

内蒙古四种牧草对土壤汞的富集能力与生理响应

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摘要: 随着内蒙古就地煤电转化发展方式的推行, 周边土壤中来自大气汞沉降逐步加剧。为了解牧草对汞的富集能力与耐性机理, 选取内蒙古草场常见四种牧草: 羊草、紫花苜蓿、披碱草和高羊茅进行研究。通过野外汞沉降模拟试验及汞胁迫盆栽种植试验, 分别测定土壤、牧草中的总汞含量及牧草的生理生化值, 分析被试牧草汞含量与土壤汞含量相关性。供试土壤为砂土, pH 在 7.43—8.53, 当土壤中汞含量不高于 $1.0 \text{ mg} \cdot \text{kg}^{-1}$ 时, 四种牧草富集系数皆大于 1。结果表明: 不同牧草对汞富集能力不同并且会受土壤中汞含量影响, 牧草对汞的富集系数随汞胁迫浓度增高而逐渐下降, 汞胁迫下牧草细胞膜脂过氧化程度提高, 牧草通过提高抗氧化酶酶活、脯氨酸含量调节细胞是其耐受汞的重要机理。

关键词: 牧草; 汞; 富集; 生理响应

Accumulation characteristics and physiological responses of four kinds of herbage to mercury in Inner Mongolia

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Abstract: Background, aim, and scope Some of large coal enterprises in Inner Mongolia have been striving to develop coal-fired power plants and promoting the integration of coal and electricity, which leads to the rapid increase of coal consumption in Inner Mongolia. During the coal combustion, the phenomenon of mercury emissions into the atmosphere appears, which results in serious impacts on environmental pollution. This paper aims to study the mercury enrichment capacity of various herbage and their tolerance to mercury through the experiment of mercury stress in herbage in Inner Mongolia. **Materials and methods** The experimental materials included *Leymus chinensis*, *Medicago sativa*, *Elymus dahuricus*, *Festuca elata*, the herbage and seeds were provided by the grassland station of the Chinese Academy of Sciences in Dongwuzhu, Inner Mongolia, the experimental soil was collected from grassland of Chinese Academy of Sciences in Dongwuzhu, Inner Mongolia. The method of microwave digestion was used to determine mercury content in plant and soil, malondialdehyde (MDA) was determined by colorimetric method, the soluble protein was determined by coomassie brilliant blue staining, the soluble sugar content was determined by anthrone method, respectively. **Results** In the case of low

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concentration of mercury, all the four kinds of herbages had certain adsorption capacity on mercury, but when the concentration of mercury in soils continued to increase, mercury enrichment effect of four kinds of herbages did not increase as expected like enrichment plants. When the mercury concentration in soil was not higher than that of the soil quality standard, the enrichment coefficient (EC) of the four herbages were all greater than 1, which reflected better ability to accumulate mercury. Average mercury adsorption efficiency of *Leymus chinensis* was 26.22%, followed by *Elymus dahuricus*, the adsorption efficiency was 24.61%, then is *Festuca elata*, the adsorption efficiency was 22.69%, the lowest was *Medicago sativa*, the adsorption efficiency was 15.57%. The correlation between heavy metals in plant and soil is related to the physiological characteristics of plant, indicating that the mercury content in herbage is related to the mercury content in soil. With the increase of mercury stress concentration, the content of malondialdehyde (MDA) in the four kinds of herbages decreased first and then rose. After reaching the lowest value, the MDA content of the four herbages increased with the increase of mercury stress concentration. Among them, *Medicago sativa* had the greatest change in MDA content under mercury stress, which was mostly affected by mercury. When the mercury stress concentration was $64.0 \text{ mg} \cdot \text{kg}^{-1}$, the proline content of the four kinds of herbages was the highest, respectively 7.48 times, 5.63 times, 3.42 times and 4.02 times of the control group. Among the four kinds of herbages, *Leymus chinensis* was most powerful for the stress resistance of mercury. **Discussion** Enrichment coefficient is the ratio of the content of certain heavy metals in plants to that in soils, which is one of the important indexes to evaluate the enrichment ability of plants to heavy metals. The greater the enrichment coefficient, the stronger the accumulation ability. Four kinds of herbages have adsorption effect on mercury in soil at various levels of mercury stress. The mercury content in herbage is correlated with mercury content in soil. MDA and proline can reflect mercury physiological toxicity to plants. **Conclusions** The mercury accumulation in plants is significantly affected by different concentrations of mercury stress. Four kinds of herbages have certain adsorption and transfer ability to mercury in soil. When the mercury stress concentration is low ($<16.0 \text{ mg} \cdot \text{kg}^{-1}$), *Medicago sativa* has the strongest adsorption capacity. After the increase of mercury stress concentration ($>16.0 \text{ mg} \cdot \text{kg}^{-1}$), *Elymus dahuricus* has the strongest adsorption capacity. The mercury enrichment coefficient decreases with the increase of mercury stress concentration, among which the maximum enrichment coefficient of *Leymus chinensis* is the highest. Mercury stress has significant effect on the physiology of different herbages. When the mercury content in soil is too high, it will produce toxic effect on the growth of four kinds of herbages. Under a certain concentration of mercury stress, the herbages can balance cell osmotic pressure by raising proline content, promoting the antioxidant enzymes activity, slowing down the increase of reactive oxygen species and membrane lipid peroxidation caused by mercury stress, thus maintaining the normal growth environment of plants. **Recommendations and perspectives** Inner Mongolia is a coal-producing province, while promoting the economic development model of coal-electricity integration, long-term planning and protection of grassland ecology should be carried out. Faced with increasing coal-fired power generation, mercury released from coal-fired power plants will also increase. Proper measures should be taken to control mercury content in gas emissions from coal-fired power plants, so as to prevent the impact of long-term and low concentration mercury emissions from coal-fired power plants on grassland vegetation from the source.

Key words: herbages; mercury; accumulation characteristics; physiological response

内蒙古地区地层发育齐全, 岩浆活动频繁, 成矿条件好, 矿产资源丰富, 仅呼伦贝尔、锡林浩特、霍林郭勒、鄂尔多斯的煤炭探明储量已达 3903.48 亿吨, 约占全国总储量的 43.5%。自 2012 年, 内蒙古采用燃煤发电这一方式, 大力推广煤电一体化

项目, 转煤输送为电输送, 使该地区燃煤量大幅增长, 汞排放量随之升高。相关研究已证实, 内蒙古地区燃煤电厂汞排放量占总排放量的 41.3%, 是大气汞排放的主要来源之一(张静静等, 2014)。大气中汞通过干湿沉降返回地面, 进入土壤, 从

而影响植物生长 (Pirrone et al, 2010)。汞是植物生长的非必需元素, 当土壤中汞胁迫浓度过高后, 会对植物产生毒害作用。通过在全区范围实地采样检测, 已知内蒙古地区土壤中汞含量范围是 $0.0277\text{—}3.0977\text{ mg}\cdot\text{kg}^{-1}$, 参考国家土壤环境质量标准 (GB 15618—2018) (土壤中汞含量不超过 $3.4\text{ mg}\cdot\text{kg}^{-1}$), 所调查地区部分土壤中汞含量已临近国家标准, 随着汞排放不断积累, 将威胁植物正常生长。内蒙古作为全国重点牧区, 草原承担着为畜牧提供大量食料的重要任务, 但其草原土壤汞污染不断加剧, 不仅威胁草原生态系统第一生产者牧草的生长, 进而还将通过食物链危及畜类乃至人类健康。目前, 国内外大量汞污染相关研究工作多以蔬菜、谷物为主 (庞欣等, 2001; 刘芳等, 2013; 陈剑等, 2016), 鲜有以牧草为研究对象的相关研究。本文通过对内蒙古地区牧草汞胁迫实验, 研究四种牧草的汞富集能力及对汞的耐受性, 以期草原牧草在汞胁迫作用下生理响应的相关研究积累有价值的信息, 同时为寻求草原土壤中汞的生物治理措施提供理论依据。

1 材料与方法

1.1 野外样地布设

样地选取在内蒙古东乌珠穆沁旗, 北纬 $45^{\circ}34'58''$, 东经 $116^{\circ}57'60''$, $10\text{ m}\times 7\text{ m}$ 样地。使用分析纯氯化汞 (HgCl_2), 根据土壤污染风险管控标准, 依照等比法设置处理水平间隔 (洪曾纯, 2012), 模拟大气汞沉降进入本底土壤后影响, HgCl_2 处理设置 10 个水平 (0 、 $0.5\text{ mg}\cdot\text{kg}^{-1}$ 、 $1.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $1.5\text{ mg}\cdot\text{kg}^{-1}$ 、 $2.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $4.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $8.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $16.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $32.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $64.0\text{ mg}\cdot\text{kg}^{-1}$), 每个处理 7 次重复。经过 14 周生长期后, 将样地植物 (主要物种为羊草 *Leymus chinensis* (Trin.) Tzvel) 整株采集。并挖取植株根部附近 5 cm 深土壤样品带回测定。

1.2 盆栽试验设计

采用盆栽汞胁迫控制试验, 供试土壤为内蒙古东乌珠穆沁旗中科院草原站草场内土壤, 土样风干、碾碎后过 2 mm 筛后混匀, 每盆 $7\text{ cm}\times 7\text{ cm}\times 7\text{ cm}$, 装土 150 g (干重)。 HgCl_2 处理设置 10 个水平 (0 、 $0.5\text{ mg}\cdot\text{kg}^{-1}$ 、 $1.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $1.5\text{ mg}\cdot\text{kg}^{-1}$ 、 $2.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $4.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $8.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $16.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $32.0\text{ mg}\cdot\text{kg}^{-1}$ 、 $64.0\text{ mg}\cdot\text{kg}^{-1}$), 每个处理 3 次重复,

土样混匀、稳定 7 天。供试种子由内蒙古东乌珠穆沁旗中科院草原站提供, 将筛选出的饱满籽粒的种子 (紫花苜蓿 *Medicago sativa* L.、披碱草 *Elymus dahuricus* Turcz.、高羊茅 *Festuca elata* Keng ex E. Alexeev) 浸泡在蒸馏水中催芽, 露白后选择长势一致的种子种于花盆中, 每盆种入 15 颗。日常以去离子水维持适宜水分, 采用自然光照。

1.3 测定方法

参考《食品中总汞及有机汞的测定》(GB 5009.17—2014) 和《土壤和沉积物汞、砷、硒、铋、锑的测定——微波消解/原子荧光法》(HJ 680—2013), 对牧草根部长及土壤进行微波消解 (仪器型号 MD6C—4H), 用原子荧光光度计 (普析通用 PF—7 原子荧光光度计), 测定总汞含量。生理指标测定参考植物生理学指导 (华东师范大学生物系植物生理教研室, 1980)。本文实验误差控制在 1%。

1.4 数据处理

植物中汞富集系数按下式计算: 富集系数 = 植物体内汞含量 / 土壤中汞含量 (刘国锋, 2006)。

数据运算采用 Excel 2007, 统计分析采用 SPSS 19.0 中的单因素方差分析法 (ANOVA), 运用 LSD、Duncan 对数据进行两两比较。方差分析之前, 先对数据进行正态分布检验及方差齐性检验。在图表中有相同字母数据为无显著性差异 ($p>0.05$), 不同字母表示数据有显著性差异 ($p<0.05$)。

2 结果与分析

2.1 牧草对土壤汞的富集能力

2.1.1 牧草汞含量与土壤汞含量相关性

植物中重金属含量与土壤中重金属之间的相关性与植物生理特征有关 (黄玉芬, 2011; 赵鲁等, 2013; Pavlish et al, 2003)。牧草中汞含量与土壤中汞含量具有相关性, 由表 1 可知: 在试验研究范围内 ($0.5\text{—}64.0\text{ mg}\cdot\text{kg}^{-1}$), 羊草、紫花苜蓿、披碱草中汞含量与土壤中汞含量呈显著正相关 ($p<0.01$), 高羊茅与土壤中汞含量无显著相关性。由图 1 可知: 当土壤中汞含量高于 $16.0\text{ mg}\cdot\text{kg}^{-1}$ 时, 高羊茅汞含量明显降低, 分析土壤中汞含量在 $0.5\text{—}16.0\text{ mg}\cdot\text{kg}^{-1}$ 时, 高羊茅汞含量与土壤汞含量的相关性, 发现 Pearson 相关性为 0.966, 显著性 (双侧) 为 0.00, 呈显著正相关

($p < 0.01$)。当土壤中汞含量过高后, 牧草受到毒害作用, 汞积累量降低。

表 1 牧草中汞含量与土壤汞含量相关性 Tab.1 Correlation between mercury content in herbage and soils		
牧草 Herbages	Pearson 相关性 Pearson correlation	显著性 (双侧) Significance level
羊草 <i>Leymus chinensis</i>	0.953**	0.000
紫花苜蓿 <i>Medicago sativa</i>	0.901**	0.000
披碱草 <i>Elymus dahuricus</i>	0.891**	0.000
高羊茅 <i>Festuca elata</i>	0.309	0.097

**：在 0.01 水平 (双侧) 上显著相关。
**：The correlation was significant at the 0.01 level (bilateral).

2.1.2 牧草的汞富集系数

富集系数 (EC) 指植物体内某种重金属含量与土壤中该种重金属原有含量的比值, 是评价植物

富集重金属能力的重要指标之一, 可以反映植物对重金属的富集能力, 富集系数越大, 表示其富集能力越强 (刘月莉, 2008)。杨保华 (2004) 对水马齿、眼子菜研究实验结果表明: 汞的富集是生物主动吸收与表面被动吸附共同作用的结果。刘雅妮等 (2014) 对汞矿废弃地带植物富集能力研究表明: 土壤汞浓度太高情况不适宜植物生长。本实验中, 四种牧草富集系数大于 1 对应土壤中汞胁迫浓度临界梯度依次为: $1.5 \text{ mg} \cdot \text{kg}^{-1}$ (羊草)、 $4.0 \text{ mg} \cdot \text{kg}^{-1}$ (紫花苜蓿)、 $2.0 \text{ mg} \cdot \text{kg}^{-1}$ (披碱草)、 $4.0 \text{ mg} \cdot \text{kg}^{-1}$ (高羊茅) (表 2), 表明在较低浓度汞胁迫的情况下, 四种牧草对土壤中汞具有一定的吸附能力, 根据土壤环境质量标准, 当土壤中汞含量浓度未超过土壤质量标准要求的范围内时, 四种牧草的富集系数皆大于 1, 具有较好的富集重金属汞能力。但当土壤中汞胁迫浓度继续增加后, 吸附汞量增加, 但四种牧草富集增量汞与环境增量不成比例, 因环境增量可无限, 富集量有限, 因而富集系数呈减小趋势, 四种牧草对于汞的富集效果不理想, 无法作为高含量土壤中富集汞的植物。

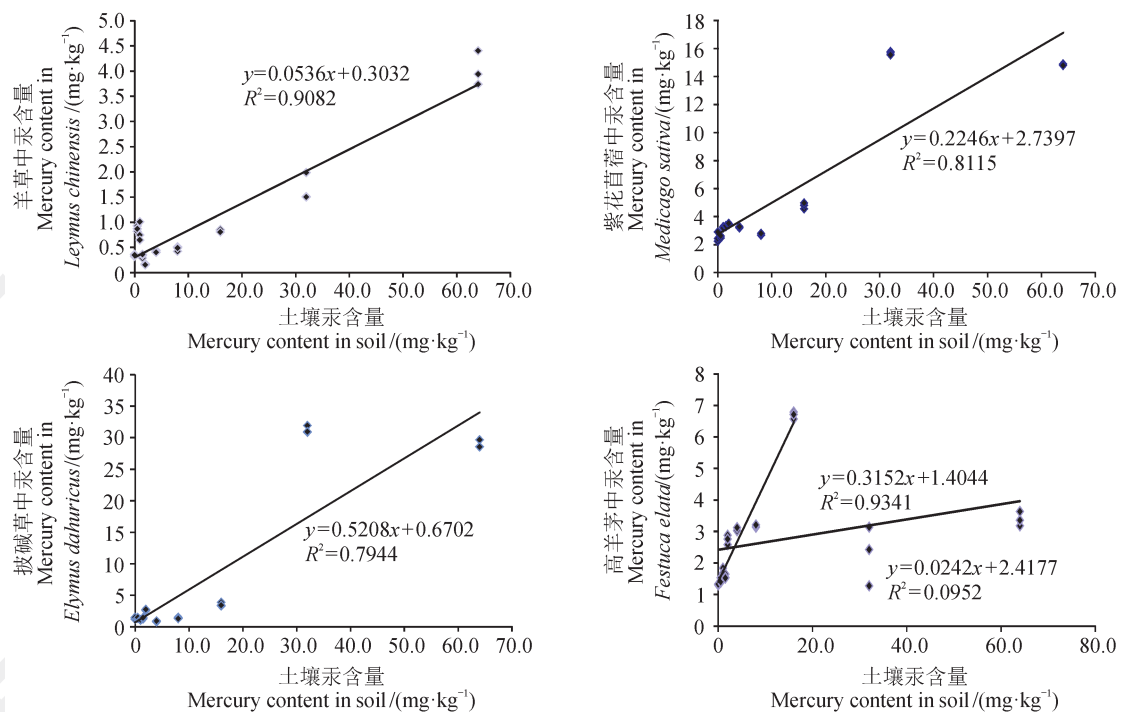


图 1 牧草汞含量与土壤汞含量的关系

Fig.1 Relation of herbage mercury content to soil mercury content

2.1.3 牧草对土壤汞的吸附能力

由表3可知: 四种牧草在各汞胁迫水平下均对土壤中的汞有吸附作用。牧草对土壤中汞的吸附效率为: 羊草>披碱草>高羊茅>紫花苜蓿, 分别是: 26.22%、24.61%、22.69%、15.57%。同种牧草比较发现: 羊草在土壤中汞浓度为 $0.5 \text{ mg} \cdot \text{kg}^{-1}$

和 $1.5 \text{ mg} \cdot \text{kg}^{-1}$ 之间时, 与对照组相比无显著性差异 ($p>0.05$)。其他浓度胁迫下, 羊草对汞吸附能力呈显著性差异 ($p<0.05$), 且当汞胁迫浓度为 $16.0 \text{ mg} \cdot \text{kg}^{-1}$ 和 $32.0 \text{ mg} \cdot \text{kg}^{-1}$ 时, 呈极显著差异 ($p<0.01$), $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时显著下降, 降至对照组的 19.40%。

表2 四种牧草的汞富集系数
Tab.2 Mercury EC of four herbage

汞胁迫浓度 Concentration of $\text{Hg}^{2+}/(\text{mg} \cdot \text{kg}^{-1})$	富集系数 EC			
	羊草 <i>Leymus chinensis</i>	紫花苜蓿 <i>Medicago sativa</i>	披碱草 <i>Elymus dahuricus</i>	高羊茅 <i>Festuca elata</i>
0.5	3.29	1.11	2.70	2.77
1.0	1.50	3.23	1.05	1.83
1.5	0.39	2.20	1.28	1.09
2.0	0.18	1.73	0.93	1.29
4.0	0.20	0.80	0.20	0.75
8.0	0.11	0.33	0.17	0.39
16.0	0.11	0.28	0.24	0.42
32.0	0.09	0.49	0.96	0.04
64.0	0.12	0.23	0.45	0.05

表3 不同牧草在汞胁迫下对土壤中汞的吸附情况
Tab.3 Adsorption of mercury in soils by different herbage under mercury stress

处理汞浓度 Concentration of $\text{Hg}^{2+}/(\text{mg} \cdot \text{kg}^{-1})$	牧草吸附后土壤中汞的残余量 Residual content of mercury in soils after the mercury were adsorbed by herbage/ $(\text{mg} \cdot \text{kg}^{-1})$				
	无植物 No plants	羊草 <i>Leymus chinensis</i>	紫花苜蓿 <i>Medicago sativa</i>	披碱草 <i>Elymus dahuricus</i>	高羊茅 <i>Festuca elata</i>
0.0	0.23+0.00a	0.19+0.04ab	0.21+0.01ab	0.18+0.02a	0.19+0.01a
0.5	0.73+0.00a	0.58+0.04ab	0.64+0.02ab	0.55+0.02b	0.65+0.03a
1.0	1.23+0.00a	1.19+0.02ab	1.19+0.03ab	1.13+0.04bc	1.06+0.01c
1.5	1.73+0.00a	1.64+0.01ab	1.62+0.02ab	1.55+0.01b	1.53+0.03b
2.0	2.23+0.00a	2.04+0.04b	2.06+0.02ab	1.82+0.01c	1.82+0.02c
4.0	4.23+0.00a	2.59+0.01d	3.93+0.01b	3.55+0.02c	2.50+0.01d
8.0	8.23+0.00a	4.77+0.02c	7.24+0.01b	4.89+0.02c	6.81+0.02b
16.0	16.23+0.00a	9.42+0.02d	12.94+0.01c	12.78+0.02c	14.97+0.01b
32.0	32.23+0.00a	16.26+0.01d	19.89+0.01bc	19.13+0.01c	20.77+0.01b
64.0	64.23+0.00a	51.77+0.02b	38.68+0.01c	35.85+0.01c	29.96+0.01d

不同字母表示不同浓度间显著性差异 ($p<0.05$)。

Different letters indicate significant differences between different concentrations ($p<0.05$).

紫花苜蓿在土壤汞胁迫浓度为 $0.5 \text{ mg} \cdot \text{kg}^{-1}$ 和 $2.0 \text{ mg} \cdot \text{kg}^{-1}$ 之间时, 与对照组相比无显著性差异 ($p>0.05$)。当汞胁迫浓度达到 $4.0 \text{ mg} \cdot \text{kg}^{-1}$ 后, 紫花苜蓿对汞吸附能力呈显著性差异 ($p<0.05$), 且当汞胁迫水平为 $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时, 吸附效率最高,

达到 39.78%。

披碱草在各浓度汞胁迫下, 对土壤中汞的吸附能力与对照组相比皆呈显著性差异 ($p<0.05$)。与紫花苜蓿相同, 在汞胁迫水平为 $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时, 吸附效率达到最高, 为 44.18%。表明披碱草可对

汞污染土壤起到修复作用，能够作为萃取植物。

高羊茅仅在汞胁迫水平为 $0.5 \text{ mg} \cdot \text{kg}^{-1}$ 时，对土壤中汞的吸附与对照组相比无显著性差异 ($p > 0.05$)，其他浓度下皆为显著性差异。且当汞胁迫水平为 $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时，吸附效率达到最高，为 53.36%。

2.2 牧草中汞的生理毒性

丙二醛 (MDA)、脯氨酸可以反映汞对植物的生理毒害。汞毒害可使线粒体上 ATP 酶活性受到抑制，影响能量的产生与供应，破坏细胞膜结构，使膜中不饱和脂肪酸产生过氧化反应，导致丙二醛含量提高 (武永军等, 2009)。脯氨酸对植物起着保护和调节作用，其含量的增加有助于细胞或组织持水，对原生质起着保护作用与保水作用，同时参与植物体内活性氧自由基的清除，减少重金属对细胞膜和蛋白质造成的损伤，对稳定和保护大分子化合物有重要作用 (Israr et al, 2006)。

由图 2 可知：随着汞胁迫浓度的增加，四种牧草体内 MDA 含量均先下降后上升。达到最低值后，随汞胁迫浓度的升高，四种牧草 MDA 含量呈上升趋势，当汞胁迫浓度为 $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时，牧草中 MDA 含量依次为对照组的 1.34 倍、2.76 倍、1.24 倍和 1.55 倍。其中紫花苜蓿在汞胁迫下 MDA 含量变化幅度最大，四种牧草中受汞干扰最严重的为紫花苜蓿。

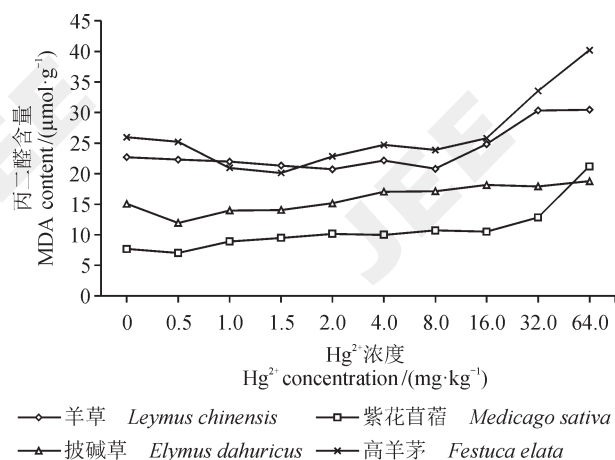


图 2 Hg^{2+} 胁迫对牧草丙二醛含量的影响
Fig.2 The effects of Hg^{2+} stress on the contents of MDA in herbage

由图 3 可知：在试验汞胁迫水平范围内，当汞

胁迫浓度为 $64.0 \text{ mg} \cdot \text{kg}^{-1}$ 时，四种牧草脯氨酸含量最高，分别为对照组的 7.48 倍、5.63 倍、3.42 倍和 4.02 倍。其中，随着汞胁迫浓度的升高，披碱草和高羊茅的脯氨酸含量先升高后略下降再急剧升高。脯氨酸含量升高，体现了植物抗氧化能力和渗透调节能力及对汞的耐受性，对植物具有保护作用。分析高浓度汞胁迫下脯氨酸含量的增长率，四种牧草中羊草对汞胁迫抗逆调节能力最强。

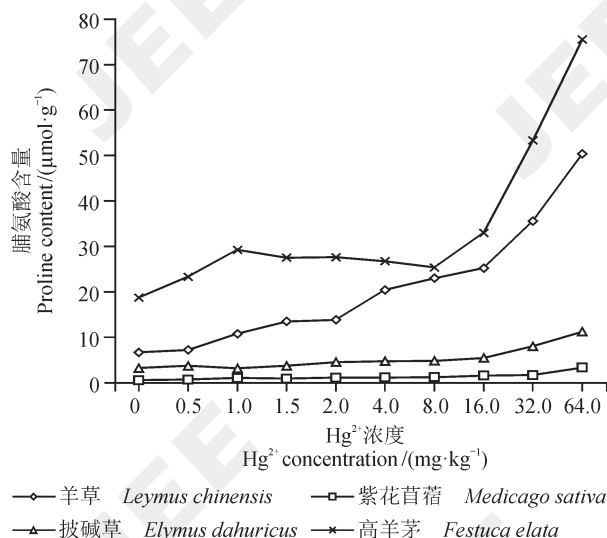


图 3 Hg^{2+} 胁迫对牧草脯氨酸含量的影响
Fig.3 The effects of Hg^{2+} stress on the contents of proline in herbage

3 结论

(1) 不同浓度汞胁迫对牧草中汞累积有显著影响，四种牧草对土壤中汞具有一定吸附转移能力，其中汞胁迫浓度较低时 ($< 16.0 \text{ mg} \cdot \text{kg}^{-1}$)，紫花苜蓿吸附转移能力最强；汞胁迫浓度增高后 ($> 16.0 \text{ mg} \cdot \text{kg}^{-1}$)，披碱草对土壤中汞吸附转移最强。四种牧草对汞富集系数随汞胁迫浓度增高而逐渐下降，其中羊草最大富集系数最高。

(2) 汞胁迫对不同牧草生理具有显著影响，当土壤中汞含量过高后，会对四种牧草生长产生毒害作用。在一定浓度汞胁迫作用下，牧草通过提高脯氨酸含量，促进抗氧化酶活性，减缓汞胁迫造成的活性氧升高及膜脂过氧化，达到平衡细胞渗透压，从而维持牧草正常生长所需环境。

内蒙古为产煤大省，煤电一体化经济发展模式推广的同时，要对草原生态进行长远的规划和

保护。面对与日俱增的燃煤发电量, 燃煤释汞量也将随之增加, 应采取适当措施, 对燃煤电厂烟气排放中的汞含量加以控制, 从源头制止燃煤电厂长期、低浓度汞排放对草原植被造成的影响。

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