

## Table of contents

List of tables: .....	1
List of figures: .....	3
List of abbreviations:.....	4
1. Introduction and literature review.....	6
1.1 General introduction.....	6
1.2 Symbiotic nitrogen fixation (SNF).....	8
1.3 Symbiotic N <sub>2</sub> fixation - a limiting process .....	9
1.3.1. Limitation of symbiotic nitrogen fixation by assimilate supply .....	11
1.3.2. Limitation of nitrogen fixation by oxygen diffusion into nodules .....	13
1.3.3. Regulation of nitrogen fixation through a N feedback effect .....	14
1.4. Indeterminate versus determinate nodules (pea and common bean) .....	15
1.5. Physiological role of phosphorus in legumes .....	17
1.6. Phosphorus deficiency .....	18
1.6.1 General effects on plant growth.....	18
1.6.2 Influence on nodule growth and functioning .....	20
1.7. Physiological regulation of symbiotic N <sub>2</sub> fixation under phosphorus limiting supply:.....	22
1.7.1 P deficiency affect plant energy conversion and assimilate formation resulting in assimilate limited nodules .....	22
1.7.2 Possible direct effects of P deficiency on nodules.....	23
1.7.3 N feedback regulation of nitrogen fixation under P deficiency .....	23
1.8 Objectives of the present work and principal layout of the experiments.....	24
2. Materials and Methods .....	26
2.1 Biological Materials .....	26
2.2 Phosphorus response experiments.....	26
2.2.1 Experimental design and plant growth .....	26
2.2.2 Phosphorus levels and application .....	27
2.2.3 Harvest procedure .....	29
2.3 Experiments on the effects of shoot manipulations on growth and nitrogen fixation under sufficient and limiting P supply.....	30
2.3.1 Plant growth and P levels .....	31
2.3.2 <sup>15</sup> N labeling of leaves .....	32
2.3.3 Leaf darkening and defoliation .....	33
2.3.4 Harvest procedures .....	34
2.4 Analytical methods .....	35
2.4.1 Dry matter determination .....	35
2.4.2 C, N and <sup>15</sup> N analyses .....	35
2.4.3 P analyses in plant material.....	36
2.4.4 Determination of amino acids, malate and sugars in nodules .....	36
2.4.5 Statistical analyses .....	37
3. Results .....	38

3.1 Experiments on the response of pea and bean plants in growth, nodulation and nitrogen fixation to increased phosphorus supply .....	38
3.1.1 Objective of the experiments .....	38
3.1.2 Plant dry matter .....	38
3.1.3 Nitrogen concentration in plants and nitrogen fixation.....	39
3.1.3.1 Nitrogen concentration in plants .....	39
3.1.3.2 Nitrogen fixation.....	40
3.1.4 P uptake and P concentration .....	42
3.1.4.1 P uptake.....	42
3.1.4.2 P concentration.....	43
3.1.5. Nodulation, nodule senescence and specific nitrogen fixation .....	44
3.1.6 Free amino acids (a.a.) in nodules .....	48
3.1.7 Organic acids in nodules .....	52
3.1.8 Sugars in nodules.....	53
3.2 Experiments on the effects of shoot manipulations on growth and nitrogen fixation under sufficient and limiting P supply:.....	55
3.2.1 Objective of the experiments .....	55
3.2.2 Dry matter increment and phosphorus concentration in the plants.....	55
3.2.3 %N concentration in plants and nitrogen fixation in dependence on the P treatments and imposed shoot manipulation in pea and bean plants .....	57
3.2.4 Influence of P treatments and shoot manipulations on nitrogen allocation from leaves to nodules .....	64
3.2.5 Influence of P treatments and shoot manipulations on free amino acids in nodules.....	67
4. Discussion .....	68
5. Summary: .....	75
References .....	79
Appendix .....	90
Acknowledgements .....	93
CURRICULUM VITAE .....	95