Protein Analysis During the Ontogeny of Normal and Male Sterile Stamenless-2 Mutant Stamens of Tomato (Lycopersicon esculentum Mill.)

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The levels and synthesis of proteins during the ontogeny of normal and male sterile stamenless-2 (sl-2/sl-2) mutant stamens of tomato (Lycopersicon esculentum) were examined. The mutant stamens contained low levels of soluble protein which were related to reduction in protein synthesis. The mutant stamens, however, possessed many polypeptides similar to the normal and synthesized a 53-kd polypeptide at stages when there are abnormalities in tapetum development. The mutant stamens also possessed a 23-kd and some low molecular weight polypeptides that were considered as degradative proteins. Normal stamens exhibited the synthesis of many polypeptides not found in the mutant, from microspore mother cell to the preanthesis stages. In addition, at the time of pollen maturation there was a greater synthesis of several polypeptides, particularly those of 42 and 37 kd. Although the causative mechanisms of male sterility in the sl-2/sl-2 mutant are not known, the synthesis, and the lack, of specific polypeptides reported here appears to be associated with pollen degeneration.

KEY WORDS: male sterility; stamenless-2 mutant; proteins; Lycopersicon esculentum.

INTRODUCTION

The interest in male sterility of crop plants lies in its usefulness in the production of hybrid seed without the need of hand emasculation (Driscoll, 1986; Kaul, 1988). There has been considerable research, in different
systems, aimed at analyzing the various factors that affect male sterility. However, the mechanisms involved in the process of pollen degeneration are not fully understood.

The homozygous recessive, stamenless-2 (sl-2/sl-2) mutant of tomato (*Lycopersicon esculentum* Mill.) produces abnormal stamens and nonviable pollen. The mutant is temperature and gibberellin sensitive and produces normal viable pollen under low temperatures (18°C day/15°C night) or by gibberellic acid treatment (Sawhney, 1983; Sawhney and Greyson, 1973). The breakdown of microsporogenesis in mutant stamens becomes apparent after the tetrad stage. At that time, many microspores, devoid of a wall, become enlarged and degenerate, and others become highly vacuolate (Bhadula and Sawhney, 1987; Sawhney and Bhadula, 1988). However, abnormalities in the tapetum of mutant anthers were evident at the microspore mother cell (MMC) stage. Unlike the normal, some tapetal cells in the mutant anther divide and form a bilayer, whereas others enlarge considerably and protrude into the pollen sac (Sawhney and Bhadula, 1988).

In an attempt to determine whether male sterility in the sl-2/sl-2 mutant is associated with the appearance of specific proteins, we reported earlier that the mutant stamens at maturity were enriched in 23- and 31-kd polypeptides, and the normal in 75- and 37-kd polypeptides (Sawhney and Bhadula, 1987). Further, the low temperature-reverted mutant stamens lacked the 23- and 31-kd polypeptides, suggesting that these polypeptides may be associated with the expression of male sterility. The mutant stamens also contained a lower level of buffer-soluble protein than the normal stamens at maturity.

This study was undertaken to investigate (1) when during ontogeny the synthesis of various polypeptides occurs in normal and stamenless-2 mutant stamens and (2) whether the appearance of specific polypeptides correlates with structural aberrations in mutant stamens.

**MATERIALS AND METHODS**

**Seed Source and Plant Cultivation**

The seed source of normal (+/+) and stamenless-2 (sl-2/sl-2) mutant of tomato and the methods of plant cultivation were similar to those described earlier (Sawhney, 1983).

Plants were grown in a greenhouse with a supplemental lighting of fluorescent tubes and incandescent bulbs for 16 hr a day. The daily temperature in the greenhouse ranged from 21 to 26°C. Plants were fertilized biweekly with a commercial fertilizer 20:20:20 (Plant Products Co., Braemlae, Ontario).