	0.137 ^{ns}	-0.224*	-0.289*	-0.081 ^{ns}	-0.289*	-0.714*
	Sig	–	–	–	0.152	0.019	0.011	0.006	0.224	0.046	0.009	0.477	0.009	.000
PL	correlation	–	–	–	1	0.204 ^{ns}	0.614*	0.451*	-0.314*	0.937*	0.616*	0.576*	0.616*	0.514*
	Sig	–	–	–	–	0.069	.000	.000	0.005	.000	.000	.000	.000	.000
RL	correlation	–	–	–	–	1	0.898*	0.622*	-0.502*	0.394*	0.853*	0.557*	0.853*	0.409*
	Sig	–	–	–	–	–	.000	.000	.000	.000	.000	.000	.000	.000
TL	correlation	–	–	–	–	–	1	0.704*	-0.546*	0.738*	0.964*	0.707*	0.964*	0.560*
	Sig	–	–	–	–	–	–	.000	.000	.000	.000	.000	.000	.000
GP	correlation	–	–	–	–	–	–	1	-0.841*	0.721*	0.848*	0.866*	0.848*	0.547*
	Sig	–	–	–	–	–	–	–	.000	.000	.000	.000	.000	.000
GI	correlation	–	–	–	–	–	–	–	1	-0.573*	-0.703*	-0.779*	-0.703*	-0.415*
	Sig	–	–	–	–	–	–	–	–	.000	.000	.000	.000	.000
SVI	correlation	–	–	–	–	–	–	–	–	1	0.800*	0.789*	0.800*	0.614*
	Sig	–	–	–	–	–	–	–	–	–	.000	.000	.000	.000
LVI	correlation	–	–	–	–	–	–	–	–	–	1	0.829*	1.000*	0.601*
	Sig	–	–	–	–	–	–	–	–	–	–	.000	.000	.000
WVI	correlation	–	–	–	–	–	–	–	–	–	–	1	0.829*	0.632*
	Sig	–	–	–	–	–	–	–	–	–	–	–	.000	.000
PVI	correlation	–	–	–	–	–	–	–	–	–	–	–	1	0.601*
	Sig	–	–	–	–	–	–	–	–	–	–	–	–	.000

* Correlation is significant at the 0.05 level

Fresh weight (FW) decreased in T1, T3, and T4 cultivars by increasing extract concentrations but slightly improved in S2 and S4. In contrast, dry weight (DW) showed an insignificant increasing trend with increasing concentrations of extracts (Figure 3; *b*; Table 1). These results were consistent with the tissue moisture content (TMC) results diagram (Figure 3; *c*).

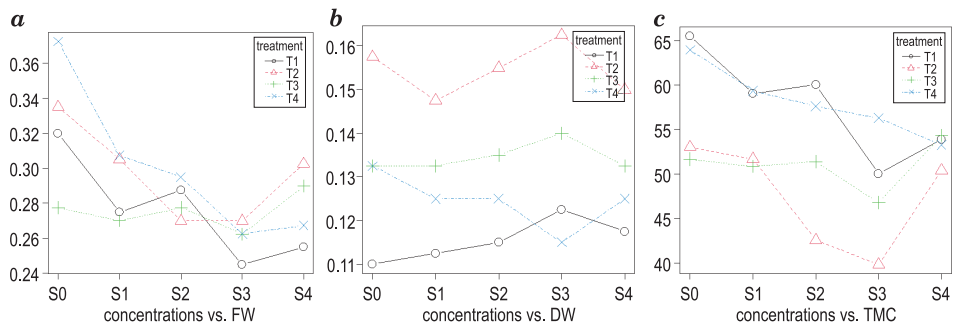

Fig. 3. Fresh weight (FW, *a*), Dry weight (DW, *b*), and Tissue moisture content (TMC, *c*) in different leaf extract concentrations

All leaf extracts significantly suppressed the growth of plumule and radicle of rice culti vars. In addition, the weight and length vigor indices (WVI and LVI, respectively) and plumule vigor index (PVI) reduced with increasing leaf extract concentration regularly (Figure 4; *a*, *b* & *c*; Table Anx 1).

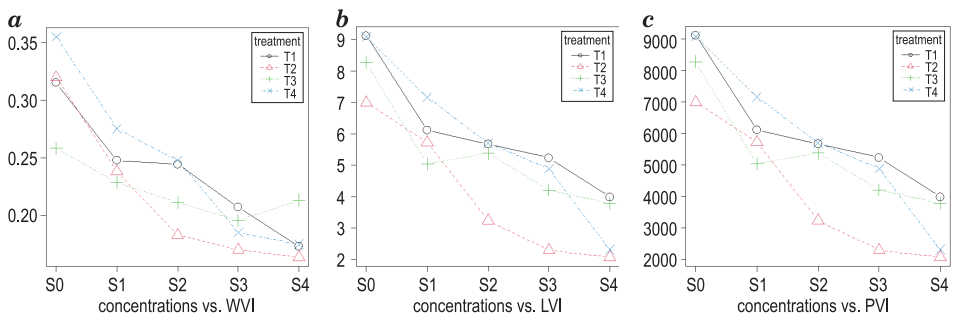

Fig. 4. Weight vigor index (WVI, *a*), length vigor index (LVI, *b*), and plumule vigor index (PVI, *c*) in different leaf extract concentrations

Note:
Leaf extract consecration levels (X-axis): in control (S0), 25% (S1), 50% (S2), 75% (S3) and 100% (S4) consecrations
Indices (Y-axis): GR – germination rate; GP – germination percentage; GI – germination inhibition; PL – plumule length; RL – radicle length; TL – total length; FW – fresh weight; DW – dry weight; TMC – tissue moisture content; WVI – weight vigor index; LVI – length vigor index; SVI – seed vigor index; PVI – plumule vigor index; T1, T2, T3, and T4 – treatments of the local rice cultivars

Pearson correlation test to assess the linear correlation among variables showed that there was a non-significant correlation among PL and RL (0.204), DW and PL (0.162), DW and GI (0.137), GR and FW (0.127), DW and WVI (0.081).

In contrast, There was a strong significant positive correlation ($r \geq 0.7$) between SVI and WVI (0.789), TL and SVI (0.738), GR and GP (0.733), GP and SVI (0.721), TL and GP (0.704), TL and WVI (0.707), RL and TL (0.898), WVI and GP (0.866), RL and LVI (0.853), RL and PVI (0.853), GP and LVI (0.848), GP and PVI (0.848), LVI and WVI (0.829), WVI and PVI (0.829), FW and WVI (0.805), SVI and LVI (0.800), SVI and PVI (0.800), PL and SVI (0.937), TH and LVI (0.964) and TL and PVI (0.964). In addition, There was a strong significant negative correlation ($r \geq -0.7$) between GP and GI (-0.841), GI and WVI (-0.779), DW and TMC (-0.714), GI and LVI (-0.703), and GI and PVI (-0.703) – Table 2.

Discussions

The present study revealed a decrease in rice seedling growth with an increase in the cottonwood leaves extract concentration. Furthermore, this inhibited behavior consisted of seed germination indices and plumule and radicle growth. These results were consistent with KHALID et al. (2021). They concluded that the fresh and dry leaves extract of *Populus nigra* negatively affected the germination and seedling growth of *Brassica campestris* and indicates some allelochemicals might be present in this tree. Also, a similar result was reported by ZUBAY et al. (2021). The allelopathic treatment of *Populus tremula* L. leaves extracts on some medicinal and aromatic plants showed that Poppy and Angelica proved to be the most sensitive species to the treatments. The allelochemicals existing in the higher concentrations of leaf extracts reduced the photosynthetic activity of seedlings and caused a decrease in the dry weight of the target plants (LI et al. 2021).

This study showed that germination and seedling growth significantly reduced in all tested crops in response to *P. deltoides* leaves leachate. The researchers found that the soil of poplar stands was rich in phytotoxic phenolic (SINGH et al. 2001, ZUBAY et al. 2021). Therefore, it is recommended to examine soil content in poplar stands or rice fields in terms of the allelochemicals present in the different soil horizons.

In our study, the allelopathic effects were severe at higher concentrations. This result does not support the MAJEED et al. (2017) that showed the higher extracts concentration of *P. deltoides* leaves had no effects on

the seedling growth parameters. The germination percentage (GP) of wheat was not influenced by the higher concentration of poplar leaves extracts and on the contrary, it was promoted by lower extract concentrations. In contrast, our results were supported by the WANG et al. (2021) that has reported a significant reduction in seed germination and seedling growth of *Amygdalus pedunculata* under concentration increase of some *Pinus* species leaves aquatic extracts. CATALAN et al. (2013) pointed out that *Populus alba* had an intense allelopathic effect and they also emphasized on the extracts' concentration level impacts on prohibition process. Moreover, inhibitory effects of *P. deltiodes* leaf extracts on crops in laboratory or field experiments were shown by SHARMA et al (2000) and SINGH et al. (2001). They also demonstrated its stimulant effects at lower concentrations. Conversely, experiments on the extracts of *Melia azedarach* leaves at all concentration levels revealed inhibitory effects on the seedling growth of wheat (MAJEED et al. 2017). As a result, the allelopathy process is strongly dependent on the type of adjacent or target plants. Consequently, allelopathic effects correlated with the target plant, the type of allelochemicals, and extract concentrations.

Based on the conclusion of the LIU et al. (2003), more inhibition of seedling growth might lead to a theory that more extractions levels contain the higher contents of allelochemicals. In other words, the inhibitory effect of allelopathic plants on other plants was strongly correlated with the concentration percentage and type of allelochemicals (MAJEED et al. 2017).

In addition, the tree-crop-soil interaction demonstrated that the reduction in the growth of the crops associated with *P. deltoides* phenolic presence was released from tree leaves in the soil in this interaction process (SINGH et al. 2001).

Since it is not unexpected that cottonwood is a deciduous tree species and the fallen leaves in autumn and winter on the field floor may be released and accumulate the allelochemicals into the soil. Therefore, soil study is strongly recommended (WANG et al. 2021) because, in field experiments, this process may cause the reduce yield of crops by falling litter under the shelter of cottonwood (KHAKET et al. 2014).

The growth reduction of crops was due to allelopathic interference of phytotoxin phenolics content of cottonwood leaves. Therefore, it is suggested that the content of *P. deltoides* leaf extract should be examined because it was shown the presence of bioactive compounds (that have a role in the allelopathic effects), alkaloids, flavonoids, steroids, terpenoids, saponins, tannins, phlorotannins, glycoside, triterpenes, and phytosteroid in the aqueous leaf extract of *Populus nigra* (INAYAT et al. 2020). In the

same experiment, *Populus tremula* L. trees synthesize compounds typically derived from the shikimate-phenylpropanoid pathway (phenol glycosides, hydroxy-cinnamates, flavonoids, and condensed tannins) (ZUBAY et al. 2021). However, terpenoids and fatty acids are also present in considerable concentrations. The most likely responsible compounds are the phenolic acids. They decompose from decaying leaves to soil.

According to the results of the present study, *Oryza sativa* L. is a sensitive crop species and has poor growth yield under the influence of cottonwood leaf extraction. Similar conclusions were obtained from the KOUL et al. (1991).

They reported a laboratory experiment to investigate the allelopathic activity of leaf leachates from eight commonly grown farm tree species (*Acacia nilotica*, *Dalbergia sissoo*, *Bauhinia variegata*, *Ficus bengalensis*, *Morus alba*, *Populus deltoides*, *Salix babylonica*, and *Leucaena leucocephala*) on the seed germination and the early growth parameters of rice. All leaf leachates inhibited seed germination and seedling growth (with a maximum reduction by *L. Leucocephala*, and *A. nilotica*). Therefore, it was recommended to use more parameters to measure allelopathic effects for suggesting some feasible crops species for a multicultural agroforestry system.

Overall, the type of extractions (leaves, roots, bark, flowers, or fruits), type of target plant, type of allelochemicals, and amount of concentration is the most efficacious factors in an allelopathy interaction. A change that accrued in any aspect could affect the allelopathic potential and reduce the germination of crops (MAJEED et al. 2017). A lower extraction concentration level of *P. deltoides* increases the total length growth of wheat (MAJEED et al. 2017), but all concentrations had high effects on reducing the growth of all rice cultivars in our study. Therefore, the target plant selection is a fundamental phase.

Declining enzyme activity, impairing mineral ions uptake, decreasing cells division and energy limitation, inhibiting photosynthesis, damage to cells, reducing the plant's ability to remove active oxygen, destroying the structure of plant cell membranes, thus weakening the protective effect has introduced as the main factors that resulting from allelochemicals activity in plants. Consequently, they could limit the seeds' germination and seedlings' growth (ABEDI and ABDI 2021, ZHANG et al. 2021, LI et al. 2021).

Conclusions

The current study demonstrated that the allelopathic effects of cottonwood should be attended seriously on tested rice cultivars because it is planting increasingly inside or around the rice fields in recent years in the north of Iran. While according to the results of this study, the fall of cottonwood leaves on rice fields is affected adversely and has inhibitory effects on the growth characteristics of rice seedlings and products. Therefore, it is necessary to consider that the cottonwood is not recommended as a suitable species for agroforestry with rice, but further research is needed to conduct mixed cultivation with other agricultural species and crops. Also, an additional study under natural field conditions is essential for evaluating the effect of local natural conditions on plant growth and an additional study on natural conditions seems to be necessary to assess the impacts of local natural conditions on plant growth, crop yield, and the physical and chemical properties of soil under field cultivation. Further research is necessary to identify the allelochemicals in other parts of the cottonwood tree (such as roots by secreting allelochemicals into the soil) that prohibit or promote the growth of rice or other crops. The results of this experiment may provide a consequential basis for further field experimental studies.

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Annex

Table Anx 1

The results of multiple comparisons of Tukey test

Dependent variable	Concentration (I)	Concentration (J)	Mean difference (I–J)	Std. error	Sig.
GR	1	2	1.99369	1.54619	.698
		3	4.25613	1.54619	.056
		4	5.18431*	1.54619	.011*
		5	7.04150*	1.54619	.000*
	2	1	-1.99369	1.54619	.698
		3	2.26244	1.54619	.589
		4	3.19063	1.54619	.247
		5	5.04781*	1.54619	.014*
	3	1	-4.25613	1.54619	.056
		2	-2.26244	1.54619	.589
		4	.92819	1.54619	.975
		5	2.78538	1.54619	.380
	4	1	-5.18431*	1.54619	.011*
		2	-3.19063	1.54619	.247
		3	-.92819	1.54619	.975
		5	1.85719	1.54619	.751
	5	1	-7.04150*	1.54619	.000*
		2	-5.04781*	1.54619	.014*
		3	-2.78538	1.54619	.380
		4	-1.85719	1.54619	.751
FW	1	2	.03687*	.00984	.003*
		3	.04375*	.00984	.000*
		4	.06625*	.00984	.000*
		5	.04750*	.00984	.000*
	2	1	-.03687*	.00984	.003*
		3	.00688	.00984	.956
		4	.02938*	.00984	.031*
		5	.01063	.00984	.817
	3	1	-.04375*	.00984	.000*
		2	-.00688	.00984	.956
		4	.02250	.00984	.161
		5	.00375	.00984	.995

Cont. Table Anx 1

FW	4	1	-.06625*	.00984	.000*
		2	-.02938*	.00984	.031*
		3	-.02250	.00984	.161
		5	-.01875	.00984	.324
	5	1	-.04750*	.00984	.000*
		2	-.01063	.00984	.817
		3	-.00375	.00984	.995
		4	.01875	.00984	.324
DW	1	2	.00375	.00650	.978
		3	.00062	.00650	1.000
		4	-.00188	.00650	.998
		5	.00187	.00650	.998
	2	1	-.00375	.00650	.978
		3	-.00312	.00650	.989
		4	-.00562	.00650	.908
		5	-.00187	.00650	.998
	3	1	-.00062	.00650	1.000
		2	.00312	.00650	.989
		4	-.00250	.00650	.995
		5	.00125	.00650	1.000
	4	1	.00188	.00650	.998
		2	.00562	.00650	.908
		3	.00250	.00650	.995
		5	.00375	.00650	.978
	5	1	-.00187	.00650	.998
		2	.00187	.00650	.998
		3	-.00125	.00650	1.000
		4	-.00375	.00650	.978
PL	1	2	.55312	.29351	.335
		3	.75937	.29351	.083
		4	1.17812*	.29351	.001*
		5	.93438*	.29351	.018*
	2	1	-.55312	.29351	.335
		3	.20625	.29351	.955
		4	.62500	.29351	.219
		5	.38125	.29351	.693

Cont. Table Anx 1

PL	3	1	-.75937	.29351	.083
		2	-.20625	.29351	.955
		4	.41875	.29351	.613
		5	.17500	.29351	.975
	4	1	-1.17812*	.29351	.001*
		2	-.62500	.29351	.219
		3	-.41875	.29351	.613
		5	-.24375	.29351	.920
	5	1	-.93438*	.29351	.018*
		2	-.38125	.29351	.693
		3	-.17500	.29351	.975
		4	.24375	.29351	.920
RL	1	2	1.16875	.44637	.077
		3	1.66562*	.44637	.003*
		4	1.97187*	.44637	.000*
		5	3.21250*	.44637	.000*
	2	1	-1.16875	.44637	.077
		3	.49687	.44637	.799
		4	.80313	.44637	.382
		5	2.04375*	.44637	.000*
	3	1	-1.66562*	.44637	.003*
		2	-.49687	.44637	.799
		4	.30625	.44637	.959
		5	1.54688*	.44637	.008*
	4	1	-1.97187*	.44637	.000*
		2	-.80313	.44637	.382
		3	-.30625	.44637	.959
		5	1.24062	.44637	.052
	5	1	-3.21250*	.44637	.000*
		2	-2.04375*	.44637	.000*
		3	-1.54688*	.44637	.008*
		4	-1.24062	.44637	.052
TL	1	2	1.72187*	.52074	.012*
		3	2.42500*	.52074	.000*
		4	3.15000*	.52074	.000*
		5	4.14687*	.52074	.000*

Cont. Table Anx 1

TL	2	1	-1.72187*	.52074	.012*
		3	.70312	.52074	.661
		4	1.42812	.52074	.057
		5	2.42500*	.52074	.000*
	3	1	-2.42500*	.52074	.000*
		2	-.70312	.52074	.661
		4	.72500	.52074	.634
		5	1.72187*	.52074	.012*
	4	1	-3.15000*	.52074	.000¬*
		2	-1.42812	.52074	.057
		3	-.72500	.52074	.634
		5	.99688	.52074	.319
	5	1	-4.14687*	.52074	.000*
		2	-2.42500*	.52074	.000*
		3	-1.72187*	.52074	.012*
		4	-.99688	.52074	.319
GP	1	2	10.12500*	2.46965	.001*
		3	17.37500*	2.46965	.000*
		4	22.50000*	2.46965	.000*
		5	30.37500*	2.46965	.000*
	2	1	-10.12500*	2.46965	.001*
		3	7.25000*	2.46965	.035*
		4	12.37500*	2.46965	.000*
		5	20.25000*	2.46965	.000*
	3	1	-17.37500*	2.46965	.000*
		2	-7.25000*	2.46965	.035*
		4	5.12500	2.46965	.242
		5	13.00000*	2.46965	.000*
	4	1	-22.50000*	2.46965	.000*
		2	-12.37500*	2.46965	.000*
		3	-5.12500	2.46965	.242
		5	7.87500*	2.46965	.017*
	5	1	-30.37500*	2.46965	.000*
		2	-20.25000*	2.46965	.000*
		3	-13.00000*	2.46965	.000*
		4	-7.87500*	2.46965	.017*

Cont. Table Anx 1

GI	1	2	-10.50816*	2.41756	.000*
		3	-15.76414*	2.41756	.000*
		4	-21.11811*	2.41756	.000*
		5	-28.05624*	2.41756	.000*
	2	1	10.50816*	2.41756	.000*
		3	-5.25598	2.41756	.201
		4	-10.60994*	2.41756	.000*
		5	-17.54808*	2.41756	.000*
	3	1	15.76414*	2.41756	.000*
		2	5.25598	2.41756	.201
		4	-5.35396	2.41756	.186
		5	-12.29210*	2.41756	.000*
	4	1	21.11811*	2.41756	.000*
		2	10.60994*	2.41756	.000*
		3	5.35396	2.41756	.186
		5	-6.93813*	2.41756	.041*
	5	1	28.05624*	2.41756	.000*
		2	17.54808*	2.41756	.000*
		3	12.29210*	2.41756	.000*
		4	6.93813*	2.41756	.041*
PVI	1	2	2358.62500*	4.70730E2	.000*
		3	3371.18750*	4.70730E2	.000*
		4	4210.43750*	4.70730E2	.000*
		5	5324.25000*	4.70730E2	.000*
	2	1	-2358.62500*	4.70730E2	.000*
		3	1012.56250	4.70730E2	.210
		4	1851.81250*	4.70730E2	.002*
		5	2965.62500*	4.70730E2	.000*
	3	1	-3371.18750*	4.70730E2	.000*
		2	-1012.56250	4.70730E2	.210
		4	839.25000	4.70730E2	.391
		5	1953.06250*	4.70730E2	.001*
	4	1	-4210.43750*	4.70730E2	.000*
		2	-1851.81250*	4.70730E2	.002*
		3	-839.25000	4.70730E2	.391
		5	1113.81250	4.70730E2	.136

Cont. Table Anx 1

PVI	5	1	-5324.25000*	4.70730E2	.000*
		2	-2965.62500*	4.70730E2	.000*
		3	-1953.06250*	4.70730E2	.001*
		4	-1113.81250	4.70730E2	.136
LVI	1	2	2.35862*	.47073	.000*
		3	3.37119*	.47073	.000*
		4	4.21044*	.47073	.000*
		5	5.32425*	.47073	.000*
	2	1	-2.35862*	.47073	.000*
		3	1.01256	.47073	.210
		4	1.85181*	.47073	.002*
		5	2.96562*	.47073	.000*
	3	1	-3.37119*	.47073	.000*
		2	-1.01256	.47073	.210
		4	.83925	.47073	.391
		5	1.95306*	.47073	.001*
	4	1	-4.21044*	.47073	.000*
		2	-1.85181*	.47073	.002*
		3	-.83925	.47073	.391
		5	1.11381	.47073	.136
	6	1	-5.32425*	.47073	.000*
		2	-2.96562*	.47073	.000*
		3	-1.95306*	.47073	.001*
		4	-1.11381	.47073	.136
WVI	1	2	.06456*	.01070	.000*
		3	.09050*	.01070	.000*
		4	.12250*	.01070	.000*
		5	.13075*	.01070	.000*
	2	1	-.06456*	.01070	.000*
		3	.02594	.01070	.120
		4	.05794*	.01070	.000*
		5	.06619*	.01070	.000*
	3	1	-.09050*	.01070	.000*
		2	-.02594	.01070	.120
		4	.03200*	.01070	.030*
		5	.04025*	.01070	.003*

Cont. Table Anx 1

WVI	4	1	-.12250*	.01070	.000*
		2	-.05794*	.01070	.000*
		3	-.03200*	.01070	.030*
		5	.00825	.01070	.938
	5	1	-.13075*	.01070	.000*
		2	-.06619*	.01070	.000*
		3	-.04025*	.01070	.003*
		4	-.00825	.01070	.938
TMC	1	2	3.30825	2.26444	.591
		3	5.64381	2.26444	.103
		4	10.28544*	2.26444	.000*
		5	5.56706	2.26444	.111
	2	1	-3.30825	2.26444	.591
		3	2.33556	2.26444	.840
		4	6.97719*	2.26444	.023*
		5	2.25881	2.26444	.856
	3	1	-5.64381	2.26444	.103
		2	-2.33556	2.26444	.840
		4	4.64163	2.26444	.253
		5	-.07675	2.26444	1.000
	4	1	-10.28544*	2.26444	.000*
		2	-6.97719*	2.26444	.023*
		3	-4.64163	2.26444	.253
		5	-4.71837	2.26444	.238
	5	1	-5.56706	2.26444	.111
		2	-2.25881	2.26444	.856
		3	.07675	2.26444	1.000
		4	4.71837	2.26444	.238
SVI	1	2	82.56250*	26.23192	.019*
		3	120.11875*	26.23192	.000*
		4	165.93125*	26.23192	.000*
		5	168.28750*	26.23192	.000*
	2	1	-82.56250*	26.23192	.019*
		3	37.55625	26.23192	.609
		4	83.36875*	26.23192	.018*
		5	85.72500*	26.23192	.014*

Cont. Table Anx 1

SVI	3	1	-120.11875*	26.23192	.000*
		2	-37.55625	26.23192	.609
		4	45.81250	26.23192	.412
		5	48.16875	26.23192	.361
	4	1	-165.93125*	26.23192	.000*
		2	-83.36875*	26.23192	.018*
		3	-45.81250	26.23192	.412
		5	2.35625	26.23192	1.000
	5	1	-168.28750*	26.23192	.000*
		2	-85.72500*	26.23192	.014*
		3	-48.16875	26.23192	.361
		4	-2.35625	26.23192	1.000

* The mean difference is significant at the 0.05 level