CHAPTER 1
GENERAL INTRODUCTION

The orchid cut-flower is a lucrative export commodity in Malaysia, accounting for about RM 150 million, in revenue (Ministry of Agriculture, 2011). Generally, orchids have a long life span, reaching a period of 4 months in some species and hybrids. However, certain conditions, (e.g. pollination) reduce the shelf-life of orchids, from several months down to a few days. Therefore, if shelf life could be extended, this would assure better quality and commercial value for the sustainability of the orchid industry and to face competition in the international market.

Dendrobiums are among the popular orchids which contribute to the flower industry in Malaysia and are exported as both cut flowers and potted plants (Kosravi *et al.,* 2009). Several hybrids nevertheless, are highly sensitive to ethylene, both from exogenous and endogenous sources, resulting in the occurrence of ethylene-induced senescence which renders the flowers unmarketable. While exposure to exogenous ethylene can be avoided and controlled through means of packaging, effects of ethylene produced by the flowers as a response to pollination still exist.

According to Arditti (1979) the earliest description of post-pollination phenomena may have been described in 1793 by Conrad Sprengel. Thereafter, studies revolved around movement of cell contents as well as pollination-associated structures. One of the major discoveries was done by Hans Fitting, who was convinced that the substance initially reported as poisonous (Muller, 1868;1887) was in fact what he would call a *Pollenhormone.* This led to the studies on plant hormones which are known to have effects on orchids following pollination. Production of ethylene following pollination, leads to a cascade of pollination-induced symptoms in the flower resulting in changes in

morphology, ultra-structure, metabolism and gene expression (Arditti, 1969; Burg and Dijkman, 1967; Avadhani *et al.*,1971; Strauss and Koopowitz, 1973; Lim *et al.,* 1975*;* O'Neill *et al.,* 1993; Thongkum *et al.,* 2009). The collective effects of post pollination symptoms prepare the orchid flower for fertilization and embryogenesis (Zhang and O'Neill, 1993).

Eradication of the post-pollination symptoms either through the use of suitable additives or inhibitors as well as by gene manipulation would add value to these orchids. Ethylene inhibitors such as aminooxyacetic acid (AOA) and silver thiosulphate (STS) act by inhibiting ethylene effects by circumventing ethylene production or signalling. Significant delay in post pollination symptoms and increased longevity has been reported in orchids as well as other flowers treated with ethylene inhibitors (Aharono and Halevy, 1970; Nowak and Vacharotayan, 1980; Serek and Reid, 1995, Abdullah, 2005). Furthermore improvements in physiological aspects of flowers such as water relations, weight and colour have also been attributed to the application of ethylene inhibitors (Kusuhara, 1996; Loubaud and van Doorn, 2004). The use of glucose and sucrose has been used to improve vase life flowers as well as their keeping quality (Nair and Tung, 1980; Suisuwan, 1986; Liao *et al.,* 2000; Verlinden and Garcia, 2003).

The integral role of ethylene in regulating pollination-induced senescence means that inhibiting ethylene biosynthesis and signalling would effectively supress post pollination symptoms. The isolation and characterization of 1-aminocyclopropane-1-carboxylate oxidase (ACCO) and 1-aminoclopropane-1-carboxylate synthase (ACCS) genes involved in ethylene biosynthesis (Holdsworth *et al.,* 1987; Zarembinski and Theologis, 1994; O'Neill and Bui, 1998) and the five ethylene receptor genes found in *Arabidopsis* (Guzman and Ecker, 1990; Hua and Meyerowitz, 1998) were discoveries that successfully paved the road for future investigations.

Despite these developments, less attention has been devoted towards pollination induced senescence compared to leaf senescence and fruit ripening. A comprehensive approach is needed to understand this economically significantly phenomenon. The overall goal of this thesis was to establish a comprehensive understanding of pollination induced senescence to address the gaps in knowledge and the lack of information. This required the testing of several hypotheses:

Hypothesis I: That pollination reduces longevity and quality of *Dendrobium* Pompadour orchids.

Hypothesis II: That treatment solutions circumvents the symptoms of pollination-induced senescence.

Hypothesis III: That pollination induced senescence is accompanied by a decrease in both soluble and insoluble proteins.

This study was undertaken to investigate pollination-induced senescence in *D.* Pompadour and the effectiveness of treatment solutions in delaying/circumventing the phenomenon. The objectives of this thesis are as follows:

1. To profile morphological and physiological changes
2. To investigate changes in starch and carbohydrate status
3. To establish the changes in cell wall enzymes and membrane leakage
4. To establish changes in protein status and compare the protein profiles of pollinated and unpollinated flowers
5. To isolate and characterise selected ethylene biosynthesis and receptor genes from pollinated flowers