

## Supplementary material:

**Table 1:** Reduction in redundancy

Total no. of ESTs	No. of ESTs forming contigs (%)	No. of contigs	No. of singletons (%)	No. of assembled sequences	Reduction in Redundancy (%)
25,495	22652 (88.84%)	2843	7001 (27.46%)	9844	61.39%

**Table 2:** Gene ontology based functional annotation and classification of Contigs-SSRs of *Humulus Lupulus*.

Gene ontology (Biological process)	SSRESTs (numbers)	Gene ontology (Molecular function)	SSR-ESTS (number)	GeneOntology(Cellular Component)	SSRESTs (numbers)
Abscisic acid biosynthetic process	1	Acetate-coa ligase activity	1	Actin cytoskeleton	1
Abscisic acid mediated signalling pathway	2	Acetolactate synthase activity	1	Anchored to membrane	1
Acetyl-coa biosynthetic process from pyruvate	2	Acid phosphatase activity	1	Apoplast	18
Actin filament depolymerization	1	Acyl carrier activity	2	ATP binding cassette transporter	1
Actin nucleation	1	Acyl-coa dehydrogenase activity	1	Catalytic activity	1
Actin polymerization	1	Adenylylsulfate kinase activity	3	Cell plate	1
Activation of protein kinase C activity by GPCR	1	Alanine-glyoxylate transaminase activity	1	Cell surface	1
Aerobic respiration	3	Alcohol dehydrogenase(nad) activity	1	Cell wall	43
Aging	2	Allene-oxide cyclase activity	1	Central vacuole	2
Amino acid transport	2	Alpha-amylase activity	2	Chloroplast	73
Ammonia assimilation cycle	3	Alpha-galactosidase activity	1	Chloroplast envelope	2
Anther dehiscence	2	Amino acid transmembrane transporter	3	Chloroplast photosystem II	5
Anthocyanin metabolic process	1	Amino acid binding	2	Chloroplast thylakoid membrane	6
Anti-apoptosis	1	Ammonia-lysate activity	1	CUL4 RING ubiquitin ligase complex	1
Arginine process to glutamate	1	Anchored to membrane	1	Cytoplasm	12
Aromatic amino acid family biosynthetic process	2	Antioxidant activity	1	Cytoplasmic large ribosomal unit	6
Asparagine biosynthetic process	2	Asparagine synthase	1	Cytosol	56
ATP biosynthetic process	1	Atp binding	28	Cytosolic large ribosomal unit	10
ATP synthesis coupled proton transport	14	Atpase activity	9	Cytosolic small ribosomal subunit	8
Auxin mediated signalling	3	Beta-amylase activity	1	Early endosome membrane	1
Auxin polar transport	2	Beta-galactosidase activity	1	Endomembrane system	33
Base-excision repair	1	Binding	6	Endoplasmic reticulum	10
Brassinosteroid biosynthetic process	11	Caffeate o-methyltransferase activity	2	Extracellular region	1
Calcium mediated signalling	6	Calcium channel activity	1	Extrinsic to membrane	1
Carbohydrate biosynthetic process	1	Calcium ion binding	5	Extrinsic to vacuolar membrane	1
Carbohydrate metabolic process	6	Calmodulin binding	5	Integral to membrane	3
Carboxylic acid metabolic process	1	Carbohydrate tranmembrane transporter	1	Intracellular	2
Carotenoid biosynthetic process	3	Carbon sulfur lyase activity	2	Intrinsic to endoplasmic reticulum	1
Cell death	1	Carboxypeptidase activity	1	Membrane	8
Cell proliferation	1	Catalytic activity	6	Mitochondrial respiratory complex	7
Cell redox homeostasis	7	Cellulose synthase activity	2	Mitochondrion	8
Cell wall loosening	1	Chalcone isomerase activity	2	Nuclear envelope	1
Cell wall modification	3	Chaperone binding	1	Nuclear speck	2
Cell wall organization	2	Chitin binding	1	Nucleic acid binding	1
Cell wall thickening	1	Chitinase activity	1	Nucleolus	3
Cellular copper ion homeostasis	1	Chloroalyl aldehyde dehydrogenase	2	Nucleosome	1
Cellular iron homeostasis	1	Chlorophyll binding	1	Nucleus	15
Cellular metabolic process	1	Chromatin dna binding	1	Peroxisome	1
Cellular respiration	1	Cobalt ion binding	1	Plasma membrane	2
Cellular response to cold	2	Conjugate hydrolase activity	1	Plastid large ribosomal unit	1
Cellular response to ethylene stimulus	1	Copper chaperone activity	4	SCAR complex	1
Cellular response to fatty acid	1	Copper ion binding	9	Vacuole	3
Cellular response to nitrogen starvation	1	Cyclic nucleotide binding	2		
Cellular response to phosphate starvation	1	Cyclin-dependent protein kinase reg.	1		
Cellular response to selenium ion	3	Cystathionine beta-lyase activity	1		
Cellular response to water deprivation	2	Cysteine synthase activity	1		
Chlorophyll biosynthetic process	2	Cysteine-type endopeptidase activity	2		
Chloroplast thylakoid membrane	1	Cytochrome-b5 reductase activity	2		
Cinnamic acid biosynthetic process	3	Diacylglycerol kinase activity	1		
Copper ion transport	1	Dihydrolate reductase activity	1		
Cytokinesis	1	Dna binding	15		
De-etiolation	1	Dna photolyase activity	1		
Defense response	3	Dna-directed rna polymerase activity	2		
Defense response to bacterium	4	Double-stranded rna binding	4		
Development process	2	Electron carrier activity	5		
DNA duplex unwinding	1	Endopeptidase inhibitor activity	1		
DNA mediated transformation	1	Enzyme inhibitor activity	1		
DNA repair	1	Epoxide hydrolase activity	1		
Double-strand break repair	1	Fatty-acyl-coa binding	1		
Embryo development in seed dormancy	14	Fatty-acyl-coa reductase activity	1		
Endocytic recycling	1	Fk506 binding	1		
Entrainment of circadian clock	1	Flavin adenine dinucleotide binding	1		
Ethylene biosynthetic process	1	Glutamate synthase(nadh) activity	2		
Extracellular transport	1	Glutathione transferase activity	1		
Fatty acid beta-oxidation	1	Glycerol-3-phosphate o-acyltransferase	1		
Fatty acid biosynthetic process	4	Gtp binding	8		
Fatty acid metabolic process	1	Heme binding	6		
Fatty acid catabolic process	1	Histidine-trna ligase activity	1		

Flavonoid biosynthetic process	5	Hydrogen ion transporting atp synthase	4
Gene silencing	2	Hydrolase activity	1
Glycolipid transport	1	Inorganic anion transporter activity	1
Heat acclimation	1	Intracellular cyclic nucleotide activated	1
Heme biosynthetic process	1	Inositol monophosphatase activity	1
Histidine biosynthetic process	1	Isopentenyl-diphosphate delta-isomerase	1
Histone deacetylation	1	Kinase activity	4
Histone H2B ubiquitination	1	Lipase activity	1
Hyperosmotic salinity response	5	Lipid binding	5
Intracellular Protein Transport	1	Lipoxygenase activity	1
Iron-sulfur cluster assembly	1	L-malate dehydrogenase activity	1
Lateral root development	1	Long-chain fatty acid-coa ligase activity	1
Lignin biosynthetic process	3	L-tyrosine:2-oxoglutarate amino-transf.	1
Lipid storage	4	Manganese ion binding	6
Lipid transport	5	Map kinase activity	1
L-methionine salvage from methylthioadenosine	2	Metal ion binding	5
L-phenylalanine biosynthetic process	2	Metalloendopeptidase activity	1
Malate metabolic process	1	Mitochondrial respiratory chain complex	2
Male gamete generation	2	Mrna binding	1
Male meiosis	2	Nucleic acid binding	4
Metabolic process	4	Nucleocytoplasmic transporter activity	1
Methionine biosynthetic process	2	Nucleotide binding	1
Microgametogenesis	1	Nucleotide-sugar transmembrane transp.	1
Mrna transcription	1	Oxidoreductase activity	6
Multicellular organismal development	3	Pantothenate kinase activity	1
Negative regulation of flower development	1	Peptidyl-prolyl cis trans isomerase	1
Negative regulation of transcription,DNA-dependent	1	Phenylalanine ammonia-lyase activity	1
Negative regulation of plant-type hypersensitive resp.	1	Phosphatidylinositol-4,5-biophosphate bi	2
Nitrate assimilation	1	Phosphoglycerate dehydrogenase activity	1
Nuclear mrna splicing,via spliceosome	3	Phosphoric diester hydrolase activity	1
Nucleosome assembly	3	Polygalacturonase activity	1
Nuclear-transcribed mrna poly(A) tail shortening	1	Polynucleotide 5'-hydroxyl-kinase	1
Nucleocytoplasmic transport	1	P-p-bond-hydrolysis-driven protein	1
Organ growth	1	Polygalacturonate 4-alpha-galacturosyl-tr	1
Oxidation-reduction process	14	Potassium ion transmembrane	1
Pentose-phosphate shunt	1	Proline dehydrogenase activity	1
Peptidyl-proline modification	1	Protein binding	33
Phenylalanine biosynthetic process	1	Protein disulfide oxidoreductase activity	3
Phosphatidyl Glycerol biosynthetic process	1	Protein disulfide isomerase activity	1
Phosphatidylcholine biosynthetic process	3	Protein homodimerization activity	1
Photosynthesis	9	Protein transporter activity	2
Photosystem II stabilization	2	Protein serine/threonine phosphatase	1
Pollen tube growth	1	Purine base transmembrane transporter	2
Pollen development	1	Rrna-binding	7
Protein folding	15	Sequence specific dna binding	21
Protein O- linked glycosylation	1	Serine o-acetyltransferase activity	1
Protein phosphorylation	5	Serine racemase activity	1
Protein targeting to vacuole	1	Serine-type endopeptidase inhibitor	2
Protein transport	1	Sequence-specific signalling pathway	1
Protein ubiquitination involved in ubiquitin-dependent	1	Signal transducer activity	2
Proteolysis	3	Small conjugating proten ligase activity	2
Proton transport	2	Snorna binding	1
Regulation of transcription,DNA-dependent	2	Steroid binding	1
Regulation of cell cycle	1	Strictosidine synthase activity	1
Regulation of programmed cell death	1	Structural constitute of ribosome	35
Respiratory electron transport chain	1	Succinate dehydrogenase (ubiquinone)	1
Response to auxin stimulus	1	Sugar-hydrogen symporter activity	1
Response to cadmium ion	4	Transferase activity	3
Response to gibberellin stimulus	1	Translation initiation factor activity	1
Response to high light intensity	1	Transmembrane transporter activity	2
Response to hypoxia	1	Triglyceride lipase activity	1
Response to karrikin	4	Triose-phosphate isomerase activity	1
Response to metal ion	9	Ubiquinol-cytochrome-c reductase	1
Response to nematode	2	Ubiquitin binding	1
Response to other organism	2	Ubiquitin protein ligase activity	1
Response to salicylic acid stimulus	1	Uracil phosphoribosyltransferase activity	1
Response to salt stress	1	Vacuole	1
Response to stress	1	Water channel activity	1
Response to water deprivation	4	Zinc ion binding	6
Ribosome biogenesis	32	1,3-beta-d-glucan synthase activity	1
RNA metabolic process	1	1-acylglycerol-3-phosphate o-acyltrn	1
Starch biosynthetic process	1	9-cis-epoxycarotenoid dioxygenase activity	1
Sterol biosynthetic process	1	Inorganic anion transporter activity	1
Transcription,DNA dependent	1	Intracellular cyclic nucleotide activated	1
Translation	25	Inositol monophosphatase activity	1
Transmembrane transport	2	Isopentenyl-diphosphate delta-isomerase	1
Transport	3	Kinase activity	4
Ubiquitin-dependent protein catabolic process	3	Lipase activity	1
Vesicle-mediated transport	1	Lipid binding	5
10-formyltetrahydrflormate biosynthetic process	1	Lipoxygenase activity	1

**Table 3:** Gene ontology based functional annotation and classification of Singlet-SSRs of *Humulus Lupulus*

Gene ontology(Biological process)	SSR-ESTs (numbers)	Gene ontology(Molecular function)	SSR-ESTs (numbers)	Gene ontology (Cellular component)	SSR-ESTs (numbers)
Abscisic acid mediated signalling pathway	3	Acetate-coa ligase activity	1	Anchored to membrane	4
Acetate metabolic process	1	Acetolactate synthase activity	1	Apoplast	14
Actin cytoskeleton organization	3	Acid phosphatase activity	1	Axon	1
Activation of protein kinase C	1	Acyl carrier activity	2	CCAAT-binding factor complex	1
Aerenchyma formation	1	Acyl-coa dehydrogenase activity	1	Cell surface	1
Aerobic respiration	1	Adenylylsulfate kinase activity	3	Cell wall	23
Aging	4	Alanine-glyoxylate transaminase activity	1	Cellular bud neck	1
Aluminium ion transport	2	Alcohol dehydrogenase(nad) activity	1	Chloroplast	78
Amino acid catabolic process	1	Allene-oxide cyclase activity	1	Chloroplast photosystem II	6
Amino acid import	1	Alpha-amylase activity	2	Chloroplast stroma	1
Amino acid transport	1	Alpha-galactosidase activity	1	Chloroplast stromal thylakoid	1
Ammonia assimilation cycle	2	Amino acid transmembrane transporter	3	Chloroplast thylakoid lumen	2
Anther dehiscence	2	Amino acid binding	2	Chromosome	1
Anti-apoptosis	1	Ammonia-lyase activity	1	Clathrin vesicle coat	1
Asparagine biosynthetic process	1	Anchored to membrane	1	CULA RING ubiquitin ligase complex	2
ATP synthesis coupled proton	4	Antioxidant activity	1	Cytoplasm	21
Auxin mediated signaling pathway	3	Asparagine synthase	1	Cytosol	38
Auxin polar transport	2	Atp binding	28	Cytosolic large ribosomal subunit	6
Biosynthetic process	4	Atpase activity	9	Cytosolic small ribosomal subunit	5
Branched chain family amino acid biosynthetic	1	Beta-amylase activity	1	DNA-directed RNA polymerase II,core	1
Brassinosteroid biosynthetic process	5	Beta-galactosidase activity	1	Endosome membrane	4
Brassinosteroid mediated signalling pathway	3	Binding	6	Extracellular space	3
Cadmium ion transport	1	Caffeate o-methyltransferase activity	2	FACT complex	1
Calcium-mediated signalling	5	Calcium channel activity	1	Golgi membrane	4
Carbohydrate metabolic process	1	Calcium ion binding	5	Integral to membrane	4
Carbohydrate biosynthetic process	1	Calmodulin binding	5	Intracellular	4
Carbohydrate metabolic process	7	Carbohydrate transmembrane transporter	1	Intracellular cyclic nucleotide activated	1
Carbohydrate transport	2	Carbon sulfur lyase activity	2	Membrane	11
Carbon fixation	1	Carboxypeptidase activity	1	Mitochondrial matrix	2
Carboxylic acid metabolic process	1	Catalytic activity	6	Mitochondrial respiratory chain comp.	5
Carotene catabolic process	1	Cellulose synthase activity	2	Mitochondrion	12
Cation transport	1	Chalcone isomerase activity	2	Nuclear speck	2
Cell death	1	Chaperone binding	1	Nucleus	35
Cell fate specification	2	Chitin binding	1	Peroxisome	1
Cell fate specification	1	Chitinase activity	1	Plant-type cell wall	3
Cell redox homeostasis	3	Chloroalyl aldehyde dehydrogenase	2	Plasma membrane	21
Cell tip growth	1	Chlorophyll binding	1	Plasmodesma	4
Cell wall modification	1	Chromatin dna binding	1	Plastid	1
Cell wall thickening	3	Cobalt ion binding	1	Serine/ threonine phosphatase complex	3
Cellular iron ion homeostasis	7	Conjugate hydrolase activity	1	Ubiquitin-protein ligase activity	2
Cellular localization	1	Copper chaperone activity	4	Vacuole membrane	2
Cellular metabolic process	3	Copper ion binding	9	1,3-beta-D-glucan synthase complex	1
Cellular respiration	1	Cyclic nucleotide binding	2		
Cellular response to cold	1	Cyclin-dependent protein kinase regulator	1		
Cellular response to ethylene stimulus	5	Cystathionine beta-lyase activity	1		
Cellular response to hydrogen peroxide	1	Cysteine synthase activity	1		
Cellular response to iron ion	1	Cysteine-type endopeptidase activity	2		
Cellular response to nitrogen starvation	1	Cytochrome-b5 reductase activity	2		
Cellular response to phosphate starvation	1	Diaclylglycerol kinase activity	1		
Cellular response to sulfate starvation	1	Dihydrolate reductase activity	1		
Chlorophyll biosynthetic process	1	Dna binding	15		
Chloroplast organization	3	Dna photolyase activity	1		
Cholesterol metabolic process	1	Dna-directed rna polymerase activity	2		
Coenzyme A biosynthetic process	1	Double-stranded rna binding	4		
Cold acclimation	1	Electron carrier activity	5		
Cotyledon development	2	Endopeptidase inhibitor activity	1		
Cyanide catabolic process	2	Enzyme inhibitor activity	1		
Cytochrome complex assembly	1	Epoxide hydrolase activity	1		
Cytochrome complex assembly	1	Fatty-acyl-coa binding	1		
Cytokinin transport	1	Fatty-acyl-coa reductase activity	1		
De-etiolation	1	Fk506 binding	1		
Defense response	2	Flavin adenine dinucleotide binding	1		
Defense response to bacterium	8	Glutamate synthase(nadh) activity	2		
Defense response to fungus	2	Glutathione transferase activity	1		
Defense response to insect	1	Glycerol-3-phosphate o-acyltransferase	1		
DNA duplex unwinding	1	Gtp binding	8		
DNA repair	2	Heme binding	6		
DNA replication	1	Histidine-trna ligase activity	1		
DNA topological change	1	Hydrogen ion transporting atp synthase	4		
Electron transport chain	1	Hydrolase activity	1		
Embryo development	5	Inorganic anion transporter activity	1		
Embryo development ending in seed dormancy	6	Intracellular cyclic nucleotide activated	1		
Embryo sac development	1	Inositol monophosphatase activity	1		
Endocytic recycling	2	Isopentenyl-diphosphate delta-isomerase	1		
Entrainment of circadian clock	1	Kinase activity	4		
Establishment of cell polarity	5	Lipase activity	1		
Ethylene mediated signalling pathway	1	Lipid binding	5		
Far-red light signalling pathway	1	Lipoxygenase activity	1		
Fatty acid biosynthetic process	3	L-malate dehydrogenase activity	1		
Fatty acid metabolic process	1	Long-chain fatty acid-coa ligase activity	1		
Fatty-acyl-coa biosynthetic process	1	L-tyrosine:2-oxoglutarate aminotransferase	1		
Flavonoid biosynthetic process	5	Manganese ion binding	6		
G1 phase of mitotic cell cycle	1	Map kinase activity	1		
Growth	1	Metal ion binding	5		
Heat acclimation	1	Metalloendopeptidase activity	1		
Histidyl-trna aminoacylation	1	Mitochondrial respiratory chain complex	2		
Hyperosmotic salinity response	8	Mrna binding	1		
Inositol triphosphate metabolic process	2	Nucleic acid binding	4		
Intracellular protein transport	2	Nucleocytoplasmic transporter activity	1		

Ion transport	1	Nucleotide binding	1
Jasmonic acid biosynthetic process	2	Nucleotide-sugar transmembrane	1
Lewis a epitope biosynthetic process	1	Oxidoreductase activity	6
Lignin biosynthetic process	3	Pantothenate kinase activity	1
Lipid metabolic process	2	Peptidyl-prolyl cis trans isomerase activity	1
Lipid storage	3	Phenylalanine ammonia-lyase activity	1
Lipid transport	2	Phosphatidylinositol-4,5-biophosphate	2
L-methionine biosynthetic process	1	Phosphoglycerate dehydrogenase activity	1
L-serine biosynthetic process	1	Phosphoric diester hydrolase activity	1
Male gamete generation	3	Polygalacturonase activity	1
Meiotic chromosome segregation	1	Polynucleotide 5'-hydroxyl-kinase activity	1
Metabolic process	6	P-p-bond-hydrolysis-driven protein	1
Mrna modification	1	Polygalacturonate 4-alpha- related	1
Multicellular organismal development	5	Potassium ion transmembrane transporter	1
Negative regulation of abscisic acid mediated	1	Proline dehydrogenase activity	1
Negative regulation of transcription,DNA	1	Protein binding	33
Nuclear mrna splicing,via spliceosome	3	Protein disulfide oxidoreductase activity	3
Nucleocytoplasmic transport	2	Protein disulfide isomerase activity	1
Nucleotide-excision repair	1	Protein homodimerization activity	1
Oxidation-reduction process	9	Protein transporter activity	2
Peptidyl-histidine phosphorylation	1	Protein serine/ threonine phosphatase	1
Peptidyl-proline modification	1	Purine base transmembrane transporter	2
Phosphatidylglycerol biosynthetic process	1	Rrna-binding	7
Photoinhibition	1	Sequence specific dna binding	21
Photoreactive repair	1	Serine o-acetyltransferase activity	1
Photorespiration	1	Serine racemase activity	1
Photosynthesis	11	Serine-type endopeptidase inhibitor	2
Plant-type cell wall loosening	1	Sequence-specific signalling pathway	1
Plant-type hypersensitive response	2	Signal transducer activity	2
Pollen germination	3	Small conjugating proten ligase activity	2
Pollen sperm cell differentiation	1	Snorna binding	1
Porphyrin-containing compound metabolic process	1	Steroid binding	1
Post-translational protein modification	2	Strictosidine synthase activity	1
Potassium ion transport	3	Structural constitute of ribosome	35
Production of ta-sirnas involved in RNA	1	Succinate dehydrogenase (ubiquinone)	1
Protein folding	12	Sugar:hydrogen symporter activity	1
Protein import into chloroplast thylakoid	1	Transferase activity	3
Protein phosphorylation	14	Translation initiation factor activity	1
Protein targeting to mitochondrion	1	Transmembrane transporter activity	2
Protein targeting to vacuole	1	Triglyceride lipase activity	1
Protein transport	1	Triose-phosphate isomerase activity	1
Protein ubiquitination involved in ubiquitin-	5	Ubiquinol-cytochrome-c reductase	1
Proteolysis	3	Ubiquitin binding	1
Purine base transport	1	Ubiquitin protein ligase activity	1
Regulation of abscisic acid mediated signalling	1	Uracil phosphoribosyltransferase activity	1
Regulation of cell cycle	1	Vacuole	1
Regulation of double fertilization forming a zygote	1	Water channel activity	1
Regulation of transcription,DNA-dependent	13	Zinc ion binding	6
Regulation to translation	2	1,3-beta-d-glucan synthase activity	1
Response to abscisic acid stimulus	2	1-acylglycerol-3-phosphate o-	1
Response to auxin stimulus	4	9-cis-epoxycarotenoid dioxygenase activity	1
Response to brassinosteroid stimulus	1		
Response to cadmium ion	1		
Response to cold	1		
Response to endoplasmic reticulum stress	1		
Response to far red light	1		
Response to jasmonic acid	2		
Response to karrikin	4		
Response to metal ion	2		
Response to oxidative stress	1		
Response to phosphate starvation	1		
Response to salt stress	3		
Response to stress	3		
Response to water deprivation	1		
Response to nematode	1		
Ribosome biogenesis	14		
RNA methylation	1		
RNA splicing	2		
Serine family amino acid metabolic process	1		
Small gtpase mediated signal transduction	2		
Sterol biosynthetic process	1		
Sulfate assimilation	1		
Transcription,DNA-dependent	3		
Translation	14		
Transmembrane protein	2		
Transport	1		
Tricarboxylic acid cycle	2		
Trichome branching	1		
Trna splicing	1		
Ubiquitin-dependent protein catabolic process	1		
Vegetative to reproductive phase transition	1		
(1->3)-beta-D-glucan biosynthetic process	1		
10-formyltetrahydrofolate biosynthetic process	1		