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酸雨对3类不同抗性种子萌发过程的影响Ⅵ
——酸雨胁迫对3类种子萌发POD活性的影响

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摘 要：试验采用不同pH值的模拟酸雨处理3类不同抗性种子，测定了酸雨胁迫对种子萌发过程膜脂过氧化及POD活性的影响。结果表明，萌发7 d时,3类种子POD活性随胁迫强度增加而先升后降;丙二醛(MDA)含量升高,且处理组高于CK;3类种子对酸雨胁迫的抵抗能力为水稻>小麦>油菜。考察3~7 d的动态变化时发现,不同种子,不同处理条件下,各项生理生化指标的动态变化曲线不尽一致,但随着时间的推移,POD活性均有不同程度的增加，MDA含量变化也表现相应的增加趋势,同时也反映出相应的剂量效应和防护效果。

关键词：酸雨；水稻；小麦；油菜；萌发；POD

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Effects of Acid Rain on Activity of Peroxidase (POD) in Seed Germination of Three Plants Under Time Stress

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Abstract: To study responses of seed germination of 3 kinds of seeds on acid rain stress, rice (O. *sativa*), wheat (T. *aestivum*) and rape (B.*chinensis var. oleifera*) seeds were used as experimental materials, in our test. The purpose was to understand the effects of acid rain on seed germination of various acid-fast plants in acidic deposition area and to determine reasons of growth and decline dynamics of crop population and its yield change. In the meantime, seven simulated acid rain (SAR) solutions of pH 2.0, 2.5, 3.0, 3.5, 4.0, 4.5 and 5.0 and neutral solution (pH6.5) as control group (CK) were used. Pretreatment seeds for the three species were placed in culture dishes with filter sheets to germinate in a culture container kept at a constant temperature of 25℃. Each treatment group involved in three dishes and each dish received 50 seeds. During germination, seeds were exposed to simulated acid rain freshly prepared everyday. The germination indexes (activity of POD, content of MDA) were all studied under acid rain stress in both experiments. The results showed that at the 7th day, the activity of POD of the three seeds increased at lower acidity levels, and then decreased at higher acidity levels, but the content of MDA increased always along with the increase of the stress strength. In this time, the second index of different acidity level was higher than that of CK. Compared with CK, the amplitude of the content of MDA was found: rice (13.4%) <wheat (213.7%) < rape (324.5%). The responses on acid rain stress of activity for POD and content of MDA of the three species indicated that there was a acidity level significant difference of each index at $P<0.05$ at the 7th day,i.e rice (no significant difference)>wheat (3.0)>rape(4.0). The experiment data further evidenced that plants could display different sensitivity under the acid rain stress, in which rice had stronger resistance than wheat, and rape was more susceptible to acid rain than rice and wheat. During 3~7 d, the results proved that the changing curves of each physiological topics were not identical. However, as time went on, all of the POD activity increased with different degrees, and the content of MDA also showed the same increase trend. On the other hand, the protective effect was related with acidity levels. Namely, at the same stress time, POD activities of all control groups of rape and wheat seeds were higher than those of CK, and increased along with the acidity levels. But POD activities of rice seed increased at the lower levels,and then decreased at higher levels.However, the content of MDA of three seeds increased always.It may be concluded that the response of the plants tested in the study was in an order with rice >wheat>rape.

Keywords: acid rain; rice; wheat; rape; germination; POD

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前文[1~3]报道了酸雨对水稻与小麦种子萌发的影响,获得了酸雨影响种子萌发的若干信息。本文进一步研究萌发过程中酸雨胁迫对水稻、小麦和油菜种子 POD 活性的影响，旨在进一步了解 3 类植物种子萌发响应酸雨胁迫的内在原因。

1 材料和方法

1.1 材料处理方法

模拟酸雨配置方法与种子萌发过程同参考文献[1]~[3]。

1.2 指标测定

静态指标测定于酸雨胁迫 7 d 后进行,POD 活性、丙二醛(MDA)含量的测定分别参照文献[4]、[5]。动态指标于酸雨胁迫后 3、4、5、6、7 d 进行。

2 结果与分析

2.1 种子萌发响应酸雨胁迫的即时效应

POD 是植物体内活性氧清除酶系，可防止细胞内过量自由基对生物大分子与质膜破坏。POD 也可在逆境或衰老后期表达为伤害效应,即参与活性氧生成,引发膜脂过氧化,乃植株衰老到一定阶段产物,抑或作为一种衰老指标 [6]。MDA 是细胞膜脂过氧化产物，表征植物受逆境伤害程度。表 1 显示，随着 pH（2.0~5.0）上升,3 类种子 POD 活性先应激升高后降低,且呈显著相关。经分析得回归方程：

水稻 $y=-2.452x^2+31.7x+4.011$, $r=0.959\,4$

小麦 $y=-2.280\,4x^2+34.251x-16.791$, $r=0.781\,3$

油菜 $y=1.612\,8x^3-25.497x^2+118.06x-32.551$, $r=0.764\,1$

3 类种子 MDA 含量随 pH 上升而降低,与 pH 呈显著负相关($\alpha=0.05$, $r_{水稻}=0.959\,9$；$\alpha=0.05$, $r_{小麦}=0.964\,8$；$\alpha=0.05$, $r_{油菜}=0.937\,4$),但水稻并未达到差异显著水平。表明高 pH 时,种子虽已产生应激反应,但不足以抵抗活性氧触发的膜脂过氧化增大，导致 MDA 增加。随 pH 降低,植物细胞发生实质损伤,其酶活性会不可逆降低,MDA 升高。由表 1 可知,所有处理组的 MDA 含量都高于 CK，且与 CK 相比,MDA 变幅为水稻(13.4%)<小麦(213.7%)<油菜(324.5%),表明种子萌发过程中，对酸雨胁迫的抵抗能力为水稻>小麦>油菜。pH2.5~5.0 范围内,相互间达到差异显著的 pH

表 1 酸雨胁迫对 3 类种子 MDA 含量及 POD 活性的影响

Table 1 Effects of acid rain on contents of MDA and POD activity of the three seeds

	pH	MDA/mmol・g-1	相对值	POD/△A470/min・g-1	相对值
Ⅰ	CK	1.339a	100.0	0.167ab	100.0
	2.0	1.518a	113.4	0.046e	27.5
	2.5	1.493a	111.5	0.114d	68.3
	3.0	1.483a	110.8	0.128cd	76.6
	3.5	1.473a	110.1	0.146bc	87.4
	4.0	1.378a	103.0	0.161ab	96.4
	4.5	1.366a	102.0	0.184a	110.2
	5.0	1.343a	100.4	0.178a	106.6
Ⅱ	CK	0.627a	100.0	0.826a	100.0
	2.0	1.968d	313.7	0.246f	29.8
	2.5	1.594a	254.2	0.311f	37.7
	3.0	1.549bc	246.9	0.362e	43.8
	3.5	1.292b	206.0	0.519d	62.8
	4.0	1.258ab	200.6	1.081c	130.9
	4.5	1.217ab	194.0	0.946b	114.5
	5.0	1.005ab	160.3	0.942b	114.0
Ⅲ	CK	0.580a	100.0	0.141a	100.0
	2.0	2.464f	424.5	0.294g	50.26
	2.5	2.443f	420.8	0.199f	141.1
	3.0	1.699e	292.7	0.180d	127.7
	3.5	1.248d	214.9	0.172cd	122.0
	4.0	1.161c	200.0	0.167c	118.6
	4.5	0.927bc	159.7	0.159bc	112.8
	5.0	0.804b	138.4	0.149ab	105.9

注：Ⅰ水稻, Ⅱ小麦, Ⅲ 油菜, 表中所列数据均为平均数。 同列不同字母表示不同处理间达差异显著水平 $P<0.05$。

值是水稻(未达到差异显著)<小麦(3.0)<油菜(4.0)，表明抗酸雨的能力为水稻>小麦>油菜。

2.2 种子萌发响应酸雨胁迫的时间效应

为了解酸雨对种子萌发生理变化的动态影响，试验中以POD活性和MDA含量为指标、设7 d为一周期，考察酸雨胁迫对种子生理过程的动态影响。在静态试验基础上，动态试验中舍弃pH2.0、2.5、4.5、5.0。

2.2.1 酸雨胁迫对3类种子萌发POD活性的影响

POD变化规律在一定程度上表征植物修复进程。图1-A显示，与CK相比3~7 d，pH3.0的POD活性呈先降后升趋势，并最终逼近CK。表明种子受酸雨胁迫后，应激修复启动，伤害在一定程度上缓和，导致POD活性随之下降，但当种子本身积累过多H_2O_2时，POD活性上升，又开始了新修复过程，缓解伤害，并逐渐恢复到正常水平。其他处理组变化趋势与pH3.0相似。图1-B显示，pH3.0和3.5的POD活性呈先上升后下降的趋势，并且逐渐背离CK。表明酶活应激上升，缓解酸雨对其伤害，随后酶活又渐降。pH4.0则一直上升，并且5~7 d超过CK。表明在此时段里，一直表现为应激上升。3个胁迫强度相比，酶活处于较高水平的时间pH3.0<pH3.5<pH4.0，表明随着胁迫强度增加，种子伤害程度也增加。图1-C中各个胁迫强度下的酶活的变化规律与图1-B相似，不同的是酶活均高于CK，表明细胞内积累过多H_2O_2，致使细胞发生一系列过氧化反应，使其明显受害。同时，伤害加剧刺激POD活性增加，由此推测，酸雨胁迫对油菜伤害较小麦严重。综上所述，对酸雨胁迫的响应为油菜>小麦>水稻，即抗酸雨能力为水稻>小麦>油菜。这一结论与前文[1-3]所述相符。

2.2.2 酸雨胁迫对3类种子萌发MDA含量的影响

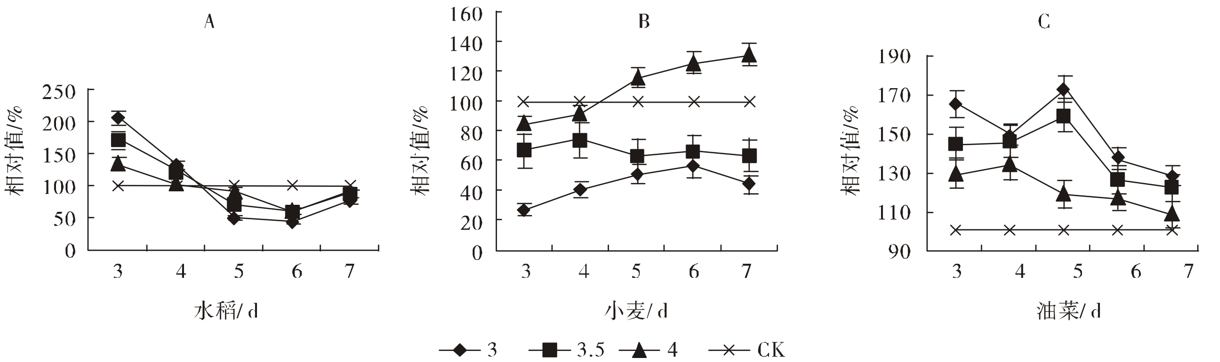

图1 酸雨胁迫对种子萌发POD活性的影响

Figure 1 Effects of acid rain on POD activity in the germination of the seeds

由图2-A知，与CK相比，在3~7 d，pH3.0~4.0酸雨处理水稻种子，MDA含量增幅为30.6%、7.9%、6.4%；45.6%、21.0%、9.0%；29.0%、14.7%、4.4%；22.2%、14.0%、6.2%；10.8%、10.0%、2.9%。呈先增后降变化规律，最终趋向正常水平，表明保护系统应激启动，有效减轻胁迫导致活性氧增加触发的膜脂过氧化，使伤害逐渐减轻。图2-B显示，在3~7 d，对于pH4.0，MDA下降，表明POD能有效清除酸雨胁迫产生活性氧，减轻膜脂过氧化。pH3.5变化规律反于该胁迫强度下POD变化规律，表明MDA含量(膜脂过氧化)受POD活性调控，即POD活性升MDA含量降，反之亦然。3~6 d，pH4.0的MDA也严格受上述POD活性的影响，而7 d时却反之，表明此刻酸雨胁迫导致植物实质伤害，POD参与活性氧生成，引发膜脂过氧化，MDA升高，酸雨对植物伤害加剧。与CK相比，3~7 d小麦MDA含量增幅为57.5%、37.1%、44.0%、55.6%、28.9%、7.0%、49.1%、31.9%、16.9%、99.8%、48.0%、18.47%、70.1%、41.9%、14.8%，比水稻同一时段的变幅增加。表明对酸雨胁迫响应为水稻<小麦，即抗酸雨能力为水稻>小麦。图1-C显示，在3~7 d，油菜所有处理组的变化规律与POD活性有关，且与上述规律相反。表明POD引发膜脂过氧化，MDA含量变化与POD酶活正相关。

2.2.3 酸雨胁迫下膜脂过氧化与POD关系

为进一步明晰酸雨胁迫下膜脂过氧化与POD活性间关系，对动态试验中的MDA-POD活性进行相关分析，得相关系数(r)于表2。分析发现，pH3.0~4.0酸雨胁迫下，3种种子的MDA-POD间相关性非常显著，且pH4.0>pH3.5>pH3.0。由此推断，各处理组通过提高植物细胞保护酶活性，清除自由基，进而减轻酸雨胁迫造成伤害，且pH4.0时修复效果明显优于pH3.5和3.0。水稻、小麦和油菜三者相比，水稻相关性明显要高于小麦和油菜。这表明，种子修复能力为水稻>小麦>油菜，即抗酸雨胁迫能力为水稻>小麦>

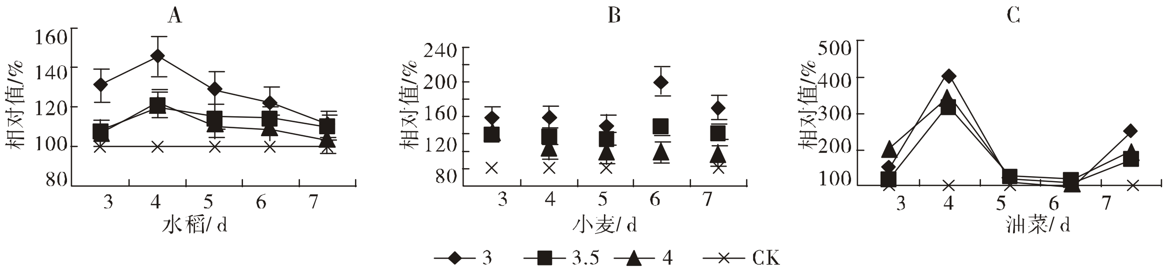

图 2 酸雨胁迫对种子萌发 MDA 含量的影响

Figure 2 Effects of acid rain on contents of MDA in the germination of the seeds

表 2 酸雨胁迫下膜脂过氧化与 POD 相关系数

Table 2 The correlation coefficients between membrane lipid peroxidation and the activity of POD under acidic rain stress

处理	3.0	3.5	4.0
水稻	0.9736	0.9777	0.9845
小麦	0.8607	0.8741	0.8941
油菜	0.7655	0.8166	0.8766

油菜。

3 结论

(1)静态试验结果显示,水稻、小麦、油菜种子 POD 活性随胁迫强度增大先升后降,MDA 含量升高且处理组的含量高于 CK。分析得知,抗酸雨胁迫能力为水稻>小麦>油菜。

(2)动态试验结果表明,胁迫强度一定时,不同种子 POD 与 MDA 的动态变化曲线不尽一致。胁迫时间一定时,所有处理组油菜 POD 活性均高于 CK,并随胁迫强度的增加上升,水稻则先增后降,小麦 pH4.0 最大,余则随胁迫强度增加而上升,水稻和小麦 MDA 含量随胁迫强度增加而增加,油菜则相反,表明抗酸雨胁迫能力为水稻>小麦>油菜。本文结果从种子萌发过程中,保护酶变化的角度揭示了 3 类植物耐酸雨分异的内在原因[7]。

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