

模拟铅污染土壤中杂草的菌根形成及对铅的吸收

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摘要:通过盆栽试验研究了 13 种杂草在模拟铅污染土壤中的菌根形成及对铅的吸收累积特点。结果表明:铅污染对杂草的生长没有明显影响。铅污染土壤中杂草的菌根侵染状况发生了显著变化,与无污染的土壤相比,苦荬菜(*Ixeris chinensis*)、早熟禾(*Poa annua*)、黑麦草(*Lolium perenne*)、野燕麦(*Avena fatua*)、野豌豆(*Vicia cracca*)、白车轴草(*Trifolium repens*)的菌根侵染率下降,而无芒稗(*Echinochloa crusgalli* var. *mitis*)、北美车前(*Plantago virginica*)、鼠曲草(*Gnaphalium affine*)和酢浆草(*Oxalis corniculata*)的菌根侵染率上升;鸡眼草(*Kummerowia striata*)、升马唐(*Digitaria ciliaris*)和婆婆纳(*Veronica didyma*)无明显变化;土壤中的孢子数除了鸡眼草、野豌豆、白车轴草和酢浆草无显著差异外,其余物种在铅污染土壤中的孢子数量与对照相比明显下降。不同的杂草物种对土壤铅的吸收和积累存在明显差异,被杂草吸收的铅主要积累在杂草根系内,向杂草地上部转移的铅比率不高。

关键词:铅污染; 杂草; 丛枝菌根真菌(AMF)

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Mycorrhizal colonization and lead uptake of weeds in lead-polluted soil

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Abstract: Mycorrhizal colonization and lead (Pb) uptake of weed species in a simulated Pb-polluted soil were studied by pot experiments in growth chamber. Thirteen species *Kummerowia striata*, *Digitaria ciliaris*, *Ixeris chinensis*, *Echinochloa crusgalli* var. *mitis*, *Plantago virginica*, *Trifolium repens*, *Veronica didyma*, *Gnaphalium affine*, *Vicia cracca*, *Avena fatua*, *Lolium perenne*, *Poa annua*, *Oxalis corniculata* from orchard were used in this experiment. Under monoculture, the growth of 13 weeds was not significantly affected when soil Pb concentration was 334.53mg/kg. Height and biomass of plants did not significantly differ from those grown in Pb elevated soil and their controls ($p > 0.05$). Compared to control, Pb concentration in roots of all species significantly increased under Pb treatment except *Amaranthus spinosus* and *Amaranthus paniculatus*. However, only 7 species of weeds were found increase in Pb concentration in shoots under Pb treatment. In Pb elevated soil, root Pb concentration in all species except *Amaranthus paniculatus*, was significantly higher than that of shoot Pb concentration, indicating that Pb was mainly accumulated in roots. Weed species differed significantly in Pb concentration of root among species, following the order *Oxalis corniculata* > *Lolium perenne* > *Gnaphalium affine* > *Vicia cracca* > *Lolium perenne* > *Plantago virginica* > *Poa annua* > *Kummerowia striata* > *Veronica didyma* > *Digitaria ciliaris* > *Echinochloa crusgalli* var. *mitis* > *Ixeris chinensis* > *Trifolium repens*.

Mycorrhizae were positive, negative or neutral influenced by Pb treatments among 13 species. Compared to the control, mycorrhizal infection rate of *Echinochloa crusgalli* var. *mitis*, *Plantago virginica*, *Gnaphalium affine*, *Oxalis corniculata* and *Eragrostis pilosa* was enhanced significantly ($p < 0.01$), *Ixeris chinensis*, *Poa annua*, *Lolium perenne*, *Avena fatua*, *Vicia*

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cracca and *Trifolium repens* was reduced significantly ($p < 0.01$), and *Kummerowia striata*, *Digitaria ciliaris*, *Veronica didyma*, *Amaranthus paniculatus*, *Amaranthus spinosus*, *Eleusine indica*, soybean and rice was not significantly affected. The vesicular numbers in mycorrhizal roots were also impacted by Pb treatments. In Pb elevated soil, vesicular numbers of *Poa annua*, *Lolium perenne*, *Avena fatua*, *Vicia cracca*, *Amaranthus paniculatus*, *Amaranthus spinosus*, *Eleusine indica*, *Eragrostis* and rice did not change, but vesicular numbers of *Veronica didyma*, *Plantago virginica*, *Gnaphalium affine*, *Trifolium repens*, *Oxalis corniculata*, *Echinochloa crusgalli* var *mitis* increased significantly, and vesicular numbers of soybean decreased. Mycorrhizal fungal spores were significantly influenced by Pb treatments. Except *Vicia cracca*, *Trifolium repens*, *Oxalis corniculata*, *Amaranthus paniculatus*, *Eleusine indica*, *Echinochloa crusgalli* var *mitis* and rice, mycorrhizal fungal spores of other species decreased significantly in Pb treatments ($p < 0.01$).

No significant relation was found between mycorrhizal infection rate and Pb uptake among weed species. Higher mycorrhizal infection rate did not enhance Pb uptake under Pb treatment.

Key words: lead pollution; weed plant; vesicular arbuscular mycorrhizal fungi

菌根是土壤中的菌根真菌与高等植物营养根系形成的一种共生体。研究表明,地球上80%的陆生植物可与菌根真菌共生形成菌根共生体^[1],菌根的形成能促进宿主植物对土壤中矿质元素P、N、K、Cu、Zn等的吸收,提高宿主根系对根部侵染病菌的抵抗能力和增强植物对干旱、高温、高盐和重金属的抗性,在植物生长发育中起着重要作用^[2~4]。菌根的形成受宿主植物的种类、土壤环境条件等因素的影响^[5~8]。许多研究表明,重金属污染对菌根的形成有明显影响,但不同的研究条件下得到的结果明显不同。如 Weissenhorn 等^[9,10]报道,土壤中重金属含量过高时显著降低了菌根真菌对三叶草、玉米根系的侵染,并影响真菌孢子的萌发。Chao 和 Wang^[11]通过盆栽试验也发现,提高镉的水平会抑制菌根真菌的侵染。而张美庆和王幼珊^[12]在灰钙土上使土壤含铅、镉、铜、砷量分别达20、100、400、200mg/kg时,3a的结果表明重金属不但没有抑制丛枝菌根真菌侵染小麦根系,反而表现出一定的促进作用。Gildon 和 Tinker^[13,14]报道被金属矿污染了的土壤(Zn达到83000mg/kg,Cd达到863mg/kg)中,自然生长的三叶草的菌根侵染率可以达到35%。

本文研究以菌根真菌侵染率为主要指标,比较分析在有铅和无铅污染的土壤上不同杂草物种菌根的形成差异和对铅的吸收特点,以期为杂草的科学利用和合理保持提供科学依据。

1 材料与方法

1.1 材料

供试土壤来自浙江省常山县胡柚果园,为第四纪红色粘土母质发育成的红壤。取0~20cm表层土壤,风干后过2mm尼龙筛备用。土壤基本理化性状测定方法为常规分析方法^[14],测试结果为有机质34.4±1.296 g/kg,速效氮48.1±3.9mg/kg,速效磷9.27±0.78mg/kg,速效钾69.6±4.6mg/kg,铅23.7±2.4mg/kg,pH5.8±0.6,阳离子交换量14.6±1.7 mmol/kg。供试的杂草物种来自常山县胡柚果园,是分布于胡柚果园的主要杂草种,包括鸡眼草(*Kummerowia striata*)、升马唐(*Digitaria ciliaris*)、苦荬菜(*Ixeris chinensis*)、无芒稗(*Echinochloa crusgalli* var *mitis*)、北美车前(*Plantago virginica*)、白车轴草(*Trifolium repens*)、婆婆纳(*Veronica didyma*)、鼠曲草(*Gnaphalium affine*)、野豌豆(*Vicia cracca*)、野燕麦(*Avena fatua*)、黑麦草(*Lolium perenne*)、早熟禾(*Poa annua*)和酢浆草(*Oxalis corniculata*)。

1.2 试验设计

采用盆栽试验的方法,盆的规格为:直径为32cm,高29cm,每盆装风干土8kg。设无铅污染土壤(不加铅,铅的背景含量为23.7±2.4 mg/kg)和铅污染土壤两个处理,铅污染土壤每盆加Pb(NO₃)₂(分析纯)2.4g,Pb(NO₃)₂以溶液态加入土壤中拌匀,使其浓度为300 mg/kg(以风干土计),具体浓度以实测为准。不加铅的土壤加入0.64g NH₄NO₃,以弥补因加入硝酸铅时造成的处理间氮素背景差异。土壤混合均匀后,加水使含水量为田间最大持水量的60%,平衡两周后测定铅含量,铅含量为334.5±30.6 mg/kg。在这两种土壤上,每一物种重复4次。因杂草对营养条件要求不高,为避免杂草疯长,本试验不施底肥。

1.3 植物生长量测定

杂草生长至4个月时取样,测定植株高度和植株地上部分和地下部分的生物量(干重)。

1.4 杂草根系丛枝菌根真菌(AMF)侵染检测和土壤孢子数量的测定

取完整杂草植株(包括地上部分和地下部分),带回实验室后立即清洗干净,剪下根部,固定于FAA溶液内。菌根侵染检测参照Giovannetti & Mosse的方法进行^[15];菌根感染率(%)=(感染根段长度/检测根长度)×100;泡囊数量以每毫米根段出现的泡囊个数表示。

1.5 土壤孢子数量的测定

土壤中孢子的数量采用湿筛倾注-蔗糖离心法测定^[16],称取相当于100g干土的新鲜土壤,倒入烧杯中,加入2000ml自来水浸泡2~3 min,玻棒充分搅匀,静置1 min后,倒入上层80目,下层320目的土壤筛中过滤,用自来水反复冲洗,把底筛中的孢子及杂物用自来水冲入离心管内加水定容,经3000 r/min离心4 min。轻轻倒去上悬液,保留沉淀物。加入45%蔗糖液,并摇匀管底沉淀物,3000 r/min离心2 min,取出离心管将上悬液抽滤到滤膜上,然后在显微镜下观察计数。

1.6 植株和土壤铅含量的测定

土壤样品去杂质后,风干、磨细、过筛(20目和100目);植物样品采回实验室后,先用自来水冲洗,再用蒸馏水冲洗,然后105℃杀青,85℃烘干磨碎待用。

土壤样品的铅采用王水和高氯酸消煮法,植物样品的铅采用干灰化湿消解法^[17],消煮溶液的铅用日本岛津AA-670型原子吸收分光光度计测定。

1.7 数据分析

试验结果采用SPSS10.0软件进行方差分析。

2 结果与分析

2.1 铅对杂草植物生长的影响

生长在铅含量为 $334.5 \pm 30.6 \text{ mg/kg}$ 的土壤上,各杂草物种的生长未明显受抑,植株高度、地上部及地下部的生物量与对照组差异不显著(表1)。

表1 铅对杂草物种的株高和生物量的影响

Table 1 The effects of Pb on plant height and biomass of different weeds

物种 Species	处理 Treatments	平均株高(cm/plant) Average height	地上部干重(g/pot) Shoot dry weight	地下部干重(g/pot) Root dry weight
<i>Avena fatua</i>	CK	59.6±6.0	31.11±8.82	13.39±5.54
	+Pb	52.8±2.4	30.28±6.19	12.83±3.04
<i>Digitaria ciliaris</i>	CK	26.9±2.4	6.31±3.01	1.55±0.24
	+Pb	27.4±3.5	5.15±2.10	1.57±0.15
<i>Echinochloa crusgalli</i> var. <i>mitis</i>	CK	46.8±5.7	12.96±2.97	3.01±0.88
	+Pb	45.9±3.3	13.85±1.45	2.45±0.62
<i>Gnaphalium affine</i>	CK	29.7±1.1	11.46±4.67	1.96±0.87
	+Pb	23.5±3.2	12.82±4.60	1.93±0.96
<i>Ixeris chinensis</i>	CK	55.7±2.9	19.98±3.68	2.44±1.48
	+Pb	56.9±3.7	18.30±5.12	2.30±1.13
<i>Kummerowia striata</i>	CK	32.1±3.2	53.47±6.22	4.01±1.14
	+Pb	31.6±5.4	54.05±9.55	4.38±1.06
<i>Lolium perenne</i>	CK	57.4±6.9	19.91±4.15	18.66±5.07
	+Pb	55.7±5.9	16.48±1.87	18.13±6.53
<i>Oxalis corniculata</i>	CK	22.6±3.9	4.99±0.36	2.34±0.39
	+Pb	23.3±2.0	5.73±0.22	1.55±0.21
<i>Plantago virginica</i>	CK	20.4±2.1	12.96±5.03	12.88±5.06
	+Pb	22.0±2.8	12.62±4.31	12.62±4.31
<i>Poa annua</i>	CK	34.9±2.4	17.84±3.66	3.06±1.09
	+Pb	33.3±3.8	16.48±1.87	3.50±1.10
<i>Trifolium repens</i>	CK	21.1±2.4	14.44±3.01	2.89±1.26
	+Pb	23.0±1.5	15.80±2.75	2.75±1.41
<i>Veronica didyma</i>	CK	17.8±3.2	11.20±2.84	3.05±1.07
	+Pb	19.7±2.5	10.31±1.66	2.96±1.02
<i>Vicia cracca</i>	CK	80.9±5.9	10.06±5.18	1.31±0.55
	+Pb	79.2±14.6	9.08±4.44	1.29±0.46

* 表中数据为4个重复的统计结果(SPSS10.0),以均值±标准差表示;ns表示植物对照与铅处理之间无显著差异;*, ** 分别表示 $p < 0.05$ 和 $p < 0.01$ 的差异显著性;下同 Data in the table were statistical results from four replications, Value are mean±sd; "ns" no significant difference; "*" significant difference $p < 0.05$; "** significant difference $p < 0.01$, the same below

2.2 铅污染土壤上不同杂草物种菌根形成的差异

表2表明,在铅污染土壤中杂草菌根的形成发生了显著变化,但不同物种的变化趋势明显不同。与无铅污染的对照土壤相比,苦荬菜、早熟禾、黑麦草、野燕麦、野豌豆、白车轴草的菌根侵染率下降($p < 0.01$),而无芒稗、北美车前、鼠曲草和酢浆草的菌

根侵染率上升($p<0.01$),只有鸡眼草、升马唐和婆婆纳的变化不大(差异不显著)。从表2还可见,鸡眼草、苦荬菜、升马唐、早熟禾、黑麦草、野燕麦、野豌豆的泡囊数没有明显变化,而无芒稗、婆婆纳、北美车前、鼠曲草、白车轴草、酢浆草的泡囊数在铅污染土壤中明显增加。土壤中的孢子数量一定程度上可反映AMF的多样性和繁殖活性,表2中的数据表明,除了鸡眼草、野豌豆、白车轴草和酢浆草无显著差异外,其余物种在铅污染土壤中的孢子数量与对照相比明显的下降($p<0.01$)。

表2 铅污染对杂草植物菌根侵染率与根际土壤中的孢子形成的影响

Table 2 Effects of Pb on mycorrhizal colonization and spore numbers of weeds

物种 Species	处理 Treatments	菌根侵染率 Percentage of mycorrhizal colonization (%)	根内泡囊数 Number of vesicular (number/mm root)	土壤中孢子数 Number of spore in soil (spore/kg dry soil)
<i>Avena fatua</i>	CK	9.50±1.29	2.0	29693±994
	+Pb	2.45±0.18**	2.0	10287±898**
<i>Digitaria ciliaris</i>	CK	46.60±15.01	5.0	28904±667
	+Pb	47.24±20.41	5.4	16359±316**
<i>Echinochloa crusgalli</i> var. <i>mitis</i>	CK	78.28±2.49	8.1	29667±861
	+Pb	91.36±6.83*	53.0	18861±432**
<i>Gnaphalium affine</i>	CK	16.38±1.82	4.0	38880±1545
	+Pb	30.37±3.87**	26.5	11870±1354**
<i>Ixeris chinensis</i>	CK	82.21±5.65	7.2	20317±867
	+Pb	74.64±6.40*	6.8	14250±562**
<i>Kummerowia striata</i>	CK	94.51±0.85	4.0	28814±1002
	+Pb	96.13±1.50	6.0	27913±532ns
<i>Lolium perenne</i>	CK	7.34±2.22	2.0	24120±659
	+Pb	1.19±0.07**	2.0	16323±9372**
<i>Oxalis corniculata</i>	CK	21.03±1.23	4.0	30780±3307
	+Pb	44.96±9.30**	51.7	29470±5448ns
<i>Plantago virginica</i>	CK	17.61±6.53	10.3	50113±911
	+Pb	31.54±2.87**	24.7	18723±4985**
<i>Poa annua</i>	CK	19.10±4.07	3.0	28280±583
	+Pb	3.14±0.74**	3.0	9735±600**
<i>Trifolium repens</i>	CK	29.55±6.98	1.0	34949±2951
	+Pb	19.83±4.71**	43.7	32532±3048ns
<i>Veronica didyma</i>	CK	7.23±1.82	1.0	28668±340
	+Pb	10.80±2.86ns	8.0	9083±1717**
<i>Vicia cracca</i>	CK	33.20±8.18	5.5	27780±412
	+Pb	14.88±3.47**	5.3	24107±3875ns

2.3 植物对铅积累的物种间差异

表3表明,铅处理下13种杂草根系铅含量与对照之间均存在极显著差异,根系有明显积累铅的趋势。铅处理的土壤中,植株地上部分的铅含量除了鸡眼草、苦荬菜、升马唐、早熟禾、婆婆纳、鼠曲草、白车轴草与对照无显著差异外,其它均有显著差异。根系铅含量与植株地上部分含量之比为0.83~28.99,说明铅主要积累在杂草根部。根部铅含量(以Pb mg/kg根系表示)酢浆草>黑麦草>鼠曲草>野豌豆>野燕麦>北美车前>早熟禾>鸡眼草>婆婆纳>升马唐>无芒稗>苦荬菜>白车轴草。而单位面积(以每盆计算)杂草的吸铅数量(吸铅量=植株含铅量(植物生物量))则以黑麦草、野燕麦、北美车前、鸡眼草和苦荬菜为最高(图1)。由于每种杂草地上部分与根系生物量的差异,黑麦草、野燕麦和北美车前等根系较发达的物种根部吸铅数量大。而鸡眼草和苦荬菜是地上部分的吸铅数量比较大。早熟禾、婆婆纳、鼠曲草、白车轴草和酢浆草根系铅含量较高的物种也是根部吸铅数量大于地上部分。

3 讨论

3.1 土壤铅污染对杂草菌根形成的影响

已有的研究表明,在土培或水培条件下,重金属或增加或降低菌根的侵染率或对菌根的侵染率无影响。Weissenhorn等^[18]发现,在重金属(Cd, Zn, Pb, 和 Cu)污染的土壤上玉米的菌根形成和孢子数量比无污染的高。Hildebrandt等^[19]也发现,在污染严重的土壤上 *V. calaminaria* 的菌根侵染率增加。但是,Chao 和 Wang^[19]报道,增加重金属(Zn, Cu, Ni, Cr, Pb 和 Cd)导致玉米的菌根侵染率降低。Weissenhorn 和 Leyval^[10]的研究表明玉米的菌根侵染率与土壤重金属污染程度无显著相关性。Diaz 等^[21]也发现,土壤中的Zn或Pb对 *Lygeum spartum* 和 *Anthyllis cytisoides* 的菌根侵染率无影响。本文研究表明,铅对

表 3 不同土壤条件下杂草不同器官内的铅含量

Table 3 Pb concentration in shoots and roots of weeds

物种 Species	处理 Treatment	地上部分铅含量 Pb content in shoot (mg/kg)	根的铅含量 Pb content in root (mg/kg)	根铅/地上部铅 Pb content in root/Pb content in shoot
<i>Avena fatua</i>	CK	6.6±0.4	13.0±1.0	2.0
	+Pb	25.5±1.2**	185.2±22.8**	7.3
<i>Digitaria ciliaris</i>	CK	74.5±2.0	79.6±2.5	1.1
	+Pb	77.7±2.3 ns	133.5±7.5**	1.7
<i>Echinochloa crusgalli</i> var. <i>mitis</i>	CK	20.3±3.9	20.5±0.8	1.0
	+Pb	47.0±5.4**	129.9±11.4**	2.8
<i>Gnaphalium affine</i>	CK	2.6±0.1	21.6±3.2	8.3
	+Pb	6.9±1.5 ns	200.6±27.4**	29.0
<i>Ixeris chinensis</i>	CK	77.4±1.5	77.3±3.0	1.0
	+Pb	79.3±1.2 ns	98.8±10.4**	1.2
<i>Kummerowia striata</i>	CK	17.6±2.3	14.6±1.0	0.8
	+Pb	23.3±2.7 ns	149.5±10.5**	6.4
<i>Lolium perenne</i>	CK	6.2±1.6	11.6±1.2	1.9
	+Pb	21.9±3.4**	226.2±62.0**	10.3
<i>Oxalis corniculata</i>	CK	12.0±0.7	17.3±3.0	1.4
	+Pb	20.0±1.4**	227.9±7.2**	11.4
<i>Plantago virginica</i>	CK	2.8±0.9	10.9±1.1	3.9
	+Pb	22.1±0.9**	171.2±13.2**	7.7
<i>Poa annua</i>	CK	21.6±2.6	24.4±2.2	1.1
	+Pb	18.2±3.3 ns	166.1±11.3**	9.1
<i>Trifolium repens</i>	CK	2.3±0.1	18.4±1.0	8.02
	+Pb	4.7±0.5 ns	92.5±12.7**	19.7
<i>Veronica didyma</i>	CK	4.6±0.3	17.4±1.0	3.8
	+Pb	5.3±0.5 ns	146.3±17.7**	27.6
<i>Vicia cracca</i>	CK	2.5±0.3	6.0±0.3	2.4
	+Pb	22.0±1.3**	191.7±13.7**	8.7

菌根侵染率的影响不同物种之间存在明显差异,铅降低苦荬菜、早熟禾、黑麦草、野燕麦、野豌豆、白车轴草的菌根侵染率,但增加无芒稗、北美车前、鼠曲草和酢浆草的侵染率而对鸡眼草、升马唐和婆婆纳的侵染率无影响。泡囊的形成可反映 AMF 在宿主根系的定殖程度,在铅污染土壤上,菌根的泡囊或增加或不变。泡囊的增加可能是对铅浓度增加的一种响应。Whitfield 等^[22]亦报道,在污染严重的土壤,泡囊数量增加,而且泡囊数量与土壤 Cd 和 Zn 提取浓度呈正相关。但菌根泡囊数量是否与植物抗重金属有关,目前仍知之甚少,需要做进一步的研究。

3.2 菌根形成对杂草吸收铅的影响

一些研究报道,在重金属污染的环境中菌根真菌可降低植物对重金属的吸收,从而增加植物对重金属污染的抵抗和促进植物生长^[10,21,23]。但是,Marschner 等^[24]报道,经过 42d 的处理后,与无菌根的处理相比,菌根增加铅在根系的积累。Joner 和 Leyval^[25,26]也发现,接种菌根真菌的处理,根系内铅的积累明显增加。砂培试验①表明,随着铅浓度增加,菌根明显增加植物对铅的吸收。但在本研究中,菌根的形成与铅吸收未见明显相关关系,需要作进一步研究。

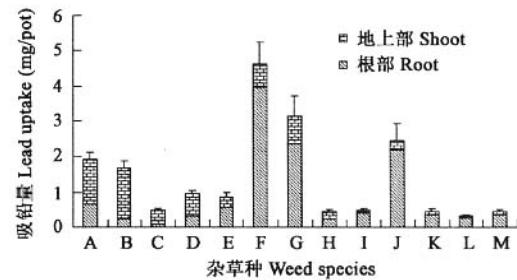


图 1 不同杂草植株不同部位摄铅量(以每盆计算)

Fig. 1 Total lead uptake in root and shoot of various weed plants (mg/pot)

A 鸡眼草 *Kummerowia striata*; B 苦荬菜 *Ixeris chinensis*; C 升马唐 *Digitaria ciliaris*; D 无芒稗 *Echinochloa crusgalli* var. *mitis*; E 早熟禾 *Poa annua*; F 黑麦草 *Lolium perenne*; G 野燕麦 *Avena fatua*; H 野豌豆 *Vicia cracca*; I 婆婆纳 *Veronica didyma*; J 北美车前 *Plantago virginica*; K 鼠曲草 *Gnaphalium affine*; L 白车轴草 *Trifolium repens*, M 酢浆草 *Oxalis corniculata*

① 吴春华. 植物多样性对铅污染土壤的响应及其生态学效应. 浙江大学博士学位论文. 2004

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