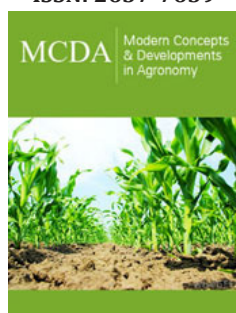


Genetics of Seed-Protein Content and Yield Related Traits in Pea (*Pisum sativum* L.)

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Abstract

Grain legumes are important for food security and nutritional value. They produce protein and protein rich foods for millions of people around the globe. They currently play a central role in sustainable agricultural production system by adding nitrogen nutrient to the soil for the subsequent crops largely through symbiosis with nitrogen-fixing soil bacteria. This sustainable provision of nitrogen fertilizer allows reducing the fertilization cost in cropping system and greenhouse gas emission. Pea (*Pisum sativum* L.) is one of the most important grain legumes which produce higher amount of seed protein and provides solution for protein deficiency and protein choices. It is also a good source of plant-based protein and has high nutritional value for human and animal consumption. With the increasing demand of plant-based protein, increasing pea seed yield and seed protein content (SPC) are the most important breeding targets today. Therefore, it is needed to elucidate the existing literature regarding genetics of pea. Here we review the existing potential quantitative and qualitative genetics underlying SPC and yield attributes in pea, currently used methods in pea breeding, and future direction for sustainable production.

Introduction

Protein-rich plant-based foods reduce detrimental health effects of millions of mothers and their children in low animal protein intake regions in the world [1]. In contrast, animal-based protein food sufficient countries such as Canada, USA, and UK are moving towards plant-based protein which creates a lot of pressure on crops with high protein content such as peas. Field pea (*Pisum sativum* L.) is one of the most important protein-rich legumes with some other agronomic benefits. It is an important green vegetable and the second most important grain legumes worldwide after common bean [2]. It provides dietary protein for millions of people globally [3]. Field pea also has some agronomic benefits such as nitrogen fixation through symbiosis with nitrogen-fixing bacteria resulting less use of nitrogen fertilizer in cropping system [4]. Due to these benefits field pea considers as a good source of protein and resources for farmers in marginal environments. Improving seed protein content (SPC) along with increased seed yield in dry peas is important to alleviate malnutrition in the world and facilitate moving towards plant-based protein food industry. However, a limited amount of research has been carried out on the genetics of SPC and its relation to beneficial agronomic traits for seed yield.

Pea is a cool-season grain legume belongs to Leguminosae family (Genus: *Pisum*, sub-family: *Faboideae* tribe: *Fabeae*). It is cultivated worldwide for both grains and vegetables with differences in morphology and nutritional status [5]. It is a self-pollinated diploid species ($2n=2x=14$) and its genome size (4300Mb) is 10 times larger than the model species *Medicago*

truncatula (500Mb). This expanded genome size in peas constitutes higher amount of repetitive DNA composing various families of mobile genetic elements [6]. In contrast, the exotic portion of genome is much lower in proportion to the whole genome compared to other legume species such as *M. truncatula*, *Lotus aponicas* (c. 472Mb) and chickpea (*Cicer arietinum* L.) (c. 740Mb). However, correlated and genetically programmed processes associated with SPC, and other agronomic traits have been reported in previous studies on legumes. A little is known about the molecular genetics in pea which limits the identification of favourable alleles and trait improvement through marker-assisted selection (MAS) breeding. The invention of high-throughput research tools and reducing cost of next generation sequencing provide durable solution to dissect the genetic architecture of model and non-model legume species [3,7]. Therefore, we can be able to bridge the knowledge gap between models and crops for trait improvement which is greatly applicable for improving SPC and yield-related agronomic traits in pea.

Quantitative genetics

Genetic parameters associated with quantitative traits such as number of loci, effect and sizes can be estimated through QTL analysis. Most of the parameters can be population specific resulting differences in identification of QTLs in different population. However, QTL analysis preceded by multiple populations in multi-environments is very common in genetic studies to determine stable QTL and correlation among traits. SPC and seed yield are complex traits and inherited mostly quantitatively. Burstin et al. [8] reported that QTLs associated with SPC, and seed yield are either consistent with plant source capacity controlling genes for production and fulfillment of seeds or seed sink strength controlling genes involved in synthesis of storage product and seed formation. Several loci associated with yield attributes and SPC has been identified in pea [8-13] (Table 1) and is currently being utilized in the respective research. However, the genetics basis of SPC and agronomic traits in peas is needed to be uncovered by more collaborative research worldwide.

Table 1: Detected QTLs associated with seed-protein content (SPC) and yield related traits in recombinant inbred lines (RILs) of pea.

Populations	Method Used	Identified QTLs/Meta QTLs	References
RILs	GenoPea 13.2 K SNP array	27	[9]
RILs	Iterative QTL mapping	261	[8]
RILs	Genotyping-by-sequencing	375	[10]
RILs	Genotyping-by-sequencing	89	[11]
RILs/F2	Bulked segregant analysis and selective genotyping	5	[12]
RILs	AFLP and SSR markers assay	36	[13]

Qualitative genetics

A few genes and markers associated with yield attributes and SPC has been identified in pea [8,14-17] (Table 2) and is currently being utilized in the variety development programmes. Starch-Branching Enzyme 1 of *rb* locus and ADP glucose-phosphorylase of *rb* locus are known to control wrinkled seed phenotype and impact seed development, yield and protein content [14,15]. Single gene such as *Le*, *Af* and *rugosus* have reported previously which are

involved in controlling plant architecture such as internode length, leaf type and seed size [8]. A subtilase gene *SBT1.1* controlling seed size by providing molecules that can act as signals to control cell division within the embryo [16]. Gali et al. [17] reported SNP markers linked to plant height, grain yield, seed starch and seed protein concentration in different chromosomes in a genome wide association study (GWAS), which have potential for marker-assisted selection towards rapid cultivar improvement.

Table 2: Identification of major genes/markers link to seed-protein content and yield related traits in pea.

Populations	Method Used	Identified Genes/Locus/Markers	References
BC11/IL2	Enzyme assay	<i>rb</i> locus	[15]
NIL3	Co-segregation	Starch-branching enzyme (SBEI)	[14]
IL/NIL	Near-infrared spectroscopy	<i>Le</i> and <i>Af</i>	[8]
EMS4 mutants	TILLING	<i>SBT1.1</i>	[16]
Diverse panel	Genotyping-by-sequencing	SNP markers	[17]

Concluding remarks and future perspective

Identification of causative polymorphism for SPC and yield attributes has been challenging in pea. Low resolution of classical quantitative genetics to dissect genetic architecture of SPC and agronomic traits in model and non-model species limits the use of this approach. Similarly, information about the genetic basis

of relationship between SPC and agronomic traits like seed yield, seed weight, days to flowering and growth habit are limited in pea. Therefore, it is essential to determine the genetic basis of trait correlation in pea for improving and stabilizing SPC while maintaining yield and other agronomic attributes. The future research directions are as follows:

- A. Develop mapping population and breeding lines utilizing available and promising germplasms.
- B. Utilize recently advanced technologies such as SNP, NGS etc. in breeding program to develop varieties with improved quality.
- C. Develop and execute research programmes towards identification of new QTLs/genes using all available germplasm globally.
- D. Develop new pea varieties and improve existing varieties based on different agro-ecological zones.
- E. Perform a meta-analysis of research on global pea production systems.
- F. Develop the best agronomic practices for pea based on different agro-ecological zones.
- G. Develop diverse pea cropping systems for increased the durability of existing QTLs/locus/genes.
- H. Develop integrated management systems that allow for increased the effectiveness of genetics and lowered the use of biocides.
- I. Evaluate the efficacy of tools and methods particularly large data analysis to improve genetic pool in the existing pea germplasm.
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