The assisted selection system (wang et al. 2011).

The key objectives in faba bean improvement program include resistance to biotic and abiotic factors, acclimatisation to targeted cropping regions and incorporation of better agronomic traits valued for higher grain production (Gnanasambandam et al. 2012; Sillero et al. 2010). Improvement in faba bean cultivars was made mostly through introduction, hybridization and selection. In Australia, faba bean production is threatened by fungal and viral diseases of which rust is the most commonly occurring disease. Faba bean rust is caused by the fungal pathogen Uromyces viciae-fabae and it is prevalent in major cultivation sites of northern New South Wales and southern Queensland (Qld) (Ijaz et al. 2016). Rust appears throughout the crop season, but early onset of disease can destroy up to 70-80% of crop (Liang 1986; Rashid and Bernier 1991).

Unlike cereals, none of the faba bean genotype has been reported immune to rust (Garcia et al. 2008; Miklas et al. 2006). However, visible necrosis was observed in some cases (Adhikari et al. 2016; Sillero et al.

2000) and it reduces disease severity. Uromyces viciae-fabae is a true biotrophic organism without any alternative host (Macleod and Galloway 2015). Faba bean is a diploid (2n = 12, genome size 13 Gb) plant species with six homologous pairs of chromosomes, out of which, five are acrocentric and one is metacentric (Bennett and Smith 1991) and it has huge proportion of repetitive nucleotide sequences (Flavell et al. 1974). The large genome size is causing an impediment towards genomic studies and associated marker development for an effective marker assisted selection system (Wang et al.

2011). However, recent revolution in the next generation sequencing have overcome hurdles and genomic regions conferring resistance against broomrape (Díaz-Ruiz et al. 2010; Gutiérrez et al. 2013), ascochyta blight (Atienza et al.

2016; Kaur et al. 2014; Ocaña et al. 2015) and various yield related agronomic traits (Khazaei et al.

2014) in faba bean have been identified. Several sources of rust resistance in faba bean have been reported from Spain (Sillero et al. 2011) and Australia (Adhikari et al. 2016; Ijaz and Adhikari 2016), but no information is available on linked markers for marker-assisted selection (MAS). A rust resistance gene, named Uvf-1, was mapped using Randomly Amplified Polymorphic DNA (RAPD) markers by Avila et al. (2003). Linkage mapping represents the relative location of various genetic markers present on the chromosome of an organism determined through markers recombination (Singh and Singh 2015).

It is the powerful tool that underpin closely linked DNA markers responsible for controlling a particular phenotype (Collard and Mackill 2008). The application of these markers to improve selection efficiency in any breeding program is highly desirable, because it allows surpassing of expensive and time consuming phenotypic tests, particularly when dealing with genes producing intermediate responses. Among genetic markers, SSRs were the most commonly used marker system for many plant species. However, during the past few years, SSRs have been replaced by the application of high-throughput Single Nucleotide Polymorphism (SNP) markers in molecular

breeding due to the availability of whole genome sequences for many plant species (Hiremath et al. 2012). RNA sequencing (RNA-Seq) or de novo transcriptome assembly has maximized the capacity of marker identification in plants species where genetic studies have been limited either due to large genome size or the absence of whole genome sequences, therefore became a method of choice in legume crops such as field pea (Franssen et al. 2011), yellow lupin (Lupinus luteus L.

) (Parra-González et al. 2012), lentils (Verma et al. 2013), chickpea (Cicer arientinum L.) (Garg et al.

2011) and faba bean (Kaur et al. 2014; Ocaña et al. 2015). These technologies underpin discovery of trait-linked SNP markers which are co-dominant in nature and amenable to high throughput system for selection in crop improvement programs (Wang et al. 2009). Two quantitative trait loci (QTL) q_rust_Doza and q_rust_Ac1655 was identified respectively in Fiord/Doza#12034 and Fiord/Ac1655 F4 populations using Illumina 1536 GoldenGate SNP array by Sudhesh et al. (2016), but the resistance linked markers were not converted to user friendly format for marker-assisted selection.

The objectives of this study were; 1) characterisation of seedling rust resistance in Fiord/Doza#12034 and Fiord/Ac1655 recombinant inbred line (RIL) populations, 2) development of PCR based KASP markers from previously reported SNP markers by Sudhesh et al. (2016), 3) development of linkage map from RNA-Seq data of Fiord/Doza#12034 RIL population to saturate resistance locus (q_rust_Doza) and 4) validation of flanking SNP

markers (using KASP technology) on the set of local and exotic cultivars/genotypes to illustrate their utility in marker-assisted breeding.