

ALLELOPATHIC EFFECTS OF DIFFERENT WEED EXTRACTS ON SEED GERMINATION AND SEEDLING GROWTH OF WHEAT

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Abstract

The present study identified wheat varieties with strong grass resistance, clarified the physiological mechanism of their anti-grassing effect, and provided a scientific basis for the application of grass-resistant wheat varieties for producing high and stable yield of wheat. Ten wheat varieties commonly used in production were selected to study the effects of *Descurainia sophia*, *Galium tricorne*, *Wild oat*, and *sativa* extracts on seed germination and seedling growth. Results showed that the *Descurainia sophia*, *Galium tricorne*, *Wild oat*, and *Vicia sativa* extracts exhibited allelopathic inhibition on wheat seed germination and seedling growth. The allelopathic indices of wheat seed germination rate, seedling height, and root length were -0.12 to -0.19 , -0.70 to -0.87 , and -0.70 to -0.91 , respectively. The weed extract had certain differences in allelopathic inhibition on wheat. The *Wild oat* extract had the strongest inhibitory effect on seed germination, whereas the *Descurainia sophia* extract showed the weakest inhibitory effect. The *Vicia sativa* extract had the strongest inhibitory effect on seedling growth, whereas the *Galium tricorne* extract showed the weakest inhibitory effect. According to the absolute value of allelopathic index, the effects of the four kinds of weed extracts on seedling growth index were systematically clustered. Results showed that the allelopathic inhibition rate of weed extracts on *Wanmai 19* was weak, and the weed resistance was strong. The weed extracts had an allelopathic effect on the root activity and activities of superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT) in the leaves and could promote the content of malondialdehyde (MDA) in the leaves. However, they had minimal allelopathic effect on *Wanmai 19*. The seedling height and root length of wheat seedlings were significantly correlated with root activity, activities of SOD, POD, and CAT in the leaves and MDA content. Thus, the effect of allelochemicals on the cell membrane in weed extracts could be the cause of inhibited wheat growth.

Key words: Wheat, Weed extract, Allelopathic effect, Root activity, Physiological indicator.

Introduction

Weeds are one of the most important biological factors that affect wheat production. In wheat fields, weeds not only compete with wheat for sunshine, space, and nutrition but also climb stalks, causing crops to fall and affecting their harvest. These problems pose a serious threat to wheat production. Because of weeds, the annual world wheat production has decreased by 15%, approximately 1.5 million tons (Jiang *et al.*, 2018; Gao *et al.*, 2016; Ramesh *et al.*, 2017). *Descurainia sophia*, *Galium tricorne*, *Wild oat*, and *Vicia sativa* are the main weeds that damage the wheat field. The number of their producing seeds are large, and their seed emergence period is uneven, making the prevention of their occurrence difficult. Thus, they have greatly influence the growth and yield of wheat (Reiss *et al.*, 2018; Li *et al.*, 2011; Cui *et al.*, 2011). The grass resistance among different wheat varieties has certain differences. Therefore, identifying wheat varieties with strong grass resistance and clarifying the mechanism of grass resistance have great application value.

Plants affect the growth and development of neighboring plants by releasing allelochemicals into the surrounding environment. This natural phenomenon is called plant allelopathy. Allelopathic substances between higher plants are released into the environment by the volatilization of stems and leaves, leaching of stems and leaves, and secretion of roots of living plants (donor plants), thereby doing good or harm to surrounding plants (recipient plants). These processes are an important chemical and ecological defense mechanism that exists widely in nature (Zimdahl, 2018; Marwat *et al.*, 2008; Li,

2014). Weeds in a wheat field can affect the growth and yield of wheat. One of the main reasons is that weeds release allelochemicals into the surrounding environment through the allelopathic effect, which affects the growth and development of wheat, thus affecting its yield.

Previous studies have shown that the water extracts from aboveground plants and roots of *Wild oat* have a significant effect on plant height, dry weight, root length, and root activity of wheat seedlings (Li *et al.*, 2018). The extract of *Descurainia sophia* can inhibit wheat seed germination and wheat seedling growth and increase malondialdehyde (MDA) content in wheat leaves, and the intensity of allelopathic inhibition effect varies among different wheat lines (Wu *et al.*, 2009; Yang & Mu, 2006). The extract of *Galium tricorne* can also inhibit seed germination and seedling growth, and the allelopathic inhibition effects of different wheat varieties vary (Zhang *et al.*, 2015b). Previous studies have shown that the weeds in the wheat field have allelopathic effects on wheat, but the reports are mostly allelopathic effects of single weeds. The comprehensive allelopathic effects of different wheat varieties on various associated weeds and the physiological mechanisms of weeds on the allelopathic effects still need to be further studied and discussed. In the present study, 10 wheat varieties commonly used in production were selected to investigate the effects of *Descurainia sophia*, *Galium tricorne*, *Wild oat*, and *Vicia sativa* extracts on seed germination and seedling growth. This study identified wheat varieties that are resistant to the allelopathic effects of weed extracts and elucidated their physiological mechanism. The results will provide a scientific basis for the application of grass-tolerant wheat varieties to produce high and stable yield of wheat.

Materials and Methods

Experimental materials: The 10 wheat varieties tested were *Yannong19*, *Yannong21*, *Jimai22*, *Lunong116*, *Kaimai 18*, *Zhengmai366*, *Wanmai19*, *Wankenmai1*, *Wanmai50*, and *Wanmai54*. The four weeds tested were *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa*, which were collected at the Planting Science and Technology Park of Anhui Science and Technology University during the flowering period. The impurities in the weed plants were removed, then set at 105°C for 1 h. After drying to a constant weight at 70°C, the plants were cut into small pieces (about 5 cm size) and pulverized into powder with a pulverizer. Then, 35 g of plant dry matter was soaked in 1,000 mL of distilled water for 48 h at room temperature and filtered twice with gauze to obtain extract with concentration of 0.035 g·mL⁻¹.

Seed germination test: Wheat seeds were disinfected with 0.1% mercuric chloride for 1–3 min, rinsed with distilled water for 3–4 times, and soaked for 3–4 h. Fifty wheat seeds were selected and placed in a culture dish with two layers of filter papers (the diameter was 15 cm). Subsequently, 10 mL of each weed extract was added. The test was repeated thrice with 0 g·mL⁻¹ as the distilled water control. The culture dish was placed indoors (20°C–25°C) for 3 days in a non-light artificial climate and cultured for 12 h each day from the fourth day for 7 days.

Seedling growth test: Approximately 300 g of dried fine sand and 86 mL of weed extract were added to the germination box (19 × 13 cm), and the treated wheat seeds were sown in the prepared germination box. Fifty wheat seeds were sown in each germination box with distilled water as the control. The cultivation was continued for 15 days in accordance with the culture conditions of the seed germination test. The experiment was set up with nine replicates: three were used to determine indicators of seedling growth, and six were used for physiological indicators of seedlings. The seedling height and main root length of wheat seedlings were measured after washing the fine sand and absorbing moisture using absorbent paper. The root activity and the contents of superoxide dismutase (SOD), peroxidase (POD), catalase (CAT) and MDA in the leaves were determined.

Determination of seed germination indicators: The number of seed germinations per day was counted during the germination period. The germination potential, germination rate, and germination indicators of seeds were calculated. Germination potential = number of germinated seeds within 3 days/number of test seeds × 100%; germination rate = number of germinated seeds within 7 days/number of test seeds × 100%; germination index = $\sum Gt/Dt$ (where Gt is the number of germinated seeds at time t and Dt is the corresponding germination days) (Chen *et al.*, 2016).

Determination of physiological indicators: The triphenyltetrazolium chloride method was used to test for root activity (Zhou *et al.*, 2012; Li *et al.*, 2000). SOD activity was determined by xanthine oxidation (Huang *et al.*, 2018). POD activity was determined in accordance with the principle of POD-catalyzed H₂O₂, and the

absorbance at 420 nm was measured (Sharma *et al.*, 2011). CAT activity was determined using the pale yellow complex formed by ammonium molybdate and H₂O₂ by colorimetry at 405 nm (Sharma *et al.*, 2011). MDA content was determined by thiobarbituric acid method (Li *et al.*, 2015).

Statistical calculation of data: The following equations were used to calculate allelopathy indicators: $RI=1-C/T(T \geq C)$ and $RI=T/C-1(T \leq C)$, where T is the processing value, C is the control value, $RI > 0$ indicates promotion, and $RI < 0$ indicates inhibition. The absolute values are consistent with the intensity of allelopathy action (Zhang *et al.*, 2015a).

The test data were collated with Microsoft Excel 2016. Statistical and correlation analyses were performed using DPS 7.55 software, and Excel 2016 was used for plotting.

Results

Germination potential, germination rate, and germination indicators of wheat seeds: Table 1 shows that under the standard germination conditions, the varieties with the highest germination potential, germination rates, and germination indicators were *Wanmai50*, *Kaimai18*, and *Zhengmai366*. The variety with the lowest germination potential, germination rates, and germination indicators was *Wanmai19*, and the differences between the varieties were 184.62%, 20.48%, and 269.17%. However, the germination rates of *Wanmai19* and *Wankenmai1* were below 90%, whereas those of other varieties were all above 90%. The four weed extracts significantly reduced the germination potential, germination rate, and germination index of wheat seeds. On the basis of the average effect of weed extracts on the allelopathic action of wheat, the germination potential, germination rates, and germination indicators of *Wild oat* extract decreased by 24.06%, 18.52%, and 32.68%, respectively, and the allelopathic indicators reached -0.28, -0.19, and -0.36, respectively, indicating significant allelopathic inhibition. The germination potential, germination rates, and germination indicators of *Descurainia sophia* extract decreased by 14.19%, 12.64%, and 22.78%, respectively, and the allelopathic indicators reached -0.14, -0.12, and -0.22, respectively, indicating low allelopathic inhibition. The *Descurainia sophia* extract had the greatest influence on the germination potential, germination rate, and germination indicators of *Lunong116*, and the least affected varieties were *Yannong19*, *Yannong19*, and *Yannong21*. The *Galium tricornis* extract had the greatest influence on the germination potential, germination rates, and germination indicators of *Wankenmai1*, *Wanmai19*, and *Wankenmai1*, and the least affected variety was *Yannong19*. The *Wild oat* extract had the greatest influence on the germination potential, germination rates, and germination indicators of *Wanmai19*, and the least affected variety was *Yannong19*. The *Vicia sativa* extract had the greatest influence on the germination potential, germination rates, and germination indicators of *Wanmai19*, *Wanmai19*, and *Zhengmai366*, respectively. The least affected variety was *Yannong19*. Given the difference of weed species, different weed extracts had different allelopathic effects on seed germination of different wheat varieties.

Table 1. Effect of weed extract on germination potential, germination rate and germination index of wheat seeds.

Variety	Extract	Germination potential		Germination rate		Germination index	
		Result (%)	RI	Result (%)	RI	Result (%)	RI
Yannong 19	WT	92.67 ± 2.31a	—	95.33 ± 1.15a	—	117.88 ± 3.15a	—
	DE	90 ± 2a	-0.028 ± 0.006a	93.33 ± 3.06a	-0.021 ± 0.001a	103.39 ± 0.48c	-0.122 ± 0.008b
	GE	87.33 ± 2.31a	-0.057 ± 0.005ab	92.67 ± 2.31a	-0.028 ± 0.004a	108.75 ± 0.68bc	-0.077 ± 0.001a
	WE	88 ± 3.46a	-0.051 ± 0.009b	92 ± 2a	-0.035 ± 0.006a	109.04 ± 3.37bc	-0.076 ± 0.009a
	VE	88 ± 3.46a	-0.05 ± 0.007ab	92 ± 4a	-0.035 ± 0.005a	110.75 ± 2.09b	-0.06 ± 0.002a
Yangnong 21	WT	96 ± 0a	—	97.33 ± 1.15a	—	118.05 ± 2.76a	—
	DE	88.67 ± 3.06b	-0.076 ± 0.006a	92.67 ± 1.15b	-0.048 ± 0.003a	107.09 ± 1.95b	-0.092 ± 0.002a
	GE	72.67 ± 3.06c	-0.243 ± 0.008bc	88.33 ± 1.53c	-0.096 ± 0.005b	79.16 ± 1.61c	-0.328 ± 0.015b
	WE	74 ± 4c	-0.229 ± 0.002b	81.33 ± 3.06d	-0.164 ± 0.01c	79.1 ± 2.24c	-0.33 ± 0.012b
	VE	68 ± 6c	-0.292 ± 0.015c	78 ± 2.65d	-0.198 ± 0.006d	73.07 ± 3.16d	-0.383 ± 0.021c
Jimai 22	WT	86.67 ± 5.03a	—	93.33 ± 2.31a	—	107.73 ± 5.05a	—
	DE	66.67 ± 3.06b	-0.23 ± 0.005a	77.33 ± 2.08c	-0.171 ± 0.005c	67.36 ± 2.74c	-0.369 ± 0.015b
	GE	66.67 ± 5.77b	-0.231 ± 0.013a	84.67 ± 1.15b	-0.093 ± 0.005a	66.09 ± 4.7c	-0.382 ± 0.019b
	WE	56.67 ± 4.16c	-0.346 ± 0.02b	72 ± 3.46d	-0.228 ± 0.01d	56.84 ± 2.25d	-0.468 ± 0.025c
	VE	66 ± 4.36b	-0.238 ± 0.003a	82 ± 3.46b	-0.121 ± 0.006b	76.98 ± 4.81b	-0.275 ± 0.025a
Lunong 116	WT	96.67 ± 3.06a	—	98.67 ± 2.31a	—	119.51 ± 3.34a	—
	DE	65.33 ± 4.16c	-0.325 ± 0.025b	72.67 ± 2.52c	-0.265 ± 0.009c	73.61 ± 5.41c	-0.386 ± 0.005b
	GE	79.33 ± 5.77b	-0.179 ± 0.004a	88.67 ± 2.52b	-0.101 ± 0.01ab	92.64 ± 1.02b	-0.224 ± 0.013a
	WE	77.33 ± 3.06b	-0.2 ± 0.013a	89.33 ± 2.31b	-0.094 ± 0.005a	90.1 ± 2.73b	-0.246 ± 0.016a
	VE	77.33 ± 6.43b	-0.198 ± 0.002a	87.33 ± 2.52b	-0.114 ± 0.007b	93.67 ± 4.65b	-0.216 ± 0.012a
Kaimai 18	WT	96 ± 2a	—	100 ± 0a	—	113.43 ± 2.37a	—
	DE	86 ± 5.29b	-0.104 ± 0.011a	90.67 ± 2.31bc	-0.093 ± 0.023b	93.68 ± 0.76b	-0.172 ± 0.013a
	GE	86.67 ± 2.31b	-0.097 ± 0.008a	93.33 ± 3.06b	-0.067 ± 0.006a	92.38 ± 2.61b	-0.185 ± 0.013ab
	WE	80.67 ± 1.15b	-0.159 ± 0.009b	88 ± 3.46c	-0.12 ± 0.01c	88.67 ± 2.66b	-0.215 ± 0.007b
	VE	80.67 ± 3.06b	-0.159 ± 0.009b	88.67 ± 2.31c	-0.113 ± 0.006c	92.79 ± 1.45b	-0.182 ± 0.005ab
Zhengmai 366	WT	96.67 ± 3.06a	—	98.67 ± 2.31a	—	122.16 ± 5.14a	—
	DE	90 ± 2b	-0.069 ± 0.001a	94 ± 2b	-0.047 ± 0.006a	93.18 ± 2.48b	-0.239 ± 0.01a
	GE	76.67 ± 3.06c	-0.208 ± 0.005b	92.67 ± 2.31bc	-0.06 ± 0.002a	68.06 ± 2.96c	-0.442 ± 0.012bc
	WE	81.33 ± 4.16c	-0.158 ± 0.007b	88.67 ± 1.15c	-0.099 ± 0.009b	71.22 ± 1.33c	-0.415 ± 0.013b
	VE	69.33 ± 3.51d	-0.282 ± 0.021c	77.33 ± 3.06d	-0.216 ± 0.014c	66.38 ± 3.15c	-0.456 ± 0.013c
Wanmai 19	WT	34.67 ± 3.06a	—	83 ± 4.58a	—	33.09 ± 5.15a	—
	DE	33.33 ± 4.16a	-0.031 ± 0.018a	58.67 ± 3.21b	-0.253 ± 0.019b	28.68 ± 4.02a	-0.131 ± 0.004a
	GE	19.33 ± 5.03b	-0.434 ± 0.176b	60 ± 2b	-0.234 ± 0.006a	22.1 ± 3.52b	-0.316 ± 0.007b
	WE	10 ± 2c	-0.711 ± 0.056d	32.67 ± 2.31d	-0.583 ± 0.03d	12.05 ± 4.01c	-0.627 ± 0.024d
	VE	12 ± 3c	-0.65 ± 0.106c	42.67 ± 2.52c	-0.46 ± 0.023c	17.75 ± 1.81b	-0.444 ± 0.031c
Wankenmai 1	WT	68.67 ± 5.77a	—	88 ± 2a	—	72.56 ± 1.78a	—
	DE	55.33 ± 4.16b	-0.193 ± 0.007a	76 ± 2b	-0.136 ± 0.002a	51.36 ± 3.29b	-0.291 ± 0.015a
	GE	44 ± 2c	-0.356 ± 0.012c	69.33 ± 3.06c	-0.212 ± 0.006c	38.64 ± 1.47cd	-0.464 ± 0.033c
	WE	39.33 ± 4.04c	-0.43 ± 0.012d	62.67 ± 2.89d	-0.289 ± 0.008d	35.28 ± 0.7d	-0.512 ± 0.044d
	VE	50 ± 2b	-0.273 ± 0.019b	73.33 ± 2.08bc	-0.165 ± 0.005b	43.14 ± 1.25c	-0.407 ± 0.014b
Wanmai 50	WT	98.67 ± 2.31a	—	99.33 ± 1.15a	—	120.18 ± 1.44a	—
	DE	86.67 ± 4.16b	-0.121 ± 0.008a	90.67 ± 4.62b	-0.087 ± 0.006a	97.63 ± 4.09b	-0.187 ± 0.005a
	GE	83.33 ± 2.08bc	-0.155 ± 0.005b	87.33 ± 3.06bc	-0.121 ± 0.006b	78.79 ± 2.02d	-0.345 ± 0.025c
	WE	69.33 ± 2.31d	-0.297 ± 0.011d	80.67 ± 2.08d	-0.188 ± 0.01d	75.32 ± 4.48d	-0.374 ± 0.031c
	VE	79.33 ± 4.62c	-0.195 ± 0.015c	84.67 ± 1.53cd	-0.147 ± 0.006c	86.31 ± 5.78c	-0.281 ± 0.011b
Wanmai 54	WT	98 ± 2a	—	98.67 ± 1.15a	—	118.55 ± 3.14a	—
	DE	80 ± 2b	-0.184 ± 0.004ab	86 ± 2b	-0.128 ± 0.005b	89.58 ± 6.5b	-0.243 ± 0.01a
	GE	82 ± 2.65b	-0.164 ± 0.01a	86 ± 4.36b	-0.129 ± 0.004b	84.6 ± 8.33b	-0.286 ± 0.006b
	WE	80 ± 2b	-0.184 ± 0.009ab	88.67 ± 2.31b	-0.101 ± 0.001a	84.63 ± 1.37b	-0.287 ± 0.083b
	VE	78.67 ± 3.06b	-0.197 ± 0.015b	86.67 ± 4.16b	-0.122 ± 0.002b	77.73 ± 5.2c	-0.344 ± 0.015c
F-value	C	507.03**	412.11**	288.49**	1050.66**	837.62**	273.22**
	E	164.03**	916.08**	208.61**	397.18**	478.93**	194.29**
	C×E	14.68**	485.83**	35.53**	218.64**	38.21**	78.58**

Notes: WT, DE, GE, WE, VE mean distilled water, *Descurainia sophia* extract, *Galium tricorne* extract, *Wild oat* extract and *Vicia sativa* extract respectively. C and E represent variety and extract treatment, respectively. RI mean allelopathy indicator. Different letters after the same column value indicate significant differences among different extractions of the same wheat variety at 0.05 level. * and ** mean significant effects at 0.05 and 0.01 levels, respectively

Wheat seedling growth: Table 2 shows that the seedling height and root length of *Yannong19*, *Jimai22*, *Kaimai18*, *Zhengmai366*, and *Wanmai54* were all above 150 cm, with *Kaimai18* and *Yannong21* having the highest seedling height and root length, respectively. The variety with the lowest seedling height and root length was *Wanmai19*, and the differences between the varieties were 182.23% and 153.25%. The four weed extracts significantly reduced the seedling height and root length of wheat. On the basis of the average effect of weed extracts on the allelopathic action of wheat, the seedling height and root length of *Vicia sativa* extract decreased by 92.98% and 88.99%, respectively, and the allelopathic indicators reached -0.91 and -0.87 , respectively, indicating significant allelopathic inhibition. The seedling height and root length of *Galium tricornis* extract decreased by 73.32% and 72.16%, respectively, and the allelopathic indicators reached -0.70 and -0.70 , respectively, indicating low allelopathic inhibition. The *Descurainia sophia* extract had the greatest inhibition on the seedling height and root length of *Kaimai18* and *Wanmai50*, respectively. The least affected varieties were *Lunong116* and *Wanmai19*. The *Galium tricornis* extract had the greatest inhibition on the seedling height and root length of *Yannong19* and *Wankenmai1*, respectively. The least affected varieties were *Lunong116* and *Wanmai19*. The *Wild oat* extract had the greatest inhibition on the seedling height and root length of *Zhengmai366* and *Wanmai54*, respectively. The least affected variety was *Wanmai19*. The *Vicia sativa* extract had the greatest inhibition on the seedling height and root length of *Zhengmai366*. The least affected variety was *Wanmai19*. Given the difference of weed species, different weed extracts had different allelopathic inhibitory effects on seedling growth of different wheat varieties.

Cluster analysis of allelopathic effects of different wheat varieties in response to weed extracts at seedling stage: The absolute value of the allelopathic index indicates the strength of the allelopathic effect. Thus, cluster analysis was used to analyze the inhibitory effects of the four weed extracts on the seedling growth indicators (seedling height and root length) on the basis of the Euclidean distance and square sum of deviations. Then, the wheat varieties were classified (Fig. 1). According to the clustering map of wheat varieties based on the allelopathic effect of *Descurainia sophia* extract, the wheat varieties were divided into two categories: one included eight varieties with strong allelopathic inhibition rates, namely, *Yannong19*, *Zhengmai366*, and *Jimai22*; the other included two varieties with low allelopathic inhibition rates, namely, *Wanmai19* and *Lunong116* (Fig. 1-A). According to the clustering map of wheat varieties based on the allelopathic effect of *Galium tricornis* extract, the wheat varieties were divided into two categories: one included six varieties with strong allelopathic inhibition rates, namely, *Yannong19*, *Wankenmai1*, and *Wanmai54*; the other included four varieties with low allelopathic inhibition rates, namely, *Wanmai19*, *Lunong116*, *Yannong21*, and *Wanmai50* (Fig.

1-B). According to the clustering map of wheat varieties based on the allelopathic effect of *Wild oat* extract, the wheat varieties were divided into two categories: one included four varieties with strong allelopathic inhibition rates, namely, *Yannong19*, *Zhengmai366*, *Wanmai54*, and *Jimai22*; the other included six varieties with low allelopathic inhibition rates, namely, *Wanmai19*, *Yannong21*, and *Wankenmai1* (Fig. 1-C). According to the clustering map of wheat varieties based on the allelopathic effect of *Vicia sativa* extract, the wheat varieties were divided into three categories: one included six varieties with strong allelopathic inhibition rates, namely, *Yannong19*, *Wanmai54*, and *Jimai22*; the second included three varieties with low allelopathic inhibition rates, namely, *Wankenmai1*, *Yannong21*, and *Lunong116*; the third included the variety with the weakest allelopathic rate, *Wanmai19* (Fig. 1-D). In summary, all four weed extracts had strong allelopathic inhibition rates on *Yannong19*, and the allelopathic inhibition rates on *Wanmai19* were weak. Therefore, when measuring physiological indicators, *Yannong19* and *Wanmai19* were used as the representative varieties.

Wheat root activity: Fig. 2 shows that the four weed extracts significantly reduced the root activity of wheat, but the *Wild oat* and *Vicia sativa* extracts had great inhibitory effect on the root activity of wheat, which decreased by 65.69% and 67.59% compared with the control. The *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts decreased the root activity of *Yannong19* by 60.82%, 62.68%, 72.39%, and 79.88%, respectively, and that of *Wanmai19* by 42.16%, 37.73%, 56.10%, and 50.00%, respectively. Therefore, the weed extracts had strong inhibition on the root activity of *Yannong19*.

Physiological indicators of wheat leaves: Fig. 3 shows that the four weed extracts significantly reduced the SOD, POD, and CAT activities in wheat leaves, especially the extracts of *Wild oat* and *Vicia sativa*. Compared with the control, the *Wild oat* extract reduced the SOD, POD, and CAT activities by 84.61%, 79.93%, and 76.77%, respectively. The *Vicia sativa* extract reduced the SOD, POD, and CAT activities by 84.12%, 79.29%, and 76.98%, respectively. However, the effects of *Wild oat* and *Vicia sativa* extracts were not significant. The *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts decreased the SOD activity of *Yannong19* by 88.87%, 91.57%, 95.14%, and 96.19%, respectively, and that of *Wanmai19* by 51.76%, 33.02%, 67.27%, and 64.25%, respectively. The *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts decreased the POD activity of *Yannong19* by 82.19%, 86.51%, 92.23%, and 93.91%, respectively, and that of *Wanmai19* by 52.21%, 33.47%, 67.71%, and 64.02%, respectively. The *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts decreased the CAT activity of *Yannong19* by 66.61%, 74.71%, 85.43%, and 88.58%, respectively, and that of *Wanmai19* by 52.36%, 34.21%, 66.79%, and 63.65%. Therefore, the weed extract had strong inhibitory effects on the SOD, POD, and CAT activities of *Yannong19*.

Table 2. Effect of weed extract on the seedlings of wheat.

Variety	Extract	Seedling height		Root length	
		Result (mm)	RI	Result (mm)	RI
Yannong 19	WT	153.79 ± 5.72a	—	153.89 ± 5.32a	—
	DE	10.27 ± 0.45b	-0.932 ± 0.052a	21.7 ± 1.1b	-0.859 ± 0.021a
	GE	7.78 ± 0.93bc	-0.95 ± 0.033a	20.68 ± 0.57b	-0.863 ± 0.029a
	WE	4.48 ± 0.29cd	-0.971 ± 0.018a	15.3 ± 0.83c	-0.901 ± 0.04ab
	VE	3.51 ± 0.32d	-0.977 ± 0.005a	11.14 ± 0.23d	-0.928 ± 0.025b
Yangnong 21	WT	141.97 ± 4.95a	—	163.32 ± 10.34a	—
	DE	33.16 ± 1.69d	-0.77 ± 0.017c	33.69 ± 0.91d	-0.794 ± 0.034c
	GE	69.86 ± 2.42b	-0.51 ± 0.013a	68.62 ± 3.84b	-0.582 ± 0.015a
	WE	41.26 ± 2.56c	-0.712 ± 0.04b	45.52 ± 2.4c	-0.722 ± 0.026b
	VE	22 ± 0.71e	-0.846 ± 0.019d	20.98 ± 0.17e	-0.87 ± 0.028d
Jimai 22	WT	157.19 ± 3.53a	—	160.23 ± 4.96a	—
	DE	13.15 ± 1.75b	-0.917 ± 0.022a	21.99 ± 0.41c	-0.865 ± 0.013b
	GE	14.09 ± 0.31b	-0.911 ± 0.031a	69.67 ± 3.05b	-0.56 ± 0.041a
	WE	10.18 ± 0.83b	-0.939 ± 0.016ab	22.78 ± 0.81c	-0.857 ± 0.02b
	VE	4.81 ± 0.39c	-0.969 ± 0.016b	13.25 ± 0.54d	-0.917 ± 0.026c
Lunong 116	WT	132.78 ± 7.44a	—	137.57 ± 1.49a	—
	DE	74.22 ± 3.31c	-0.435 ± 0.038b	59.55 ± 1b	-0.567 ± 0.005a
	GE	125.85 ± 6.75b	-0.056 ± 0.011a	56.86 ± 2.86b	-0.587 ± 0.028a
	WE	21.79 ± 0.99d	-0.837 ± 0.019c	35.7 ± 1.84c	-0.741 ± 0.041b
	VE	20.43 ± 0.8d	-0.846 ± 0.052c	28.46 ± 2.08d	-0.793 ± 0.039c
Kaimai 18	WT	158.23 ± 1.64a	—	159.07 ± 3.9a	—
	DE	6.36 ± 0.46c	-0.96 ± 0.019b	21.38 ± 0.67c	-0.866 ± 0.043b
	GE	23.9 ± 0.55b	-0.848 ± 0.028a	41.03 ± 1.14b	-0.743 ± 0.016a
	WE	25.46 ± 2.29b	-0.839 ± 0.038a	38.9 ± 1.83b	-0.756 ± 0.029a
	VE	3.7 ± 0.43c	-0.977 ± 0.003b	13.17 ± 1.01d	-0.917 ± 0.012c
Zhengmai 366	WT	153.68 ± 3.9a	—	162.29 ± 1.91a	—
	DE	11.17 ± 0.48b	-0.927 ± 0.01a	25.29 ± 1.09b	-0.844 ± 0.01a
	GE	10.57 ± 0.75b	-0.93 ± 0.021a	27.6 ± 1.2b	-0.827 ± 0.031a
	WE	3.72 ± 0.19c	-0.976 ± 0.009ab	17.03 ± 1.27c	-0.895 ± 0.009b
	VE	1.62 ± 0.15c	-0.989 ± 0.001b	7.97 ± 0.45d	-0.951 ± 0.007c
Wanmai 19	WT	56.06 ± 1.39a	—	64.49 ± 5.55a	—
	DE	27.04 ± 1.27c	-0.535 ± 0.011b	27.64 ± 0.9c	-0.562 ± 0.015c
	GE	37.55 ± 2.26b	-0.327 ± 0.008a	34.42 ± 1.91b	-0.429 ± 0.018a
	WE	18.35 ± 1.37d	-0.679 ± 0.03c	31.97 ± 1.66b	-0.504 ± 0.04b
	VE	20.04 ± 1.34d	-0.632 ± 0.042c	22.36 ± 1.11d	-0.643 ± 0.024d
Wankenmai 1	WT	98.43 ± 1.23a	—	115.94 ± 1.27a	—
	DE	6.62 ± 0.54b	-0.933 ± 0.05a	11.8 ± 0.77d	-0.896 ± 0.021b
	GE	6.58 ± 0.06b	-0.934 ± 0.051a	15.89 ± 2.52cd	-0.864 ± 0.019b
	WE	10.08 ± 1.12b	-0.898 ± 0.044a	25.29 ± 0.59b	-0.777 ± 0.044a
	VE	6.04 ± 0.42b	-0.939 ± 0.024a	16.95 ± 1.35c	-0.85 ± 0.03b
Wanmai 50	WT	111.68 ± 3.44a	—	155.02 ± 2.67a	—
	DE	6.78 ± 0.31d	-0.94 ± 0.035c	16.02 ± 1.24d	-0.896 ± 0.042c
	GE	39.09 ± 2.01b	-0.646 ± 0.014a	41.05 ± 1.16b	-0.735 ± 0.031a
	WE	15.73 ± 0.27c	-0.859 ± 0.05b	27.81 ± 1.2c	-0.821 ± 0.025b
	VE	4.97 ± 0.5d	-0.955 ± 0.014c	12.86 ± 0.73d	-0.917 ± 0.015c
Wanmai 54	WT	152.94 ± 2.29a	—	160.66 ± 4.66a	—
	DE	11.98 ± 0.44c	-0.922 ± 0.016ab	21.81 ± 0.59b	-0.864 ± 0.017ab
	GE	16.07 ± 1.34b	-0.896 ± 0.054a	23.05 ± 1.27b	-0.855 ± 0.042a
	WE	8.73 ± 0.47cd	-0.943 ± 0.027ab	14.79 ± 0.38c	-0.907 ± 0.029bc
	VE	5.32 ± 0.45d	-0.965 ± 0.01b	10.57 ± 0.88d	-0.934 ± 0.005c
F-value	C	613.2**	378.95**	237.29**	202.64**
	E	14414.95**	278.38**	12588.84**	185.48**
	C×E	575.48**	77.93**	265.52**	24.18**

Notes: WT, DE, GE, WE, VE mean distilled water, *Descurainia sophia* extract, *Galium tricornis* extract, *Wild oat* extract and *Vicia sativa* extract respectively. C and E represent variety and extract treatment, respectively. RI mean allelopathy indicator. Different letters after the same column value indicate significant differences among different extractions of the same wheat variety at 0.05 level. * and ** mean significant effects at 0.05 and 0.01 levels, respectively

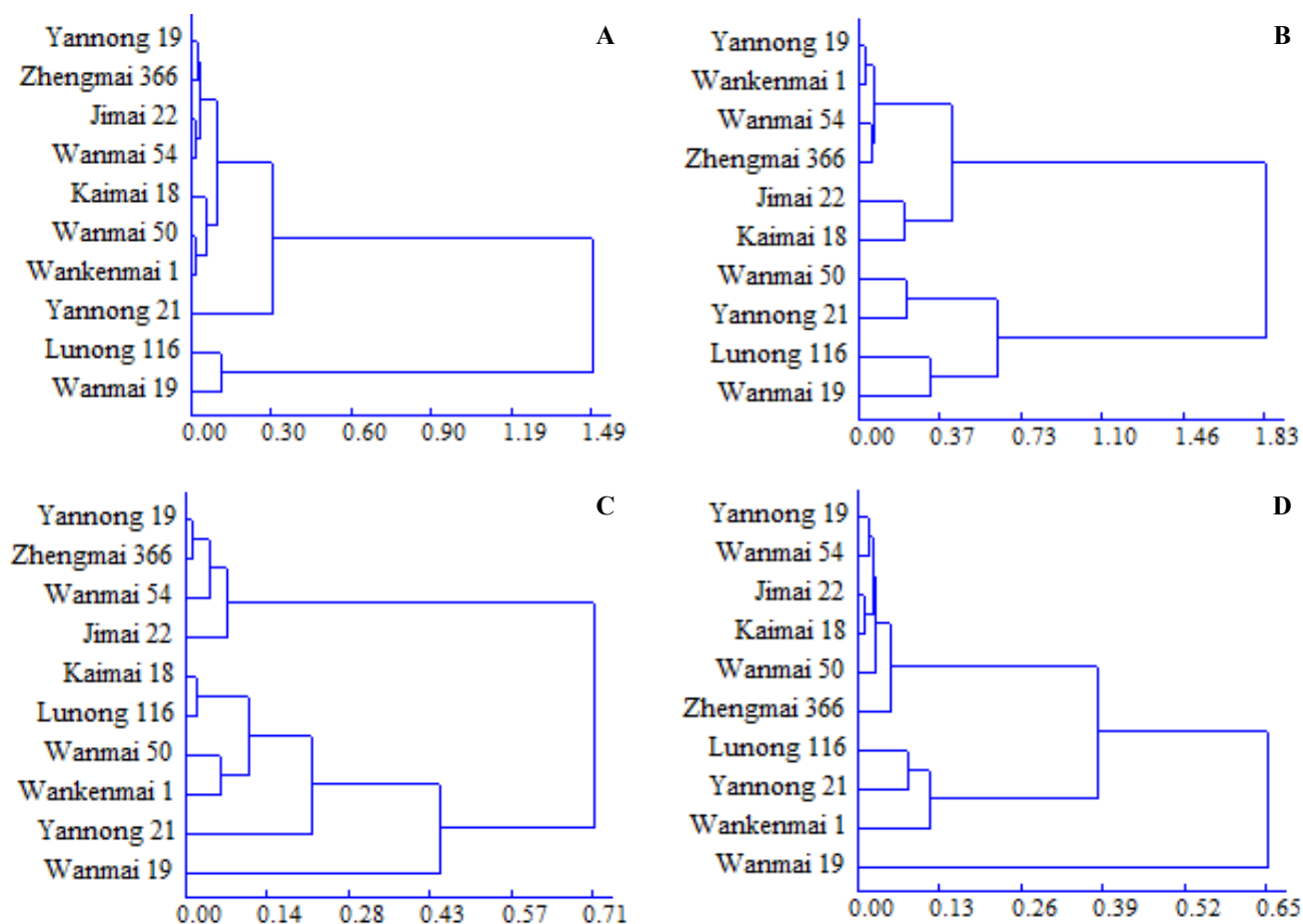


Fig. 1. Clustering map of allelopathic effects of different wheat varieties in response to weed extracts at seedling stage.

Note: A, B, C, and D are the clustering maps of wheat varieties based on the allelopathic effect of *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts, respectively.

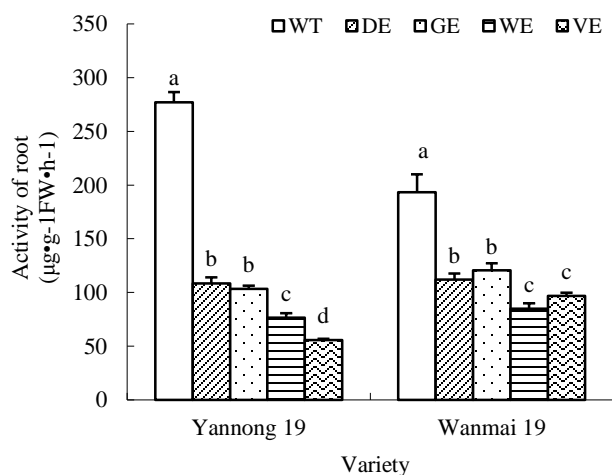


Fig. 2. Effect of weed extracts on the activity of root.

Fig. 3 shows that the four weed extracts significantly increased the MDA content in wheat leaves. The *Wild oat* and *Vicia sativa* extracts had great inhibitory effects on MDA content in leaves, which increased by 74.67% and 74.01%, respectively compared with the control. The *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts increased the MDA content of *Yannong19* by 51.82%, 79.45%, 106.05%, and 122.63%, respectively, and that of *Wanmai19* by 19.18%, 9.11%, 48.84%, and

34.00%, respectively. Therefore, the weed extracts remarkably increased the MDA content of *Yannong19*.

Allelopathic effect analysis of different weed extracts on the physiological indicators of wheat seedlings:

Table 3 shows that the weed extracts had an allelopathic inhibitory effect on the root activity and activities of SOD, POD, and CAT and promoted the MDA content in leaves allelopathically. The allelopathic effect of *Descurainia sophia* and *Galium tricornis* extracts was weaker than that of *Wild oat* and *Vicia sativa* extracts. The *Vicia sativa* extract had the strongest allelopathic effect on *Yannong19*, whereas the *Descurainia sophia* extract had the weakest effect. The *Wild oat* extract had the strongest allelopathic effect on *Wanmai19*, whereas the *Galium tricornis* extract had the weakest effect. In general, the allelopathic effect of weed extracts on *Yannong19* was greater than that on *Wanmai19*.

Correlation analysis of allelopathic indicators between wheat seedling growth indicators and internal physiological indicators:

The seedling height and root length of wheat seedlings were significantly positively correlated with root activity and SOD, POD, and CAT activities in leaves and significantly negatively correlated with MDA content. The correlation between seedling height and physiological indicators was slightly greater than that between seedling height and root length (Table 4).

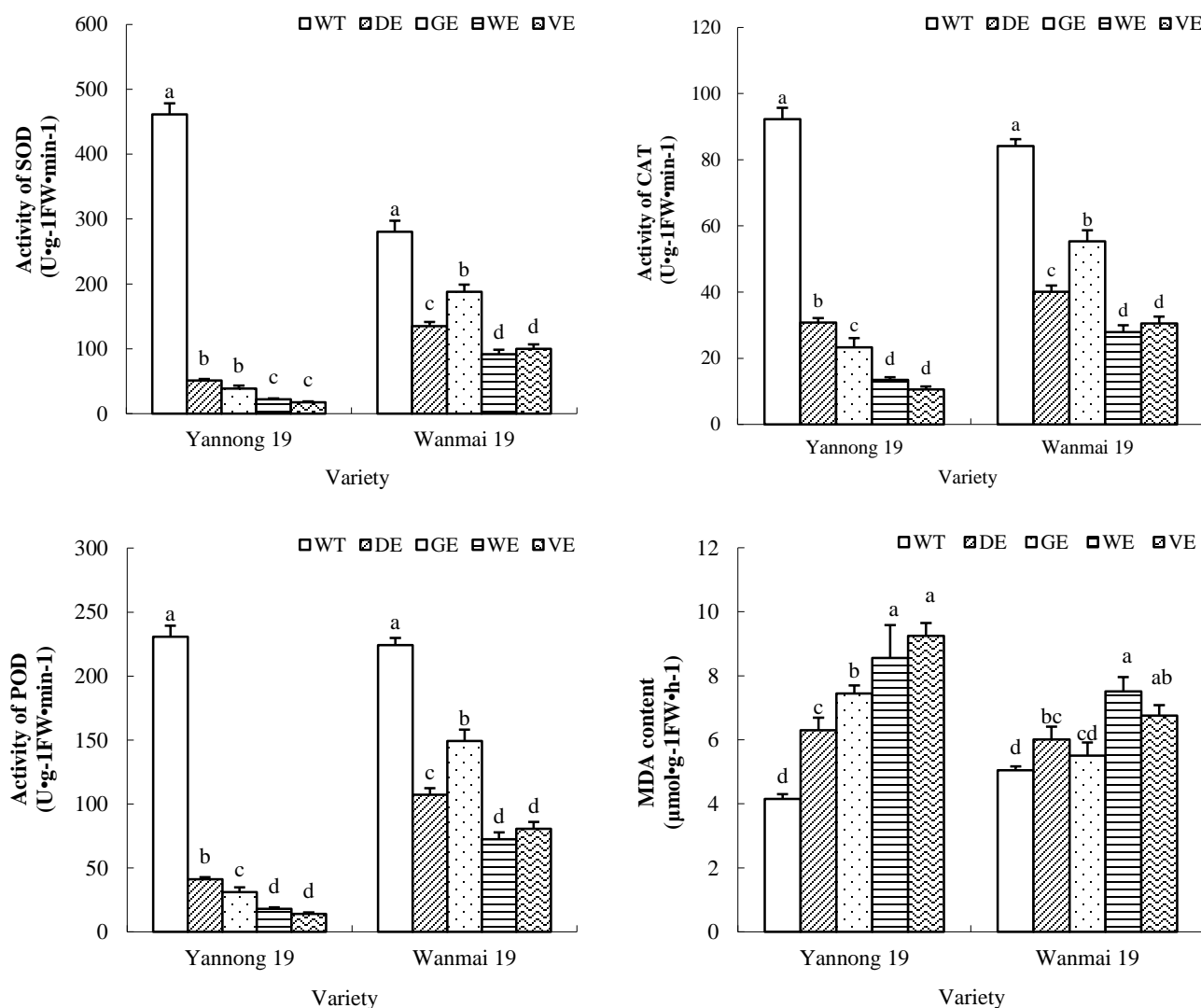


Fig. 3. Effect of weed extracts on the physiological indexes of wheat leaves.

Table 3. Allelopathic effect of weed extract on the physiological indexes of wheat.

Variety	Extract	Activity of root ($\mu\text{g}\cdot\text{g}^{-1}\text{FW}\cdot\text{h}^{-1}$)	Activity of SOD ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	Activity of POD ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	Activity of CAT ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	MDA content ($\mu\text{mol}\cdot\text{g}^{-1}\text{FW}\cdot\text{h}^{-1}$)
Yannong 19	DE	$-0.608 \pm 0.009\text{a}$	$-0.888 \pm 0.009\text{a}$	$-0.822 \pm 0.015\text{a}$	$-0.665 \pm 0.028\text{a}$	$0.517 \pm 0.038\text{c}$
	GE	$-0.627 \pm 0.003\text{a}$	$-0.916 \pm 0.008\text{ab}$	$-0.865 \pm 0.013\text{b}$	$-0.747 \pm 0.025\text{b}$	$0.797 \pm 0.12\text{b}$
	WE	$-0.724 \pm 0.02\text{b}$	$-0.951 \pm 0.005\text{bc}$	$-0.922 \pm 0.008\text{c}$	$-0.854 \pm 0.015\text{c}$	$1.059 \pm 0.204\text{a}$
	VE	$-0.799 \pm 0.004\text{c}$	$-0.962 \pm 0.003\text{c}$	$-0.939 \pm 0.006\text{c}$	$-0.886 \pm 0.01\text{c}$	$1.231 \pm 0.185\text{a}$
Wanmai 19	DE	$-0.42 \pm 0.045\text{a}$	$-0.517 \pm 0.033\text{b}$	$-0.517 \pm 0.033\text{b}$	$-0.517 \pm 0.033\text{b}$	$0.193 \pm 0.105\text{bc}$
	GE	$-0.376 \pm 0.019\text{a}$	$-0.33 \pm 0.032\text{a}$	$-0.33 \pm 0.032\text{a}$	$-0.33 \pm 0.032\text{a}$	$0.09 \pm 0.06\text{c}$
	WE	$-0.558 \pm 0.056\text{c}$	$-0.673 \pm 0.018\text{c}$	$-0.673 \pm 0.018\text{c}$	$-0.673 \pm 0.018\text{c}$	$0.488 \pm 0.07\text{a}$
	VE	$-0.498 \pm 0.03\text{b}$	$-0.642 \pm 0.032\text{c}$	$-0.642 \pm 0.032\text{c}$	$-0.642 \pm 0.032\text{c}$	$0.341 \pm 0.091\text{ab}$

Table 4. Correlation of allelopathic indicators between wheat seedling growth indicators and internal physiological indicators.

Index	Activity of root ($\mu\text{g}\cdot\text{g}^{-1}\text{FW}\cdot\text{h}^{-1}$)	Activity of SOD ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	Activity of POD ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	Activity of CAT ($\text{U}\cdot\text{g}^{-1}\text{FW}\cdot\text{min}^{-1}$)	MDA content ($\mu\text{mol}\cdot\text{g}^{-1}\text{FW}\cdot\text{h}^{-1}$)
Seedling height	0.9232**	0.9984**	0.991**	0.9246**	-0.8778**
Root length	0.8829**	0.9454**	0.9319**	0.8496**	-0.859**

Discussion

Effects of weed extracts on wheat seed germination and seedling growth: The most common allelochemicals in plant extracts include organic acids, phenols, and terpenoids. Organic acids, especially phenolic acids, are the most important allelochemicals (Qasem & Issa, 2018; Zavallonia *et al.*, 2011; Wu *et al.*, 2008). Many studies have shown that low concentrations of phenolic acids can promote crop growth, whereas high concentrations will inhibit it (Liu *et al.*, 2016). The present study found that different weed extracts had different allelopathic effects on seed germination of different wheat varieties because of the difference of weed species, which was consistent with the findings of Zhang *et al.*, 2015b on *Descurainia sophia* and *Galium tricornis*. Zhang *et al.*, 2015b argued that the total phenolic acid content in crop straw extracts was significantly or extremely significantly negatively correlated with related indicators of wheat seed germination (germination rates, germination potential, and germination indicators) and seedling growth indicators (root fresh weight) and that the correlation between the total phenolic acid content and fresh weight on the ground did not reach a significant level (Zhang *et al.*, 2015b). Therefore, the difference in allelopathic inhibition of different weed extracts may be caused by the difference in the content of phenolic acid in different weed extracts.

On the basis of the results of seed germination, the *Descurainia sophia*, *Galium tricornis*, *Wild oat*, and *Vicia sativa* extracts had allelopathic inhibition on wheat seed germination. The seed germination rate decreased by 12.64%–18.52%, and the allelopathic index of wheat seed germination rate was –0.12 to –0.19. The *Wild oat* extract had the strongest inhibitory effect, whereas the *Descurainia sophia* extract had the weakest inhibitory effect. On the basis of the seedling growth, the four weed extracts significantly reduced the seedling height and root length of wheat. The wheat seedling height decreased by 73.32%–92.98%, and the allelopathic index of seedling height was –0.70 to –0.87. The root length was decreased by 72.16%–88.99%, and the allelopathic index of root length was –0.70 to –0.91. The *Vicia sativa* extract had the strongest inhibitory effect, whereas the *Galium tricornis* extract had the weakest inhibitory effect. Thus, the inhibitory effect of weed extract on seedling growth was much greater than its inhibition on seed germination, indicating that weed extract had a strong inhibitory effect on wheat growth. Therefore, the weeds in the wheat field should be eliminated as early as possible. Otherwise, the remaining weed residues can have a strong inhibitory effect on the growth of wheat in the field. Moreover, the plant residues falling from the weed plants that have not been eliminated completely exert a strong inhibitory effect on the growth of wheat.

In this study, the allelopathic inhibition of weed extracts on wheat varieties was extremely significant. All four weed extracts had strong allelopathic inhibition rates on *Yannong19*; however, the allelopathic inhibition rates on *Wanmai19* were weak. Therefore, *Wanmai19* has good grass resistance potential. The allelopathic resistance of

different wheat varieties to weeds was different, which may be caused by genetic differences among wheat varieties, and this difference is also the theoretical premise for identifying wheat varieties resistant to weeds.

Analysis of internal physiological mechanism of weed extract on wheat: The mechanism responsible for the allelopathic effect of plants is complex. An important reason is that the allelopathic action of plants alters the physiological activities or material transport of cells by enhancing or weakening the permeability of organelle membranes, which in turn causes the change of a series of physiological metabolic processes such as cell division, photosynthesis, protein, and chlorophyll synthesis, thus affecting the growth of seedlings (Cirillo *et al.*, 2018; Khattak *et al.*, 2015; Ge *et al.*, 2015; Daniel & Oliver, 2004). MDA is a cytotoxic substance. The increase of MDA content indicates that the cell membrane of the plant is damaged, and the membrane permeability is increased. SOD, POD, and CAT are common antioxidant enzymes in plants. Their contents and activities reflect the scavenging ability of oxygen free radicals and disproportionation reaction of H₂O₂ in vivo (Yin *et al.*, 2018). In this study, all four weed extracts significantly reduced the root activity of wheat and the activities of SOD, POD, and CAT in leaves and increased the MDA content in leaves. The seedling height and root length of wheat seedlings were extremely significantly positively correlated with root activity and activities of SOD, POD, and CAT and extremely significantly negatively correlated with MDA content. Thus, the main reason for the inhibitory effect of weed extracts on the growth of wheat seedlings was that they damaged the cell membrane and reduced the activity of antioxidant enzymes, which in turn affected the growth of wheat. The effect of *Wild oat* and *Vicia sativa* extracts on the physiological indicators of seedlings was greater than that of *Descurainia sophia* and *Galium tricornis* extracts. So the *Wild oat* and *Vicia sativa* extracts also had greater effect on seedling growth index, root length, and seedling length than *Descurainia sophia* and *Galium tricornis* extracts. This finding indicated that the effect of allelochemicals on plant cell membrane was the root cause of inhibited plant growth. In this study, only the effects of weed extracts on plant cell membrane were studied. The changes of physiological metabolism processes such as leaf photosynthesis and synthesis of protein and chlorophyll caused by cell membrane permeability change remain to be further studied.

Conclusions

In this study, the weed extracts had certain differences in the allelopathic inhibition on wheat seed germination and seedling growth. The *Wild oat* extract had the strongest inhibitory effect on seed germination, whereas the *Descurainia sophia* extract had the weakest effect. The *Vicia sativa* extract had the strongest allelopathic effect on the growth of seedlings, whereas the *Galium tricornis* extract had the weakest effect. All weed extracts had an allelopathic inhibitory effect on the root activity and activities of SOD, POD, and CAT, and they could promote the MDA content in leaves allelopathically.

The effect of extracts on internal physiological indicators was an important reason for inhibiting wheat growth. According to the cluster analysis of the allelopathic effect of allelopathic indicators, the allelopathic inhibition rate of *Wanmai19* was weak, and its resistance to grass was strong, which has great application value.

Acknowledgments

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