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The response of glutathione reductase and glutathione-S-transferase to environmentally- and chemically-induced stress; amelioration by polyamines in leafy spurge (*Euphorbia esula* L.)

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Abstract:

Research is underway to establish key processes in the growth and development of leafy spurge that may be amenable to new and innovative methods for controlling this invasive weed that will be effective, cheaper and more environmentally friendly than present methods. These methods could be applied in the event that future circumstances render present methods ineffective. Basic physiology, biochemistry and genetics are areas of active research within our unit. Control of enzyme systems involved in the plant's response to stress (imposed by biotic or abiotic means) are possible candidates. Glutathione (GSH), a major constituent of all plant cells, consists of three amino acids linked together (a tri-peptide) that aids in controlling the plant's response to foreign chemicals and other stress, and behaves in many instances as an antioxidant. Glutathione reductase (GR) and glutathione S-transferase (GST) are key enzymes that regulate the action of GSH. The objective of our research is to characterize GR and GST in leafy spurge, determine their responses after induced stress of various kinds and establish ways to regulate them to the disadvantage of leafy spurge. In this report we also show that diclofop-methyl (DM) can be applied to leafy spurge to induce symptoms very similar to natural senescence. GR activity from leaves of untreated leafy spurge plants are consistently higher and somewhat more variable than GST activity (using 2,4-dichloronitrobenzene as a substrate). Activities ranged from approximately 120 to 170 nmol of product/min/mg protein for GR and approxi-

mately 50 to 120 nmol of product/min/mg protein for GST. Daily activities were more linear for GST than for GR. The activity of GR and GST from plants sprayed with 5 mM of the senescence-inducing compound DM increased within a few hours to nearly maximum levels by 26 h, and leveled off by 42 h. Activities of GR and GST from leafy spurge leaves treated 42 hrs with 5mM DM ranged from 250 to 320 nmol of product/min/mg protein and 175 to 2 10 nmol of product/min/mg protein, respectively. Polyamines are also natural constituents of plants that have many functions, most of which are still speculative, but are actively being investigated worldwide. Leafy spurge plants pre-treated 0.5 to 1 h with 20 mM polyamines (putrescine, spermidine or spermine) showed less visual damage when sprayed with 5 mM DM compared to plants sprayed with 5 mM DM alone. GR and GST activities were at essentially control levels in plants pre-treated with any one of the three polyamines, and less severe visual damage occurred to the leaves of the polyamine-treated plants than to plants sprayed only with DM. The increases in GR and GST activities induced by DM are similar to those of leafy spurge plants stressed by drought or iron deficiency. The ability of these polyamines to counteract the DM-induced activity of GR and GST suggest that they may play an important role in amelioration of the effects of biotic and abiotic stressors. Research is currently underway to attempt to establish whether these polyamines are functioning as anti-senescent agents; perhaps as antioxidants. We are still in the early stages of learning how to control the action of GSH, polyamines and their related constituents. This research should lead us forward in efforts to eventually control growth and development of leafy spurge and other perennial weeds.