

# TABLE OF CONTENTS

	Page
<b>Summary</b> . . . . .	<b>iii</b>
<b>Acknowledgements</b> . . . . .	<b>xi</b>
<b>Table of Contents</b> . . . . .	<b>xiii</b>
<b>List of Figures</b> . . . . .	<b>xvii</b>
<b>List of Tables</b> . . . . .	<b>xxi</b>
<b>Abbreviations</b> . . . . .	<b>xxiii</b>
<b>Glossary of Mutants</b> . . . . .	<b>xxv</b>
<b>List of Genes</b> . . . . .	<b>xxvii</b>
<b>Introduction</b> . . . . .	<b>1</b>
Ovule initiation . . . . .	4
Gibberellins . . . . .	8
<i>Gibberellins and ovule development</i> . . . . .	11
Brassinosteroids . . . . .	13
<i>Brassinosteroids and ovule development</i> . . . . .	14
CUP-SHAPED COTYLEDON Proteins . . . . .	15
<i>CUC during gynoecium and ovule development</i> . . . . .	18
References . . . . .	19
<b>Objectives</b> . . . . .	<b>31</b>
<b>Chapter 1 Regulation of ovule initiation by gibberellins and brassi- nosteroids in Arabidopsis</b> . . . . .	<b>35</b>
Abstract . . . . .	37
Introduction . . . . .	38
Results . . . . .	40
<i>GAs act independently of BRs to control ovule initiation</i> . . . . .	40
<i>BRs promote an increase in GA levels in inflorescences</i> . . . . .	43
<i>GA promotion of ovule initiation does not rely on ANT</i> . . . . .	45
Discussion . . . . .	50

<i>Molecular mechanism to control ovule primordia formation by GAs and BRs</i> . . . . .	52
<i>ANT would not be related to BR or GA pathways in ovule initiation</i> . . . . .	53
Experimental procedures . . . . .	54
<i>Plant material assays</i> . . . . .	54
<i>Hormonal treatments and ovule number determination</i> . . . . .	54
<i>Construction of pANT:ANT-YPet and 35S:ANT and plant transformation</i> . . . . .	55
<i>qPCR analysis of gene expression</i> . . . . .	56
<i>In situ RNA hybridization</i> . . . . .	56
<i>Quantification of GAs</i> . . . . .	56
<i>Western blot analysis of GFP-RGA</i> . . . . .	57
Acknowledgements . . . . .	57
References . . . . .	57
Supplementary Information . . . . .	63
<i>Supplementary figures</i> . . . . .	63
<i>Supplementary tables</i> . . . . .	66
<b>Chapter 2</b> <b>Gibberellins regulate ovule number through a DELLA–CUC2 complex in Arabidopsis</b> . . . . .	<b>69</b>
Abstract . . . . .	71
Introduction . . . . .	72
Results . . . . .	75
<i>GAI requires CUC2 to mediate ovule number</i> . . . . .	75
<i>GAI interacts with CUC1 and CUC2</i> . . . . .	81
<i>DELLA and CUC2 are not reciprocally regulated</i> . . . . .	84
<i>CUC2 does not participate in BR-mediated ovule initiation</i> . . . . .	86
<i>Identification of GAI–CUC2 gene targets in ovule primordia initiation</i> . . . . .	87
Discussion . . . . .	90
<i>CUC1 and CUC2 interactions with GAI have different biological significances</i> . . . . .	91
<i>CUC and GAs/BRs in ovule initiation</i> . . . . .	91
<i>Working model of DELLA regulation of ovule primordia initiation</i> . . . . .	92
<i>GAI–CUC2 targets ovule primordia determination genes</i> . . . . .	94

Experimental procedures . . . . .	95
<i>Plant materials and growth conditions</i> . . . . .	95
<i>Generation of constructs and transgenic plants</i> . . . . .	96
<i>GA and PBZ treatments and determination of ovule number and         ovary length</i> . . . . .	96
<i>Histological procedures and confocal laser scanning microscopy</i> . . . . .	97
<i>Yeast two-hybrid assays</i> . . . . .	97
<i>qPCR analyses</i> . . . . .	98
<i>Co-IP assays</i> . . . . .	98
<i>ChIP, ChIP-Seq, and ChIP-qPCR analyses</i> . . . . .	99
Acknowledgements . . . . .	100
References . . . . .	101
Supplementary Information . . . . .	108
<i>Supplementary figures</i> . . . . .	108
<i>Supplementary tables</i> . . . . .	116
<b>General Discussion . . . . .</b>	<b>123</b>
Updated model for the control of ovule primordia initiation: the role of GAs . . . . .	125
Plausible hypotheses . . . . .	127
<i>From DELLA and CUC to CKs</i> . . . . .	127
<i>Fucosylation of DELLA proteins</i> . . . . .	129
<i>Downstream gene targets of DELLA in the placenta</i> . . . . .	130
References . . . . .	130
<b>Conclusions . . . . .</b>	<b>135</b>