Certification

The effects of the oligosaccharides and *Ascophyllum nodosum* extract on soybean gene expression

By

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ABSTRACT

THE EFFECTS OF OLIGOSACCHARIDES AND *ASCOPHYLLUM NODOSUM* EXTRACT ON SOYBEAN GENE EXPRESSION

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To understand the underlying mechanisms and bioactive components within ANE, the effects of ANE and its compositional oligosaccharides on the soybean transcriptome profile were compared. All treatments commonly up-regulated various pathogenesis-related genes and vital plant secondary metabolic processes, like flavonoid and terpenoid biosynthesis, which possess anti-microbial properties. Genes for catabolic processes were also up-regulated in all treatments except for oligo-alginate. Unexpectedly, all treatments except oligo-alginate coincidentally inhibited genes in various energy consuming processes, such as protein, lipid, fatty acid and DNA synthesis; inhibition of, processes involved in cell cycle, cell growth and division were also observed. These decreases in energy consuming processes, and enhancement of energy producing processes and stress responses, are likely produced via the activation of SnRK1 (sucrose non-fermenting related kinase1) or deactivation of TOR (target of rapamycin) pathways.

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Contents

1 Demands for improvement on crop productivity

1.1 The challenge to feed ten billion people

There is a growing trend showing a large number of people changing their diets wherein animal proteins are increasing as a proportion of people’s total protein intake (FAOSTAT, 2015). However, for most animal foods derived from livestock, such as milk, eggs, chicken, pork and beef, the protein conversion efficiency is still quite low (Smil, 2002). This means that to satisfy this higher demand for meat and meat products, there is a need to sharply increase crop production for the feeding of said livestock. This dietary transition has already occurred in developed countries. For instance, in the industrialized nations of the West, average protein intakes are generally excessive, whereas protein intake is still inadequate for millions of people in Asia, Africa and Latin America (Smil, 2002). Data from FAOSTAT (Food and Agriculture Organization of the United Nations Statistics Division) show that although the global undernourishment has been declining, as of 2015, 10.8% of the world’s population were still suffering from malnutrition because of a lack of access to affordable food. Although the adequacy of the average dietary supply has been slowly increasing since 1961, simultaneously, domestic food prices have been increasing (FAOSTAT, 2015). This, suggests that millions of lower income people still have insufficient access to the food supply. It has been predicted that in the next half century, the global population will approach ten billion (FAOSTAT, 2015). To feed ten billion people, whilst satisfying these dietary changes, providing proper food access to poor people and at the same time minimizing the detrimental effects of agriculture on the environment will be extremely challenging, requiring the use of rigorously sustainable and productive agricultural practices.

Scientists haves estimated that crop production will need to be doubled to meet this challenge (Foley et al., 2011). Human agriculture has markedly influenced the natural environment almost from its inception and at present, account for approximately 38% of earth's terrestrial surface, is the largest use of land on the planet (Foley et al., 2011). Humanity is always trying out new methods to meet its fundamental needs, the agricultural revolution of recent decades is a good example of this and has greatly increased agricultural output. For example, it is thought that at least 30 to 50% of crop yields are due to commercial fertilizers' nutrient input (Stewart, Dibb, Johnston & Smyth, 2005), regardless of the frequent criticism when discussing their effects on the environment. In the United States, on average, fertilizer takes up to one-third of the energy put into crop production (Amenumey & Capel, 2014). Considering the urgent demand for increased crop production, the unavoidable detrimental effects of inorganic fertilizers and limited agricultural land availability, it is proposed that 90% of the future growth in crop production will be generated via higher yields and increased cropping intensity, with the remainder coming from land expansion (Bruinsma, 2009). Thus, enhancing crop productivity in an environmentally sustainable way is crucial.

1.2 The important role of legumes and their productivity constraints

Legumes were domesticated more than 3000 years ago, and are second only to the *Graminiae* in their importance to humans (Graham & Vance, 2003). They are well known for their ability to fix nitrogen with the assistance of symbiotic bacteria, which provides nitrogen to subsequent crops. It has been estimated that the agriculturally important legumes fix 40 to 60 million tonnes of the nitrogen annually, significantly reducing the application of inorganic nitrogen fertilizers and decreasing some of the adverse effects of fertilizers on the environment (Graham & Vance, 2003). Soybean (*Glycine Max*) is an important member of the legume families. It is a major source of oil production, provides humans with proteins of plant origin and is heavily used in the feeding of livestock, which in turn increases the availability of animal protein for human consumption. From 1961 to 2005, soybean was the primary contributor to world oil production. In 2013 soybean was eighth in a ranking of top global commodities in the world, and more than 42 million tons of the world's oil was derived from soybean (FAOSTAT, 2015). Not surprisingly, the soybean has always been in high demand. For example, the data from FAOSTAT show that soybean production has increased from 115 million tons in 1993 to 308 million tons in 2014 (FAOSTAT, 2015). Unfortunately, this increase in production was mainly due to increases in harvested area rather than a slow increase in plant productivity. So, from 1993 to 2014, the annual growth of the yield was only 0.99%, whereas, the annual increase of the harvested area was 3.27% (FAOSTAT, 2015). In comparison, the yield of other staple crops like wheat and rice, during this period enjoyed annual growth rates of 1.24% and 1.12%, respectively, whereas the annual growth of the harvested area for these crops was 0% and 0.49%, respectively. Moreover, in terms of the yield, within the five top soybean producing nations, the productivity varied drastically: Argentina produced 2.74 metric tons per hectare while India only produced about one metric ton per hectare (Masuda & Goldsmith, 2009), indicating improvement in productivity in some developing countries will greatly increase the world soybean production overall.

Soybean productivity constraints in different countries are often due to different combination of reasons. For example, developed countries are more likely to embrace the implementation of modern agricultural technology, such as, precisely controlled irrigation systems and the cultivation of abiotic and biotic stress resistant cultivars, whereas these approaches are often beyond the reach of most farmers in developing countries (Graham & Vance, 2003). This partially explains the still low productivity of crops in the developing world. In general, crops are often under abiotic and biotic stresses and so are prevented from expressing their full genetic potential in nature (J. S. Boyer, 1982). For example, in 1998, the top ten soybean producing countries lost 28.5 million tonnes of soybean, valued at $6.29 billion US, because of various disease infections (Wrather et al., 2001). There are several ways to combat this kind of adversity and so increase crop productivity, through both traditionally deployed artificial selection and the more recently developed genetics based breeding (Tester & Langridge, 2010) to cultivate crops that are resistant to various unfavorable conditions, or by application of fertilizers and pesticides to eliminate and reduce both abiotic and biotic stresses. The latter approaches have already been put to extensive use, the data from FAOSTAT show that the worldwide application of nitrogen fertilizer had significantly increased from 79 million tonnes in 2002 to 92 million tonnes in 2009 (FAOSTAT, 2015) and approximately 1.8 billion people engaged in agriculture were involved in pesticide application. The United States alone utilize more than one billion pounds of pesticides each year and globally, around 5.6 billion pounds of pesticides are used annually (Alavanja, 2009). This implies a significant risk to both biodiversity and human health. Although, fertilizer and pesticide application have been significantly increased worldwide, access to even these more standard measures are nevertheless rather limited, especially for most subsistence farming carried out in the developing countries. The same situation also hold true for the cultivation of genetically modified crops, even though many scientists believe that transgenic plant varieties are probably the most promising tool for augmenting agricultural production and productivity (Herrera-Estrella, 2000). Since the bulk of human population growth is occurring, and will continue to occur in the developing world, it is critical for these nations to have access to the same easily utilized and effective approaches to improvement in crop productivity available in the developed world. One of these approaches is the exploitation of an abundance of various seaweed resources.

2 The brown seaweed: *Ascophyllum nodosum* and its extract, ANE

2.1 The increasing focus on seaweed resources

Recently, attentions has increasingly shifted towards various traditionally used seaweed species and their corresponding extracts, valued for their promotion of both plant growth and yield as well as their ability to relieve stress in varied crops (Khan et al., 2009). Seaweed is a promising resource abundant in nature and includes the macroscopic, multicellular marine algae that commonly inhabit the coastal regions of the ocean. Based on their pigmentation, about 9,000 species of macroalgae have been broadly classified into three groups called brown, red, and green algae (Khan et al., 2009). More focus has been directed towards the exploitation of seaweed. As an example, since 1984, the global production of seaweeds has grown by 119% (Zemke-White & Ohno, 1999). The extensive application of seaweed in agriculture is a technique whose history extends back to ancient times when groups as diverse as the Ancient Greeks, Chinese, and Vikings directly applied seaweed as mulches to the crops (Aitken & Senn, 1965). Various seaweeds and their corresponding products have also been extensively researched, among which, the brown seaweed *Ascophyllum nodosum* has been the most studied species.

2.2 The classification and distribution of *Ascophyllum nodosum*

Ascophyllum nodosum is a slow growing brown algae (Gibb, 1957); it was originally named *Fucus nodosus L.* classified as the Fucaceae family, order Fucales. Members of this order possess 32 chromosomes, and large number of sub-species show genetic variations (Baardseth, 1970). *Ascophyllum nodosum* has been recorded in many parts of the North Temperate Zone, where the occurrence of *Ascophyllum nodosum* largely depends on the frequent alternation of high and low salinity in an area before it can establish itself (Gibb, 1957). In Canada, *Ascophyllum nodosum* mainly dominates the rocky intertidal shores of the Canadian Maritime regions (Ugarte & Sharp, 2012).

2.3 *Ascophyllum nodosum* used as manure

In coastal regions in the past, farmers used to apply *Ascophyllum nodosum* as manure. By 1970, in Ireland, Scotland, France, England and Iceland, it was still being harvested for this purpose and was either directly applied to fields or allowed to first decompose in heaps (Baardseth, 1970). At the time, evidence suggested that this seaweed fertilizer was superior to chemical fertilizers. Application of seaweed fertilizer was shown to enhance uptake of magnesium, nitrogen and calcium in melon leaves, increase yields of grapes and tomato, increase the sugar content of sweet corn and melons, promote initiation of flower buddings in sweet pepper and, prolong the life of the blooms of Christmas plants (Aitken & Senn, 1965).

2.4 *Ascophyllum nodosum* extract (ANE)

Previously, both its bulky and its slow action limited the extensive use of this seaweed in agriculture. Recently, however, extracts of *Ascophyllum nodosum* have been produced, which conveniently transportable and easy to use. In fact, in different regions, seaweed extracts have been produced from various seaweed species; among these, *Ascophyllum nodosum* has been the predominant species harvested from the North Atlantic Ocean for the purpose of extract production (Verkleij, 1992).

2.5 The effects of ANE on plant growth and plant stress resistance

Ascophyllum nodosum extract (ANE) is typically produced through alkaline extraction under high temperature and pressure (Stirk & Staden, 1997). Similar to application of whole *Ascophyllum nodosum*, the application of ANE exhibits multiple effects on various plants, be it by foliar application or direct soil irrigation. For example, application of ANE promotes growth of various plant species (particularly root growth), increases yields, facilitates soil microbial activity, thus increasing soil bioactive content, and enhances plant resistance to abiotic and biotic stresses as discussed below.

2.5.1 ANE as a plant growth promoter

Examples of the ability of ANE to promote plant growth are numerous. For instance, application of ANE to *Brassica napus* increase root dry weight (Jannin et al., 2013) and, increased the root length, root area, total root volume and especially the root number of four different strawberry cultivars, as well as increasing, the fresh root weight and biomass of the two carrot cultivars and, heightening microbial colony counts after an ANE soil drench (Alam, Braun, Norrie & Hodges, 2013; Alam, Braun, Norrie & Hodges, 2014). Application of ANE was also observed to increase the root and leaf elongation in *Arabidopsis thaliana* (Rayorath et al., 2008), and promoted the daily growth of the red algae *Kappaphycus alvarezii* (Loureiro, Reis, Berrogain & Critchley, 2012). Application of ANE also heightened carrot yields by 20% at 0.5g/L in the field, and that of eggplant by 84.38% at 2g/L; it also increased the weight of lettuce and the curd diameter of cauliflower

(Abetz & Young, 1983; Alam et al., 2014; Bozorgi, 2012). These beneficial effects on growth and yield were correlated with an increase in cytokinin-like activity, accumulation of auxin in roots and decreased degradation of chlorophyll contents after ANE treatment (Blunden, Jenkins & Liu, 1997; Khan, Hiltz, Critchley & Prithiviraj, 2011; Rayirath et al., 2009; Rayorath et al., 2008). Irrigation of spinach roots with different concentrations of ANE before harvest, also increased total plant phenolic and flavonoid contents, and enhanced the Fe2+ chelating ability; onions treated with ANE from a cold process extraction exhibited higher phenolic and flavonoid contents, and potato displayed higher flavonoid content (Fan et al., 2011; Lola-Luz, Hennequart & Gaffney, 2014).

2.5.2 ANE improve plant resistance to abiotic and biotic stresses

It has been demonstrated that ANE can either directly or indirectly increase plant resistance to abiotic and biotic stresses. In one study, an alcoholic extraction of *Ascophyllum nodosum* from the Pacific coast displayed a direct antibacterial function (Vacca & Walsh, 1954). Further to this, application of ANE and its component organic fractions increased the freezing tolerance of *Arabidopsis thaliana* under both laboratory and greenhouse conditions (Rayirath et al., 2009) and induced resistance in *Arabidopsis thaliana* to a bacterial pathogen in a jasmonic acid-dependent manner (Subramanian et al., 2011). Moreover, ANE can also inhibit fungal disease in greenhouse cucumber possibly through the activation of disease-related enzymes (chitinase, β-glucanase, peroxidase, polyphenol oxidase, phenylalanine ammonia lyase (PAL) and lipoxygenase (LOP)) in plants (Jayaraman, Norrie & Punja, 2011). The growth promoting and stress-alleviating effects of ANE have been extensively elucidated, but both the bioactive components within ANE and the mechanism behind their action are still poorly understood.

2.6 Proposed beneficial components in *Ascophyllum nodosum*

2.6.1 Nutrients in ANE

Although, ANE has been shown to be multifunctional, how it works to promote plant growth and stress resistance remains unexplained. In the past, the reasons for using seaweeds such as *Ascophyllum nodosum* as crop growth promoters were, firstly, that it was generally believed that most seaweeds contained relatively large amounts of nitrogen, phosphorus and potassium, the major components of any plant fertilizer, and , secondly, it was assumed that the high level of organic matter contained in the seaweeds could maintain both moisture and mineral levels in the upper soil layers where plant roots reside, thereby relieving water deficiencies and making minerals available to roots (Aitken & Senn, 1965). In truth, these factors are undoubtedly significant when *Ascophyllum nodosum* is used in large amounts. However, multiple effects can be achieved even when application of ANE are carried out at very low concentrations. Therefore, the beneficial effects of nutrients and moisture-containing organic matter need to be reassessed when considering the positive effects of ANE.

2.6.2 Phytohormones in ANE

Since plant hormones are functional at very low concentrations, the growth-promoting effects of ANE were initially thought to possibly be due to the presence of a plant hormone-like substance in *Ascophyllum nodosum* and its extract ANE (Zodape, 2001). This assumption was ultimately justified by the demonstration of cytokinin- and auxin-like effects on various explants after differing commercial ANE treatments. Specifically, when carrots, soybean callus, mung bean rootings and other explants were used as bioassays, the presence of the cytokinin- and auxin-like activities were revealed in different commercially used ANEs (Brain, Chalopin, Turner, Blunden & Wildgoose, 1973; Stirk & Staden, 1997). Further to this, several plant hormones such as IAA, cytokinins, and ABA were directly detected in *Ascophyllum nodosum* and its commercial extracts. For example, ABA, IAA and cytokinins (Kingman & Moore, 1982) were found in crude aqueous *Ascophyllum nodosum* solutions. Moreover, after analysis of various ANE samples, Wally et al., (2013) reported, the presence of cytokinins in amounts of approximately 10ng/g dry weight of sample, ABA in amounts of approximately 5ng/g and IAA in amounts of 20 to 25ng/g. Similar studies also detected the same phytohormones also at low concentrations (G. L. Boyer & Dougherty, 1988; Jannin et al., 2013). Tarakhovskaya, Maslov, & Shishova, (2007) suggested that the concentrations and the biological functions of these plant hormones were similar to that of the higher plants after reviewing the ubiquitous presence of such hormones in algae. Therefore, the benefits of phytohormones in ANE also need to be reassessed.

2.6.3 ANE mechanism reassessment

The recommended application rate for ANE in the field is only 3 ml/liter and 1-2 liter/acre, which further stress the requirement for more study into the effects of nutrients and plant hormones in ANE on plant growth. Also, there has been little explanation offered regarding the effects of ANE application on plant stress resistance and so there is a need for further study on the action of ANE in regards to both plant growth promotion and plant resistance to both biotic and abiotic stresses.

2.7 The constituents of *Ascophyllum nodosum*

As the effects of extracts taken from *Ascophyllum nodosum* remain consistent regardless of when, where and how the seaweed is harvested, it is assumed that the bioactive components of ANE are likely derived from the primary constituents of *Ascophyllum nodosum* itself.

2.7.1 The composition of *Ascophyllum nodosum*

Typically, the storage products found in brown algae are laminaran (β-1, 3-glucopyranoside, predominantly) with the outer cell wall comprised of an amorphous embedding matrix made up of an alginate and fucoidan matrix. In the inner cell wall, the fibrillary skeleton is structured from an alginate and fucoidan matrix interconnected with cellulose fibers (Davis, Volesky & Mucci, 2003). Laminaran generally represents up to 35% of the brown algal dry weight; alginates take up 17 to 45% of total dry weight and the remaining 5 to 20% of the dry weight is comprised of fucoidan (Vera, Castro, Gonzalez & Moenne, 2011). Several studies have shown that the main organic components of *Ascophyllum nodosum* are alginate, laminaran and fucoidan, although their proportional contents were shown to be relatively dynamic (Baardseth, 1970; Haug & Larsen, 1958; Rioux, Turgeon & Beaulieu, 2007; Rioux, Turgeon & Beaulieu, 2009).

2.7.2 The obligate fungus, *Mycophycias ascophylli*

Most intriguingly, *Ascophyllum nodosum* is not just a stand along alga. *Ascophyllum nodosum* is best known for its obligate relationship with a fungus, *Mycophycias ascophylli*, which lives within the intercellular spaces, inside the algal thallus. In fact, in nature, *Ascophyllum nodosum* has always being found to be infected with *Mycophycias ascophylli* (Selosse & Tacon, 1998). Extensive studies have shown that fungal hyphae are present and abundant in all parts of the thallus of *Ascophyllum nodosum*, and in all other tissues, though there are variations in fungal morphology dependent on which region of the seaweed the fungus is growing. Furthermore, by observing the behaviour between the *Mycophycias* *ascophylli* and *Ascophyllum nodosum* it has been confirmed that *Mycophycias ascophylli* is a symbiotic partner of *Ascophyllum nodosum* (Deckert & Garbary, 2005). In fact, the reproductive cycles of the two symbionts are connected, with colonization of the seaweed by *Mycophycias ascophylli* occurring soon after *Ascophyllum nodosum* zygote formation (Garbary, 2009; Xu, Deckert & Garbary, 2008). Previous research has also shown that the thallus of *Ascophyllum nodosum* provides laminaran for *Mycophycias ascophylli* as a carbon source (Fries, 1979).

Although there is still little information available concerning the significant of this fungus in *Ascophyllum nodosum,* a recent study has highlighted the importance of this symbiont for the promotion of plant growth (Sarah & Garbary, 2015). Very little research has been undertaken to uncover the composition of this particular symbiont, but a typical fungal cell wall mainly consists of chitin and β-glucan. Within this cell wall, the chitin chain associates by way of microfibrils covalently bound to the primary constituent, β-glucan. These then associate with various glycoproteins, forming a network that confers stiffness to the cell wall. As plants are subject to attack from a wide array of fungal pathogens, plants' crucial structural components have, over time, evolved ways to detect such attacks on their tissues. Intriguingly, some pathogenic fungi have developed the capacity to elude plant recognition by converting chitin to chitosan via deacetylation during host invasion (Sanchez-Vallet, Mesters & Thomma, 2014).

2.7.3 Possible oligosaccharides in ANE

Ascophyllum nodosum is a mosaic of the seaweed and its fungal symbiont, *Mycophycias ascophylli.* Thus, the primary constituents of *Ascophyllum nodosum* are a blend of various polysaccharides from both seaweed and fungus, including laminaran, alginate, fucoidan, chitin and β-glucan. Since the extract production from *Ascophyllum nodosum* involves the use of alkaline solutions at high temperatures and pressures, these polymers will be present in ANE mainly in the form of oligosaccharides, with the chitin being deacetylated to form chitiosan (Kumar, 2000). So the primary components of ANE are expected to be oligo forms of laminaran, aliginate, fucoidan, chitin, chitosan and β-glucan. Thus, for a more complete understanding of ANE's effects on plants, the effects of these oligosaccharides on plants also need to be assessed and compared.

2.7.4 Brief summary of oligosaccharides in ANE

Many studies have shown that oligosaccharides, including those found in ANE, can have various beneficial effects on plants. In one review (Vera et al., 2011), it was concluded that seaweed-derived polysaccharides such as alginates, fucoidan, laminarin and (their associated oligosaccharides) can induce an initial oxidative burst resulting in subsequent activation of SA, JA and/or ethylene signaling pathways in terrestrial plants leading to increases in expression of pathogen-related (PR) proteins showing antifungal and antibacterial properties. These pathways also increase production of defense enzymes that participate in the synthesis of secondary metabolites such as terpenes, terpenoids and alkaloids that carry out antimicrobial activities.

In conclusion, in order to reassess the mechanism underlying the effects of ANE on plants, the effects of these corresponding oligosaccharides on plants need to be compared and contrasted with the effects of ANE. In present study, the similarities and differences of oligosaccharides and ANE treatments on soybean seedlings were compared at the transcriptomic level.

3 Material and methods for planting and treatments

3.1 Preparation of ANE and oligosaccharides

Acadian® 100% liquid seaweed concentrate derived from *Ascophyllum nodosum*, containing 0.1% N, 0% P, and 5.0% K respectively was diluted to 2ml / L for soybean seeds soaking and 3ml / L to use for spraying soybean seedlings later on.

Oligo-alginate, oligo-chitin, and oligo-chitosan were kindly provided by Dr. Yuguang Du (Institute of Process Engineering, Chinese Academy of Sciences), with oligo-chitin and chitosan both possessing an average polymerization of 4 (see Figures 1 and 2, showing the MS spectrum for structural information). 1000 ppm of stock solutions were made for all oligosaccharides by dissolving them directly into distilled water.

3.2 Soybean seeds soaking and plants growing

Soybean seeds, cultivar Hidasta, were obtained from the Halifax Seed Company on April 28, 2015 and were soaked at room temperature for 36 hours before planting. For seeds soaking, 500 similar sized seeds were selected and evenly distributed in twenty-five petri dishes. Each treatment consisted of 20 ml of solution used for soaking the soybean seeds. The five treatments were as followed: 2ml/L of ANE, 26.7 ppm of oligo-alginate, 20 ppm of oligo-chitin and 33.3 ppm of oligo-chitosan and distilled water used as control.

Plastic pots each with a diameter of 23.25cm, were filled with Pro-mix LP 15 and used for the soybean planting. 20 pots, each with 4 germinated seeds, were assigned to each treatment. After completion of the planting, each pot was watered and, all 100 pots were then placed outside of the greenhouse in Saint Mary's University.

10X diluted Hoagland solution were used as the nutritional supply for the plants during the experimental period, which ran from May 28th, 2015 to July 17th, 2015. The solution was made according to Hoagland & Arnon, (1950) and was used to drench the soil in each pot every day except the rainy days. Half strength Hoagland solution was used for all five treatments three days before leaf collection as yellowing of the leaves was observed using the 10X diluted solution.

3.3 ANE and oligosaccharide spraying treatments

Distilled water, ANE, oligo-alginate, oligo-chitin and oligo-chitosan were used to spray 45 day-old soybean seedlings grown between May 28th, 2015 and July 12th, 2015. The five spraying treatments were as follows: 3 ml\ L of ANE, 40 ppm oligo-alginate, 30 ppm oligo-chitin, 50 ppm oligo-chitosan and distilled water used as a control. 100 ml of each solution were used for each pot until all the leaves were wet.

3.4 Soybean leaf collection

12 hours after spraying treatments, in each treatment, first and second compound leaves (those closest to the ground) from one plant were collected, and put into a 1.5 ml Eppendorf tube. All tubes containing approximately 100 mg of leaf material each, were immediately frozen in the liquid nitrogen and transferred to the -80℃ refrigerator for later RNA extraction.

4 Methods for mRNA sequencing

4.1 RNA extraction, qualification, and transportation

RNA was extracted using the QIAGEN RNeasy® Plant Mini Kit (20), Cat. No. 74903 following the manufacturer's instructions. After RNA extraction, Nanodrop was used to qualify the RNA concentration determined by the OD 230, OD 260, OD260/OD280 and OD260/OD230 values. Qualified RNA samples were then transferred to a Biomatrica RNA stable tube kit (#93221-001) and vacuum dried before being shipped to Novogene (China) for mRNA sequencing.

4.2 mRNA sequencing workflow

From RNA extraction to final sequencing, each step, influences the quality of the data, which in turn directly impacts both the results and subsequent data analysis. Therefore to guarantee the reliability of the data, quality control (QC) was performed at each step of the procedure (see Figure 3 for workflow).

4.2.1 Total RNA Sample QC

RNA samples were required to pass through four QC procedures before being used for cDNA library construction: Firstly, samples were run through agarose gel electrophoresis to test for RNA degradation and potential contamination. Secondly, samples were put through Nanodrop tests to check RNA purity using OD260/OD280 values. Next, Qubit was utilized to quantify RNA concentration. Finally, samples were run through the Agilent 2100 to verify RNA integrity.

4.2.2 Library construction and sequencing

After QC procedures were completed, mRNA was enriched using oligo (dT) beads. The mRNA was then fragmented randomly in fragmentation buffer, followed by cDNA synthesis using random hexamers and reverse transcriptase. After first-strand synthesis, a custom second-strand synthesis buffer (Illumina) was added with dNTPs, RNase H, and Escherichia coli polymerase I to generate the second strand by nick-translation. The final cDNA library was ready after a round of purification, terminal repair, A-tailing, ligation of sequencing adaptors, size selection, and PCR enrichment. The workflow for cDNA library construction is shown in Figure. 4. Library concentration was first quantified using a Qubit 2.0 fluorometer (Life Technologies) and then diluted to 1 ng/µl before checking insert size on an Agilent 2100 and quantifying to greater accuracy via quantitative PCR (Q-PCR) (library activity >2 nM). Libraries were fed into HiSeq machines for sequencing.

4.3 Raw data description

The original raw data from the Illumina HiSeqTM PE125/PE150 sequencer were transformed to sequence reads by base calling. Raw data were recorded in an FASTQ file, which contains sequence information (reads) and corresponding sequencing quality information. Every read in FASTQ format was stored in four lines as follows:

```
@HWI-ST1276:71:C1162ACXX:1:1101:1208:2458 1: N: 0: CGATGT

NAAGAACACGTTCGGTCACCTCAGCACACTTGTGAATGTCATGGGATCCAT

+ #

55???BBBBB?BA@DEEFFCFFHHFFCFFHHHHHHHFAE0ECFFD/AEHH
```

Line 1 begins with a '@' character and is followed by the Illumina Sequence Identifiers and an optional description, line 2 is the raw sequence read, line 3 begins with a '+' character and is optionally followed by the same sequence identifier and description. Line 4 encodes the quality values for the sequence in line 2

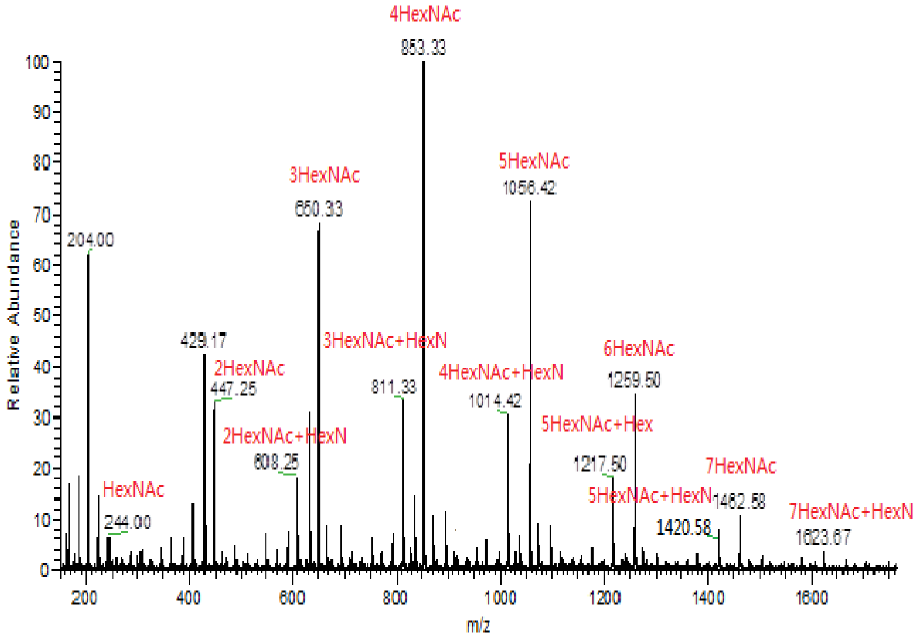


Figure 1: Structural information for oligo-chitin.

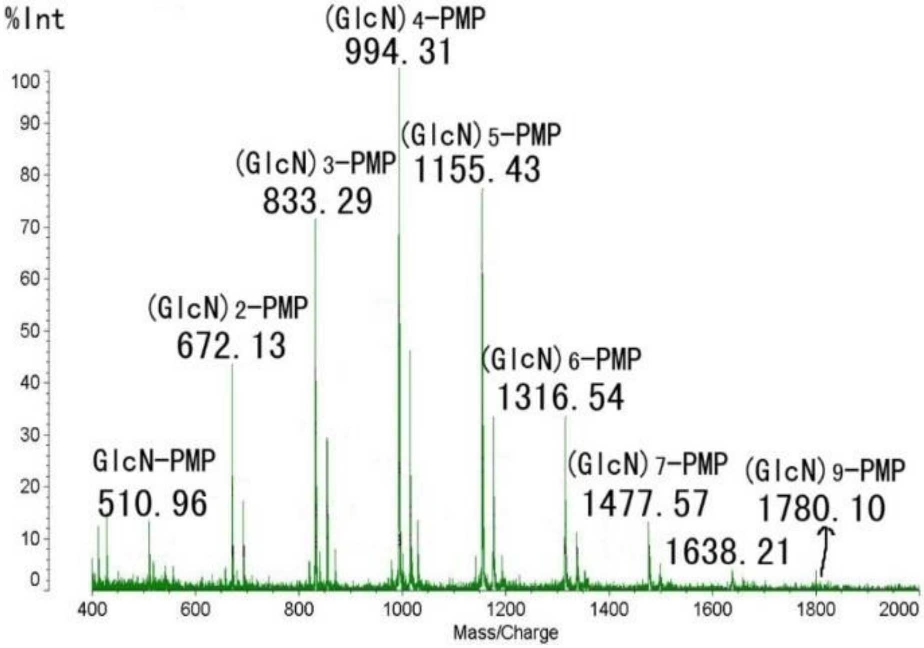


Figure 2: Structural information for oligo-chitosan.

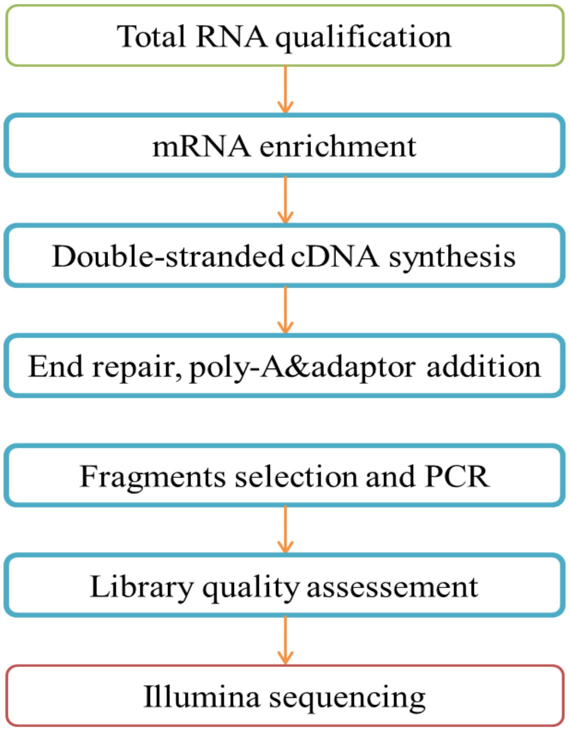


Figure 3: mRNA sequencing workflow.

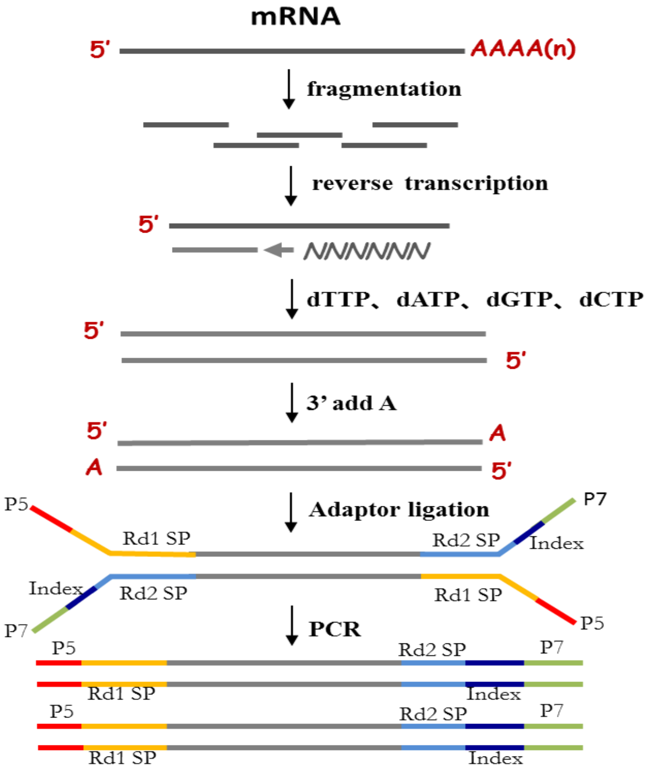


Figure 4: Library construction workflow.

5 Methods for data processing and analysis

5.1 Raw data filtering

Sequencing error rate and base quality depend on the sequencing machine, reagent availability, and the quality of the samples themselves. Error rate increases as the sequencing reads are extended and sequencing reagents become more and more scarce and so the first six bases have a relatively high error rate due to the random hexamers used in priming cDNA synthesis. Raw reads were filtered to remove reads containing adapters or reads of low quality so that downstream analyses were based on clean reads. The filtering process consists of discarding reads with adaptor contamination or reads where uncertain nucleotides constitute more than 10 percent of a read (N > 10%); reads were also discarded when low-quality nucleotides (base quality less than 20) constituted more than 50 percent of the read.

5.2 Clean data mapping to soybean genome

Glycine Max genome fasta file and its corresponding annotation file were downloaded from ftp://ftp.ensemblgenomes.org/pub/release31/plants/fasta/glycine_max/dna/Glycinemax.V1.0.31.dna.toplevel.fa.gz and ftp://ftp.ensemblgenomes.org/pub/release31/plants/gtf/glycine_max/ Glycine_max.V1.0.31.gtf.gz respectively. The Hisat2 alignment program (Kim, Langmead, & Salzberg, 2015) was downloaded from ftp://ftp.ccb.jhu.edu/pub/infphilo/hisat2/downloads/ hisat2-2.0.3-beta-Linux_x86_64.zip and installed on Ubuntu 14.04 to map the clean reads to the soybean genome. The parameters were set at default.

5.3 Distribution of the mapped reads on the soybean genome

Qualimap2 (Okonechnikov, Conesa, & García-Alcalde, 2015) is a platform independent application used to facilitate the visualization of next generation sequence data. In this study, Qualimap2 was downloaded from http://qualimap.bioinfo.cipf.es/ and was then used to visualize the distributions of the mapped reads on the soybean genome. The parameters were set as default.

5.4 Gene expression quantification

Gene expression level is measured by transcript abundance. The greater the abundance, the higher the gene expression level. In this RNA-sequence analysis, the gene expression level was estimated by counting the reads that mapped to gene or exons. The coordinates of a gene or exons on the soybean genome were annotated in the soybean genome annotation file downloaded from ftp://ftp.ensemblgenomes.org/pub/release31/plants/gtf/glycine_max/ Glycine_max.V1.0.31.gtf.gz. Read count from mRNA sequencing is not only proportional to the actual gene expression level but also dependent on gene length and sequencing depth. To make the expression level of same gene to be comparable in different samples and different genes in same sample to be comparable, the RPKM was used to measure the gene expression level. In mRNA sequencing, RPKM stands for reads per kilo base of transcript per million of mapped reads, which is the most common used method of estimating gene expression levels. This technique, takes into account the effects of both gene length and sequencing depth as shown below:

RPKM = Number reads / (Gene length/1000 * Total number reads/1,000,000)

Where Number Reads equals the number of reads mapped to a gene sequence, Gene Length is the length of the gene sequence and Total Number Reads represents the total number of mapped reads in a sample.

The quantification of gene expression levels in this study was determined by the GFOLD (generalized fold change) program (Feng et al., 2012). GFOLD program consists of two sub-programs, one is gfold –count, which is for counting the number of mapped reads on the soybean genome; second one is gfold –diff, which is responsible for calculating the RPKM value for each gene and generating a gfold (generalised fold change) value to define the differentially expressed genes when dealing with two different conditions. Specifically, for counting the number of reads that mapped to a specific gene in soybean genome, the gfold –count needs to take two inputs, one is the mapping results from Hisat2 and the other one is the soybean genome annotation file.

5.5 Determination of differentially expressed genes

When comparing the expression level of genes between two different conditions, the gfold -diff program was used. Specifically, read count result for each condition were estimated by gfold –count program. By taking the corresponding read count results of two conditions and based on the gene length, gfold –diff program then calculated RPKM values for the genes in each condition. Based on these RPKM values in two conditions, gfold –diff program then considered the posterior probability of the fold changes of genes and generate gfold (generalised fold change) values for each gene. The main benefit of gfold values is in providing a biologically meaningful ranking of the genes. The gfold value is zero if the gene does not show differential expression between two conditions. For one specific gene, if the log2 (RPKM in sample 2/ RPKM in sample1) is treated as a random variable, a positive gfold value x means that the probability of the log2 (RPKM in sample 2/ RPKM in sample1) being larger than x is 1 - SC (significant cut off). A negative gfold value x means that the probability of the log2 (RPKM in sample 2/ RPKM in sample1) being smaller than x is 1 -SC. In this study, the -SC parameter was set as 0.01, which means the probability of the log2 (RPKM in sample 2/ RPKM in sample1) being smaller/greater than gfold value is 99%. The absolute value of gfold being equal or greater than 1 was used in this study to define the differentially expressed genes between two samples. So, all the differentially expressed genes defined in this study at least have a two-fold change of expression.

5.6 Gene expression pattern analysis

The effects of different treatments on the physiological changes within soybean seedlings are essentially wrought through the regulation on the expression of responsible genes. The most efficient method for assessing the similarities and differences of the ANE and oligosaccharide treatments regarding gene influences is to look at the altered gene expression patterns under different treatments. Specifically, to assess the overall relationships of different treatments, a gene was chosen whose expression changed under at least one of the treatments compared with water. Expression levels of these genes (RPKM) were extracted from the results of gfold –diff. The collective set of theses gene IDs and its corresponding expression levels (RPKM) were used to construct a gene expression matrix. The RPKM values in each row of the matrix were normalized for the Z score (z = (x - mean)/std), so values in each row will get the mean of the row subtracted, then divided by the standard deviation of the row. For visualization of this gene expression matrix, the python visualization library seaborn 0.70 was used to make clustering heat maps. The colors in the heat map indicated the relative expression level of the genes. By doing so, it is easier to compare the effects of the different treatments on one specific gene. The similarity of effects on the expression of a collection of genes by different treatments could be inferred from the cluster result. The method for calculating the cluster was set as default.

To test which of the oligosaccharides treatment is more similar to that of ANE, it is natural to focus on the genes that regulated by ANE treatment and then examine the expression of same list of genes under treatments of oligosaccharides. Therefore, treatment of ANE regulated, up and down regulated gene IDs were extracted. The expression levels of these three gene sets under all treatments were retrieved from the corresponding gfold –diff results. Gene expression matrix were then constructed for these three set of genes. Heat map were then used to visualise the expression patterns of these genes under different treatments. The results from the cluster indicate the relationships of the gene expression patterns among different treatments. This baseline made it easier to establish relationships between both the three gene sets themselves and between oligosaccharide treatments and ANE treatment in regard to gene influences.

5.7 Commonly regulated genes between treatments of ANE and oligosaccharides

The number of the commonly regulated genes between treatments of ANE and oligosaccharides are another indicators for their similarity. To further explore these particular genes, corresponding Venn diagrams were drawn using jvenn, an online software program (Bardou, Mariette, Escudié, Djemiel, & Klopp, 2014). Specifically, in terms of commonly regulated (includes up- and down-regulated) genes, gene IDs of each treatment-regulated genes were extracted and used as input for the jvenn platform. Similarly, gene IDs of up-regulated and down-regulated genes under different treatments were extracted and analyzed independently. The results of Venn diagram is represented with overlapped circles, however, the size of the overlapped circle is not proportional to the size of commonly regulated genes. The number of commonly and uniquely regulated genes in all treatments were labelled with numbers.

5.8 Function annotations of genes influenced by ANE treatment

To further explore the functions of these commonly regulated genes between ANE and oligosaccharides treatments, the IDs of these genes were extracted and their functions were annotated. Specifically, up- and down-regulated genes in treatment of ANE consists of ANE uniquely regulated genes and genes that regulated in other treatments as well. Based on these gene IDs, the protein sequences of these genes were retrieved from Ensembl Plants Biomart (http://plants.ensembl.org/biomart/martview/ddac2d59a96f65c9aa7d51e4cace 43 fa). These protein sequences were then blast against three complementary protein database: Swiss-Prot, TrEMBL and NCBI nr, respectively by using the NCBI blast program (Altschul, Gish, Miller, Myers, & Lipman, 1990) installed locally. The blast searching results of these three database were then combined to comprehensively annotate the function of these genes. Treatment of ANE up- and down-regulated genes were then further classified into various biologically meaningful categories and regulations (gfold values in this case) of these genes by treatments of oligosaccharides were also included. The detailed annotation results were attached in the Appendix 1 and 2 for ANE up- and down-regulated genes, respectively. For tabular results in the Appendix, genes labelled with soybean gene IDs were listed in the second column. The first column were the names of treatments that regulated these genes. Since the Swiss-Prot protein database is most reliable among the searched database, the third column in the tables were the corresponding Swiss-Prot gene IDs. Column 4, 5, 6 and 7 were the gfold values for the genes under different treatments

(ANE, oligo-alginate, oligo-chitin and oligo-chitosan, respectively). Column 8 was the basic function for one specific gene. Column 9 were the biological processes that one specific gene involved in, which were extracted from the UniProt protein database, to facilitate the categorization. The information from the tabular results were summarized in the form of pie chart, to facilitate the understanding of the common effects that treatments of ANE and oligosaccharides on the soybean seedlings.

5.9 Comparison of treatments regulated biological processes

Differentially expressed genes after treatments of ANE and oligosaccharides reflects the general differing influences of each treatments on the soybean seedlings. The mRNA sequencing results often produce thousands of differentially expressed genes under different experimental conditions. It is usually interesting to see which biological pathways are involved in the up or down regulated genes. To understand that, the pathways that differentially expressed genes involved in were analyzed. Specifically, based on the gene symbols, the sequences of these differentially expressed genes were retrieved from the Ensembl Plants Biomart (http://plants.ensembl.org/biomart/martview/ddac2d59a96f65c9aa7d51e4cace43fa). Kobas 2.0 (Xie et al., 2011) program was then used, which is an update of KOBAS (KEGG Orthology-Based Annotation System), which annotates an input set of genes with putative pathways and disease relationships based on mapping to genes with known annotations. It also allows for both ID mapping and cross-species sequence similarity mapping, which fully take advantages of the expanded available database. After annotation, it then performs statistical tests to identify significantly enriched pathways. In this experiment, Kobas2.0 software and related datasets were downloaded from http://kobas.cbi.pku.edu.cn /download.do and installed on Ubuntu 14.04

to identify the enriched pathways that different treatments regulated. The results of that could facilitate the interpretation of the effects that different treatments on the soybean seedlings at a larger biological level. Specifically, for the annotation part, the sequences of all detected-, up- and down-regulated genes under different treatments were separately searched against the known pathway database by using the kobas -annotate program. To identify the enriched pathways in different treatments, kobas -identify program was then used by taking the results from annotation part as inputs. The parameters were set as default.

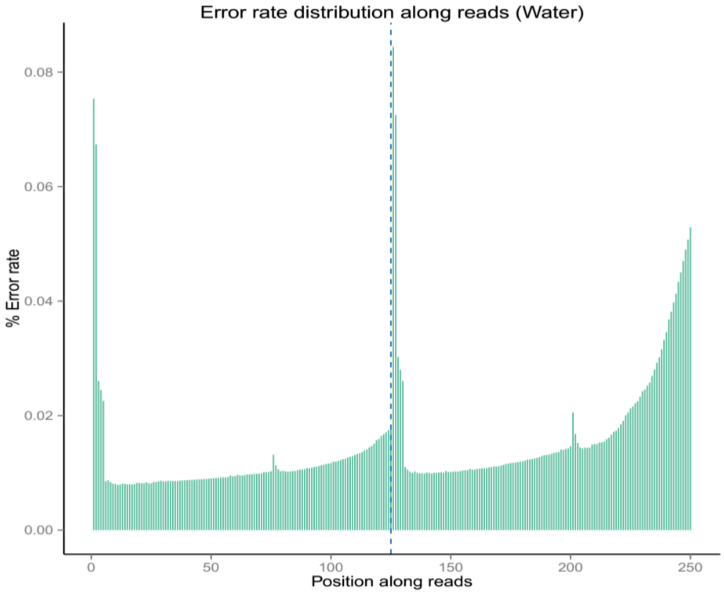
To further comprehend the effects of treatments on the soybean seedlings general metabolic processes, MAPMAN software were employed. Specifically, gene sequences of regulated genes by different treatments were first retrieved from the Ensembl Plants Biomart, then run through the Mercator pipeline (Lohse M et al., 2014) to obtain mapping results. The mapping results came from the searching of several default databases (TAIR Release 10, SwissProt/UniProt Plant Proteins, TIGR5 rice proteins, Clusters of orthologous eukaryotic genes database, Use conserved domain database) imbedded in the Mercator platform with the retrieved gene sequences. Upon the completion of searching, the characterized genes were then mapped to biological data points that involved in metabolic processes. The parameters in the searching and mapping steps were set as default. MAPMAN software were then used to integrate mapping results and the corresponding gene expression information by visualising them in various biological processes.

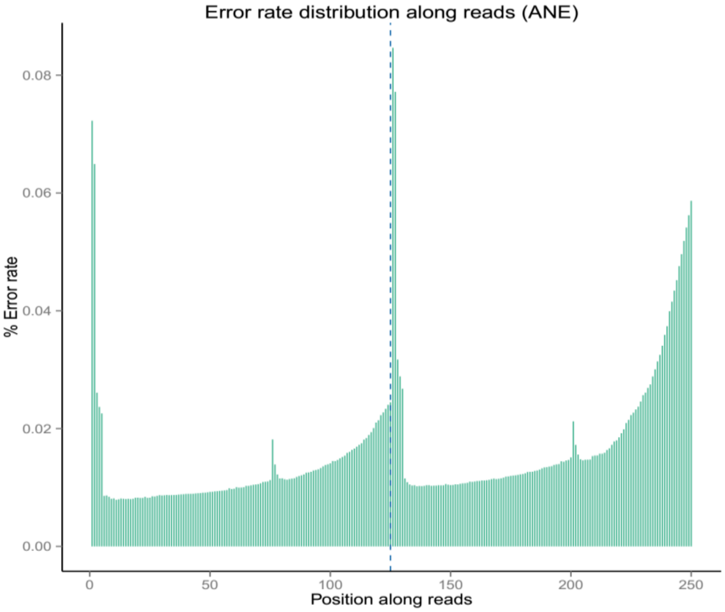
6 Results

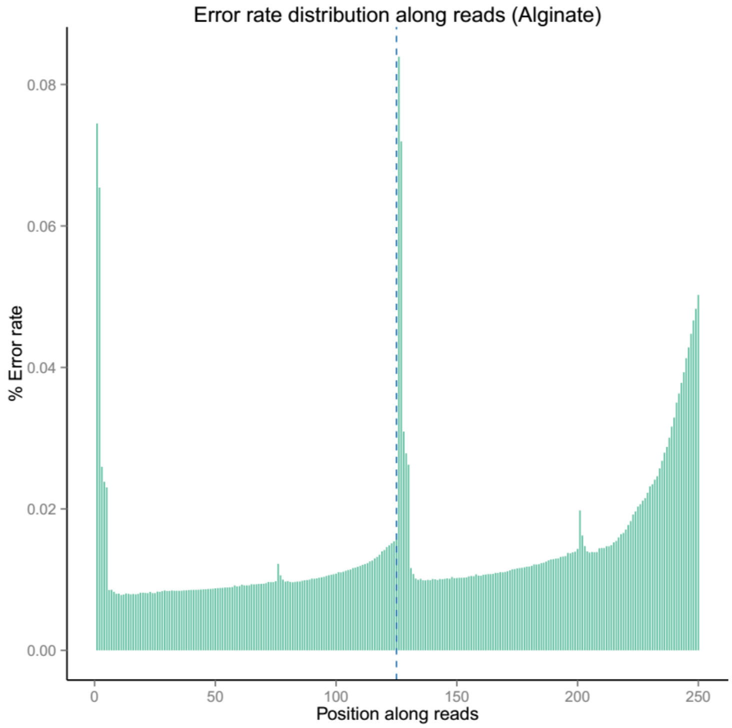
6.1 Sequencing quality

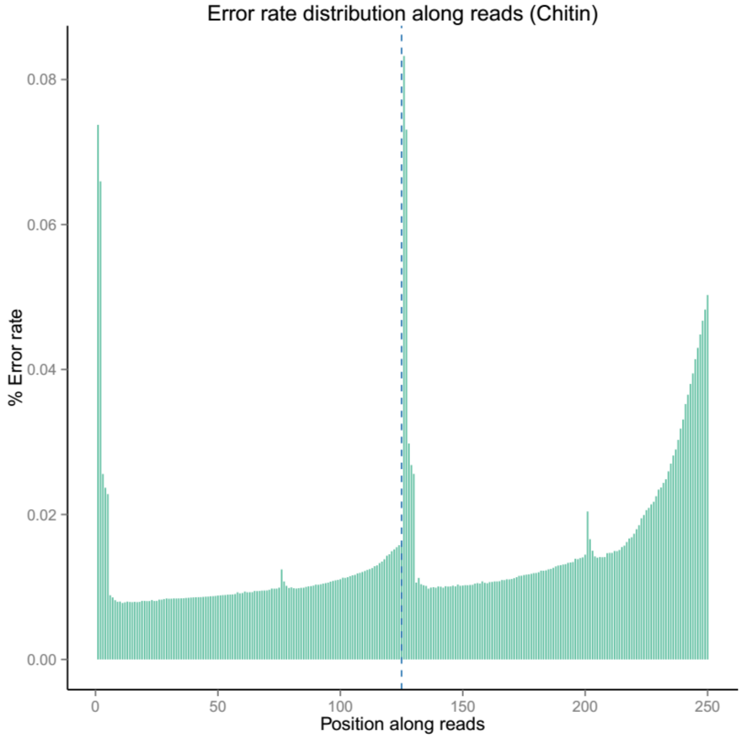
6.1.1 Sequencing error rate

As shown in Figure 5, sequencing error rates for all different sequenced samples averaged below 0.01%.






Error rate distribution along reads (Alginate)


Error rate distribution along reads (Chitin)

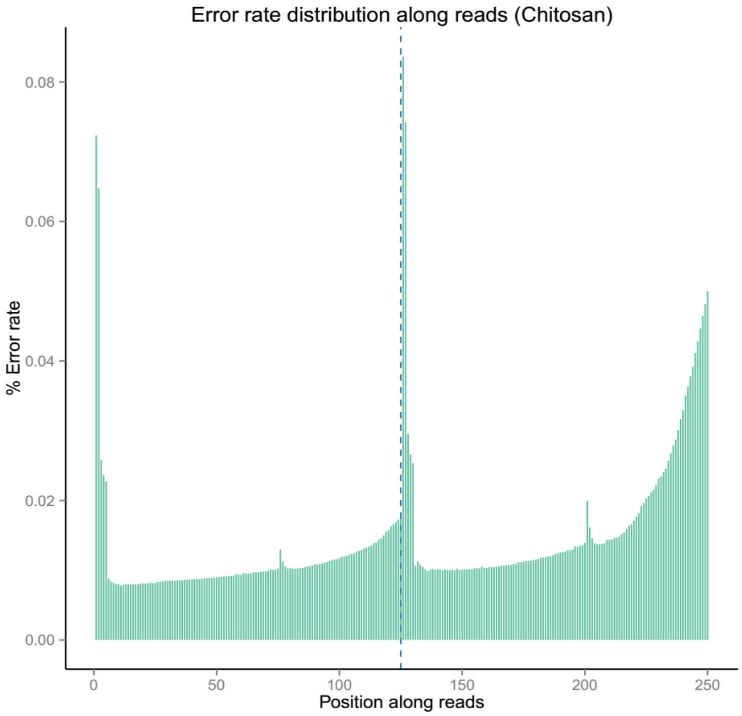


Figure 5: Distributions of sequencing error rate of all samples (water, ANE, oligo-alginate, oligo-chitin, and oligo-chitosan, respectively).

6.1.2 Data classification

The raw data composition in this experiment is shown in Table1. The clean reads of all sequenced samples made up more than 96% of total sequenced reads. After trimming the adaptor and filtering the low-quality reads, base quality, and GC content were calculated and summarized (see Table 2). As illustrated in Table 3, Q30 showed a base calling error rate below 0.1%, and all sequenced samples show values greater than 91% of the Q30 value.

Table1: Data composition

Samples	Clean reads (%)	Containing N (%)	Low quality (%)	Adapter related (%)
Water	96.33	0.01	0.98	2.68
ANE	96.93	0.01	0.82	2.25
Oligo-alginate	97.51	0.01	0.77	1.71
Oligo-chitin	97.12	0.01	0.82	2.05
Oligo-chitosan	96.11	0.01	0.88	3.00

Table2: Data production

Sample	Raw reads	Clean reads	clean bases	Error rate (%)	Q20 (%)	Q30 (%)	GC content (%)
Water	53286956	51332588	6.42G	0.02	96.81	92.49	41.95
ANE	66992712	64934636	8.12G	0.02	96.5	91.71	41.69

Oligo-alginate	56519982	55113642	6.89G	0.01	97.18	93	42.81
Oligo-chitin	63866520	62029010	7.75G	0.01	97.05	92.88	42.42
Oligo-chitosan	79288946	76204800	9.53G	0.01	96.91	92.73	40.49

Clean Bases: clean reads number multiple read length, saved in G unit

Error Rate: average sequencing error rate, which is calculated by Qphred= -10log10 (e) as illustrated in Table 3.

Table3: Base Quality and Phred score relationship with the Illumina CASAVA v1.8 software:

Base calling error rate	Base calling correct rate	Q-score
1/100	90%	Q20
1/1000	99.90%	Q30
1/10000	99.99%	Q40

Q20: percentage of bases whose correct base recognition rates are greater than 99% in total.

Q30: percentages of bases whose correct base recognition rates are greater than 99.9% in total.

GC content: percentages of G and C in total bases

6.2 Mapping results

6.2.1 Statistics of mapping results

The mapping statistics for different samples are summarized in Figure 6. The statistics clearly show that over 90% of the clean reads derived from samples from all five treatments aligned to the soybean genome

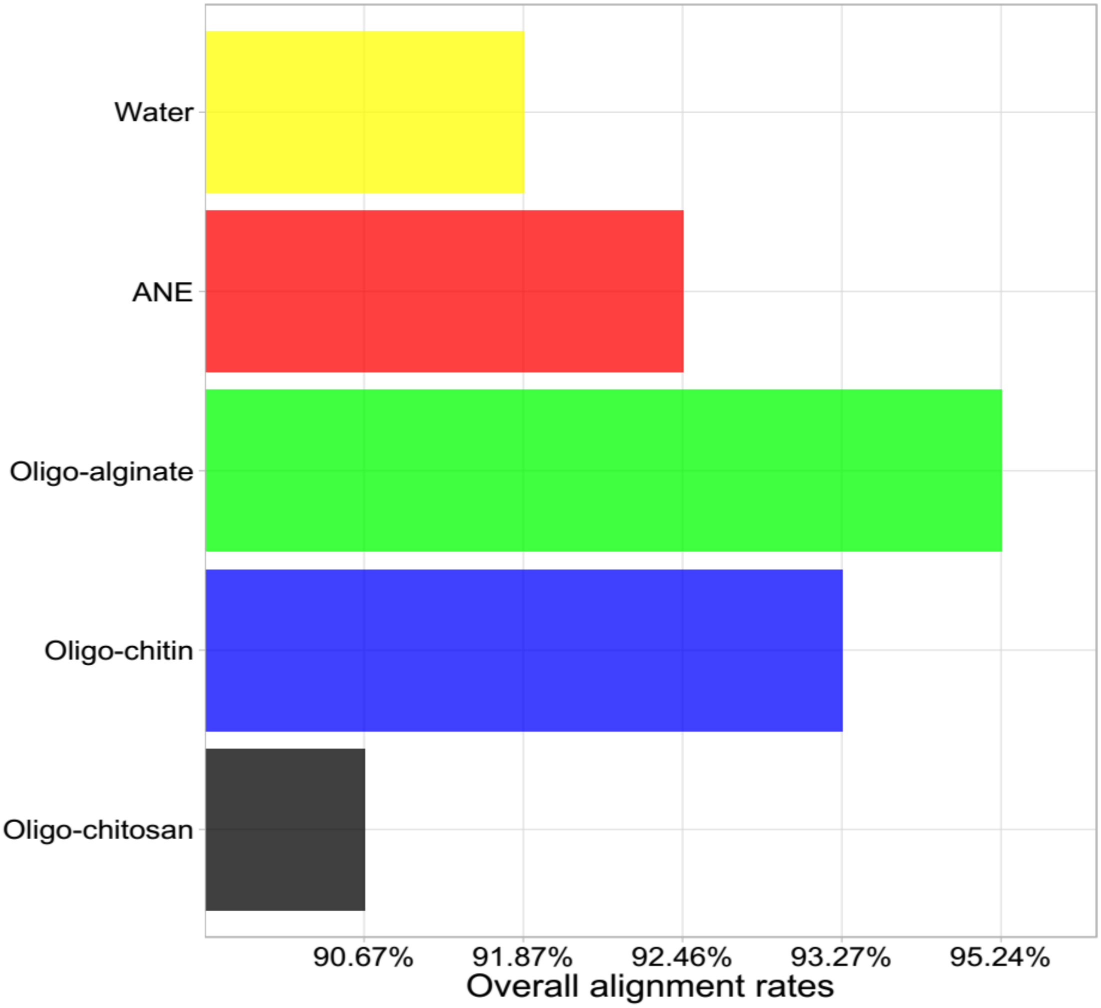


Figure 6: Proportion of mapped clean reads on soybean genome.

6.2.2 Distribution of the mapped clean reads on the soybean genome

The distributions of the mapped reads on the regions of the soybean genome are summarized in the following graphs (see Figure 7 through 11 for water, ANE, oligo-alginate, oligo-chitin and oligo-chitsoan treatments, respectively). As seen below, the mapped clean data were enriched in the exon region (pink in the figures) for samples of all treatments, with a small fraction of clean reads being mapped to intergenic and intronic regions (denoted by blue and green, respectively).

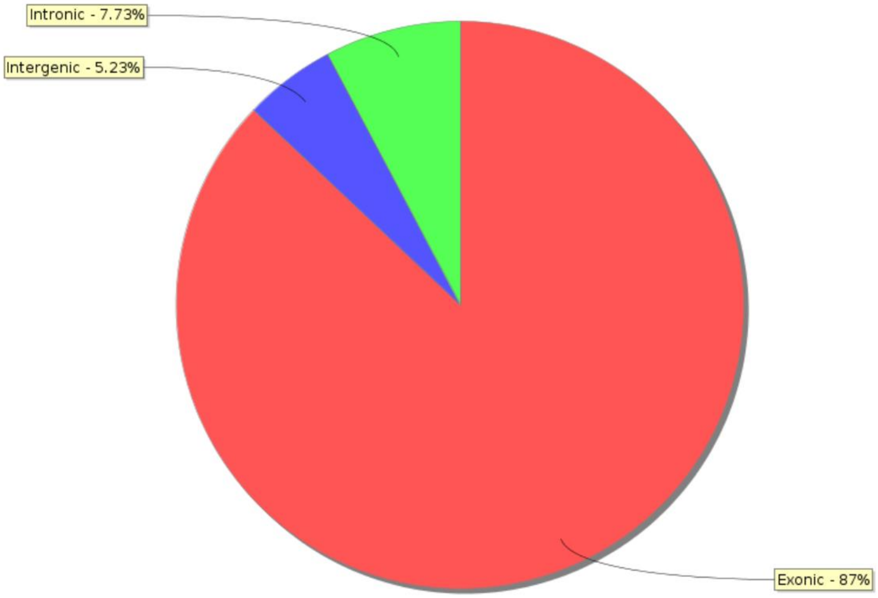


Figure 7: Proportion of mapped reads on different regions of the soybean genome (water treatment).

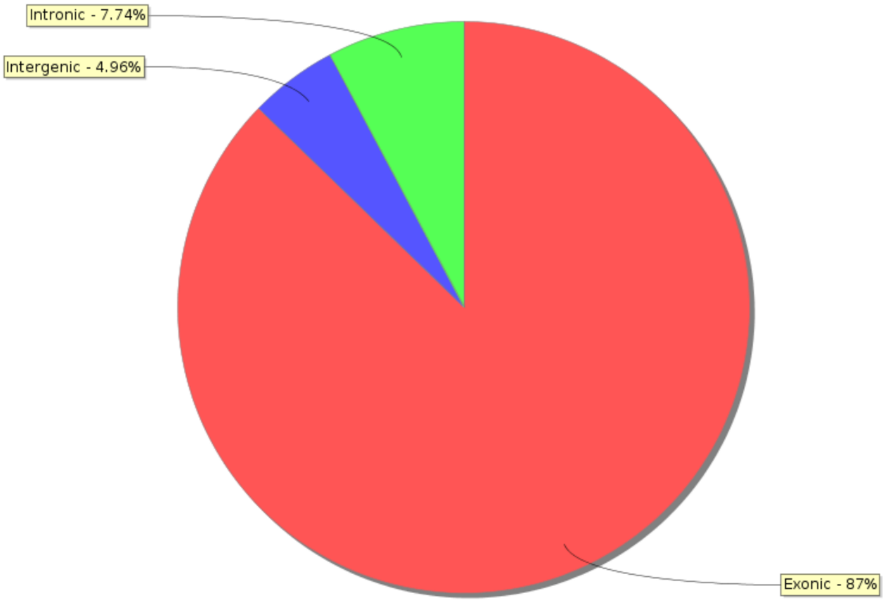


Figure 8: Proportion of mapped reads on different regions of the soybean genome (ANE treatment).

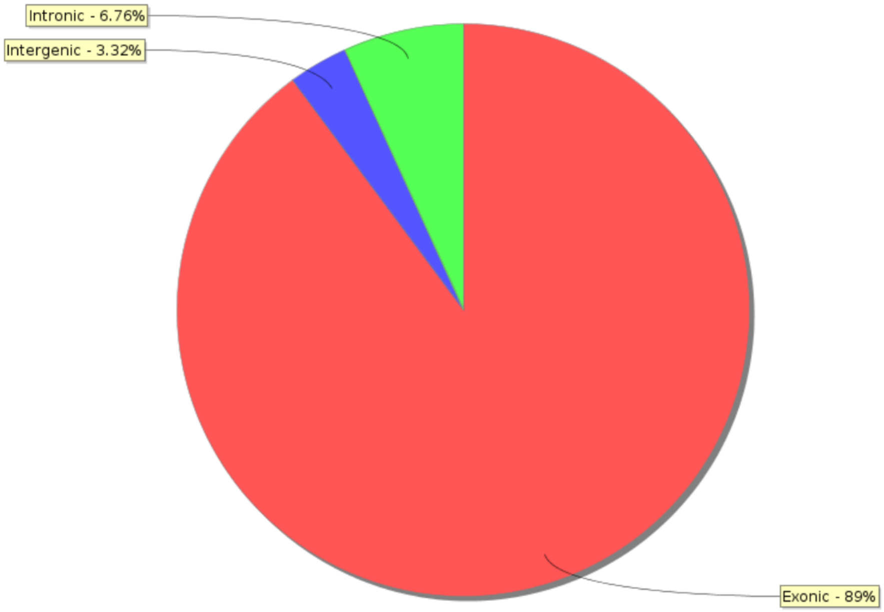


Figure 9: Proportion of mapped reads on different regions of the soybean genome (oligo-alginate treatment).

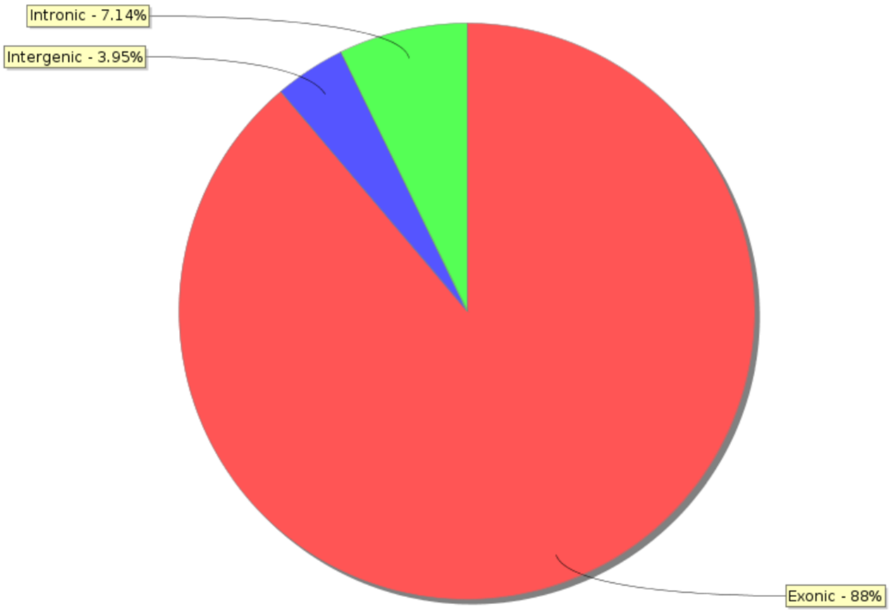


Figure 10: Proportion of mapped reads on different regions of the soybean genome (oligo-chitin treatment).

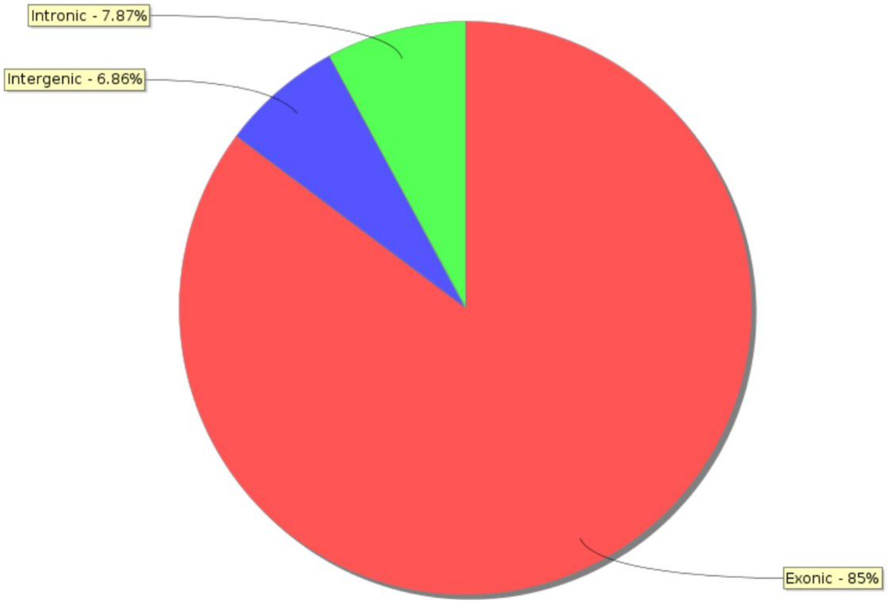


Figure 11: Proportion of mapped reads on different regions of the soybean genome (oligo-chitosan treatment).

6.3 Gene expression quantification

The gene expression profile for different treatments is summarised below (Figure 12). The gene expression levels were measured in RPKM. The x axis represents different samples and the y axis is the log10 scaled RPKM plus 0.001 value. As the results suggested, gene expression profiles for all samples generally follow a normal distribution.

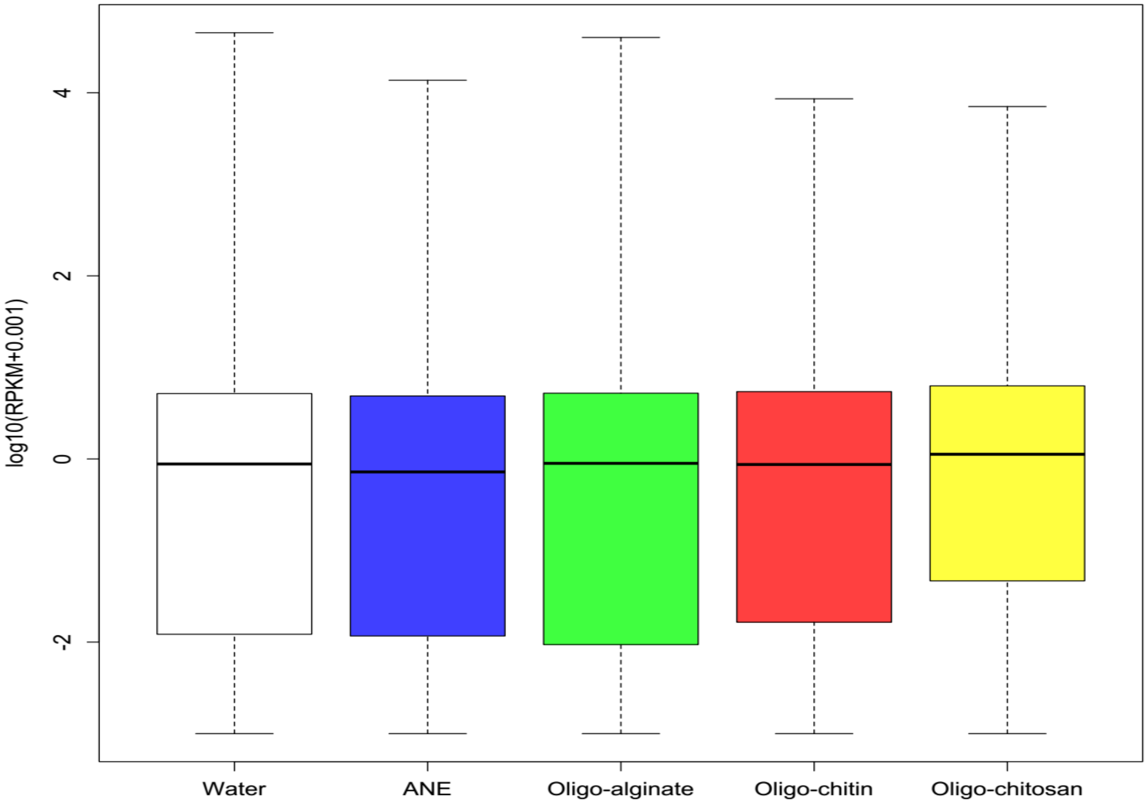


Figure 12: Distribution of gene expression levels of different treatments.

6.4 Number of differentially expressed genes for various treatments

The numbers of differentially expressed genes for the four experimental treatments, in comparison with the water treatment, are summarized below (Figure 13). All treatments showed more down-regulation of genes than up-regulation of genes. The oligo-alginate treatment regulated expression in approximately 1000 genes and ANE regulated expression in approximately 2000 genes, whereas the oligo-chitin and oligo-chitosan treatments regulated expression in approximately 3000 genes each. Up-regulated genes are represented in blue and down-regulated genes are represented in white.

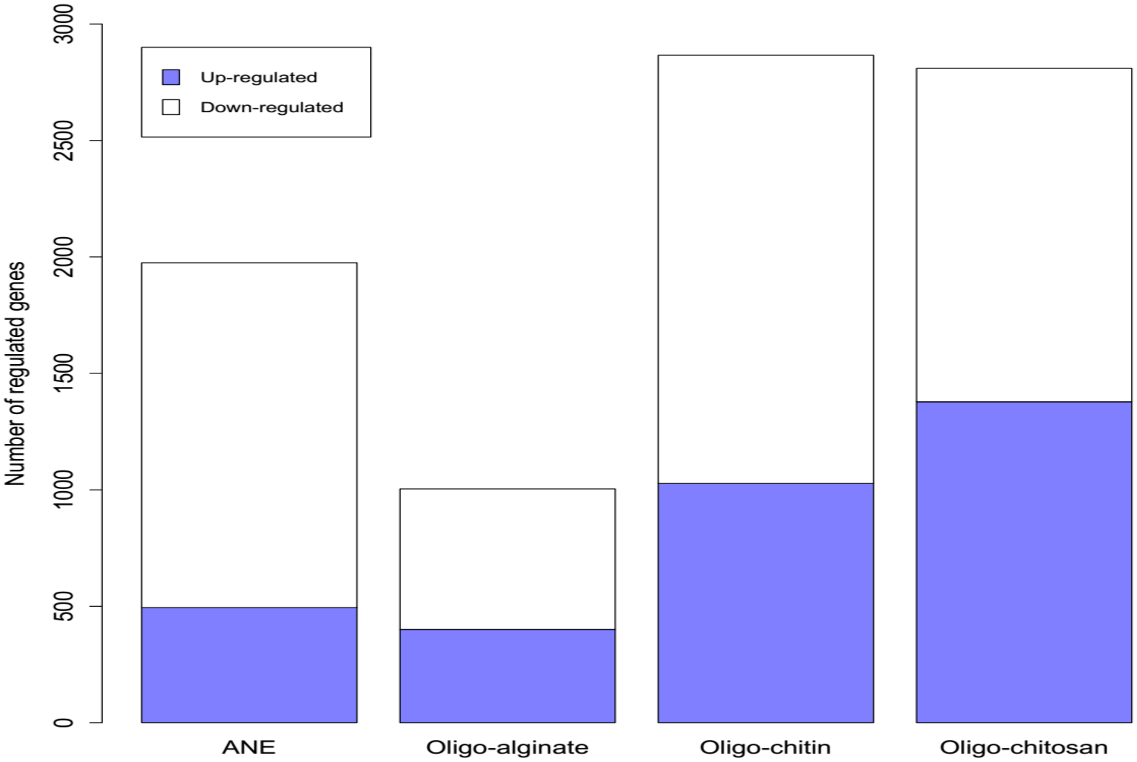


Figure 13: Number of differentially regulated genes after treatment.

6.5 Similarity of treatments based on gene expression pattern.

6.5.1 Expression patterns of all treatments' regulated genes

As shown in Figure 14, the differentially expressed genes of all samples were extracted and the level of their expression was standardized and is denoted by different colors. The scale bar in the legend indicates the Z score. The darker the color, the higher the expression. Similar expression patterns of genes under different treatments were then clustered together. As indicated below, treatments of ANE, followed by oligo-chitin and finally by oligo-chitosan down-regulated the expression of a large number of genes. Treatment with oligo-chitosan also uniquely up-regulated the expression of a large number of genes.

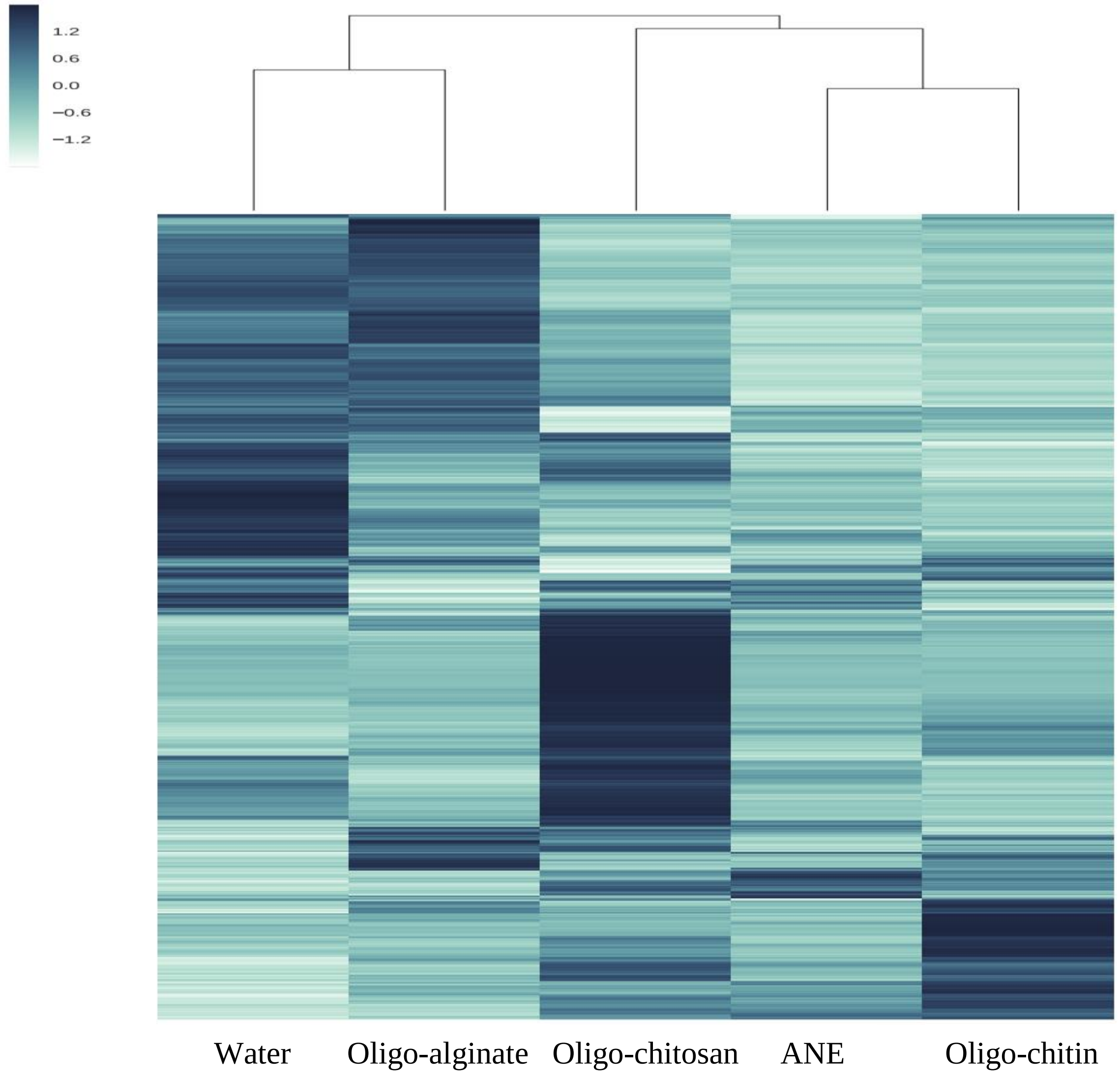


Figure 14: Relationships of different treatments based on collectively regulated genes.

6.5.2 Expression patterns of regulated genes by ANE treatment

ANE treatment altered the expression of 1975 genes compared with water treatment. As shown in Figure 15, treatment with ANE down-regulated the expression of a larger number of genes compared with the number of up-regulated genes. ANE treatment up-regulated genes were similarly up-regulated with treatments of oligo-chitin followed by oligo-chitosan. ANE down-regulated genes were also largely down-regulated following oligo-chitin treatment.

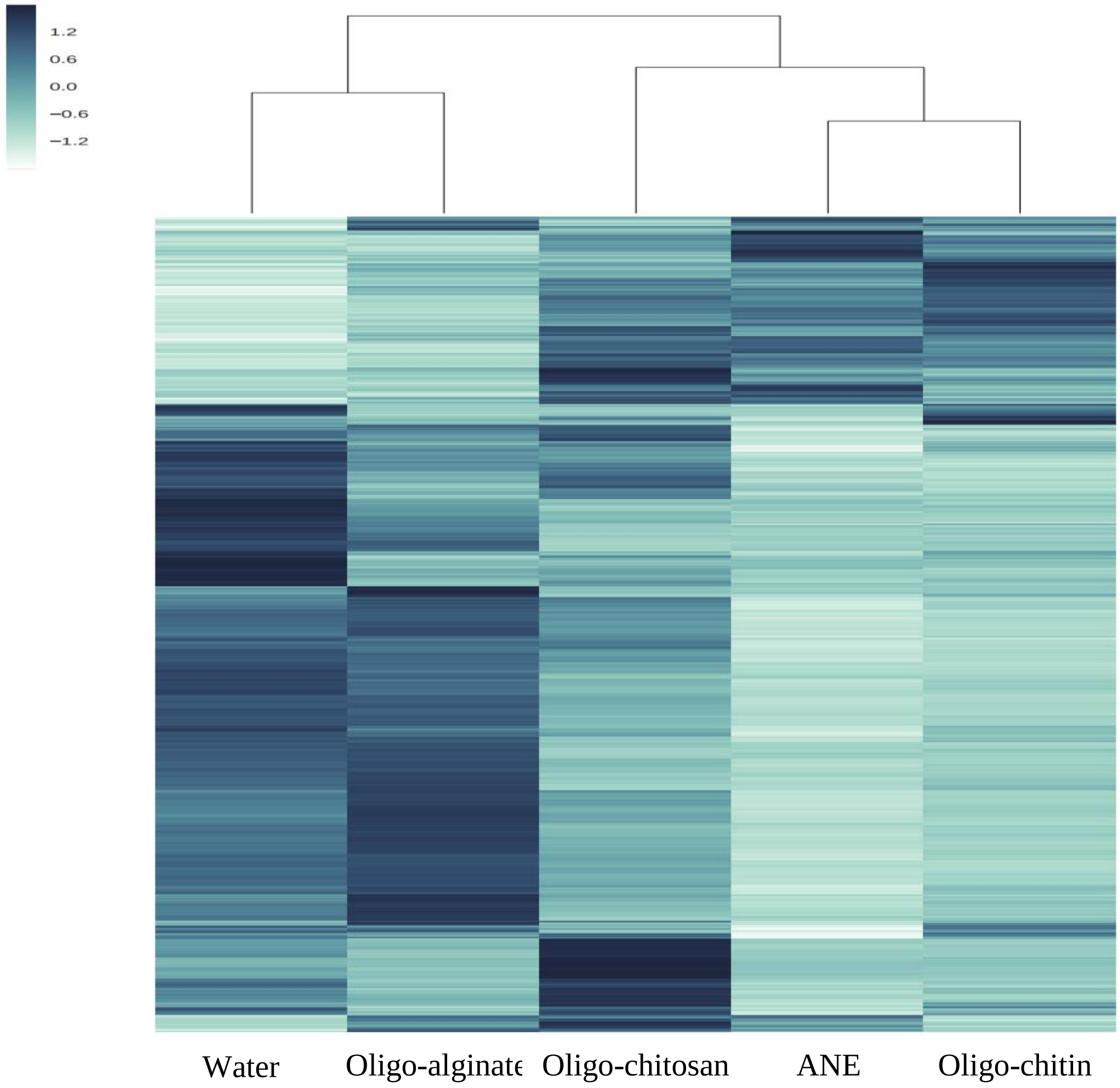


Figure 15: Relationships of different treatments based on ANE regulated genes.

6.5.3 Expression patterns of up-regulated genes by ANE treatment

494 genes were up-regulated after ANE treatment compared with water. As shown in Figure 16, only a small fraction of ANE treatment up-regulated genes were also up-regulated in the oligo-alginate treatment, whereas these same ANE up-regulated genes were also largely up-regulated in the oligo-chitin and oligo-chitosan treatments.

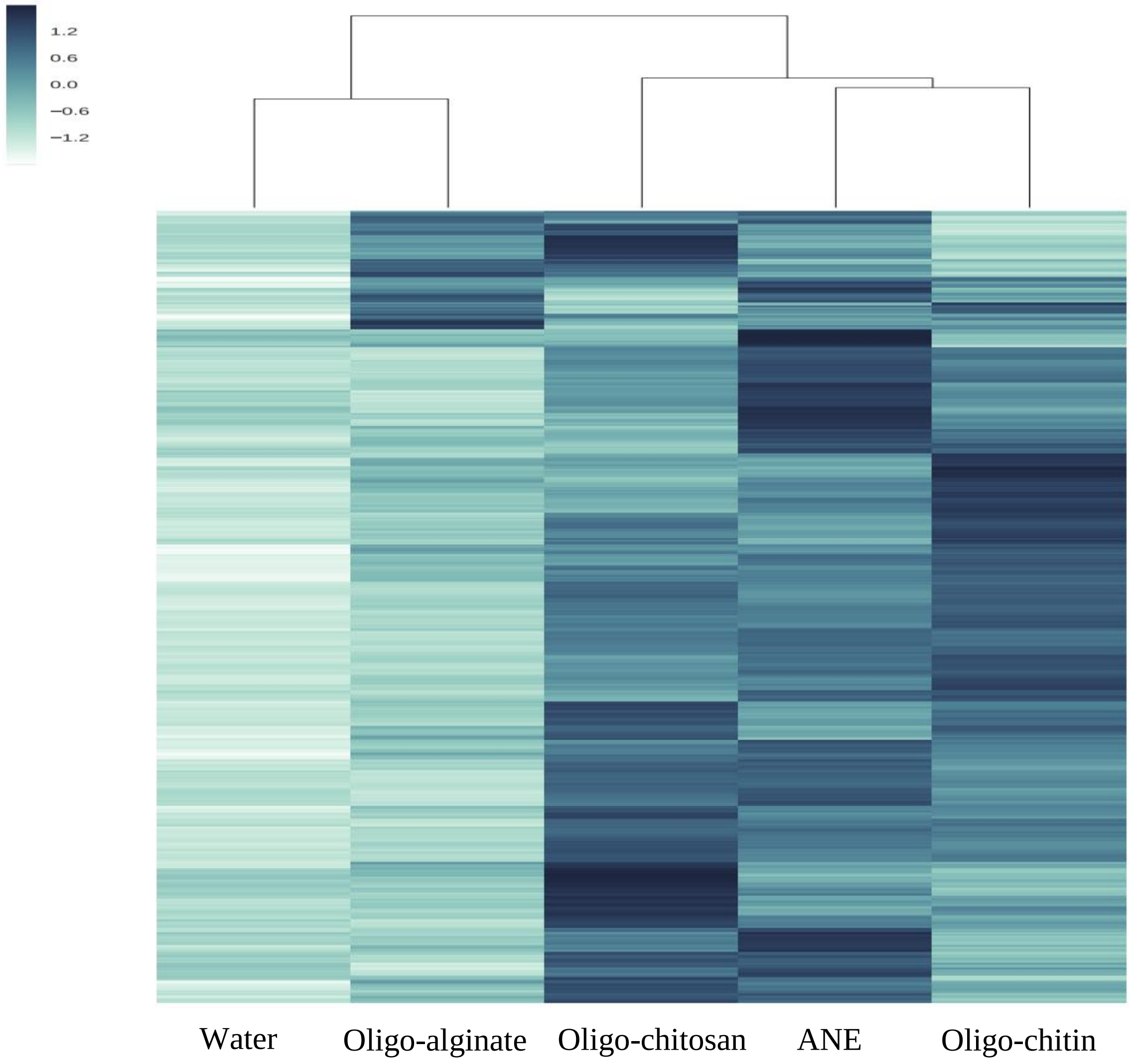


Figure 16: Relationships of different treatments based on ANE up-regulated genes.

6.5.4 Expression patterns of down-regulated genes by ANE treatment

ANE treatment down-regulated expression of 1481 genes compared with water treatment. Many of these ANE down-regulated genes were also down-regulated under oligo-chitin and, to a lesser extent, oligo-chitosan treatment as shown in Figure 17. Only a small fraction of the ANE down-regulated genes overlapped with those down-regulated under oligo-alginate treatment.

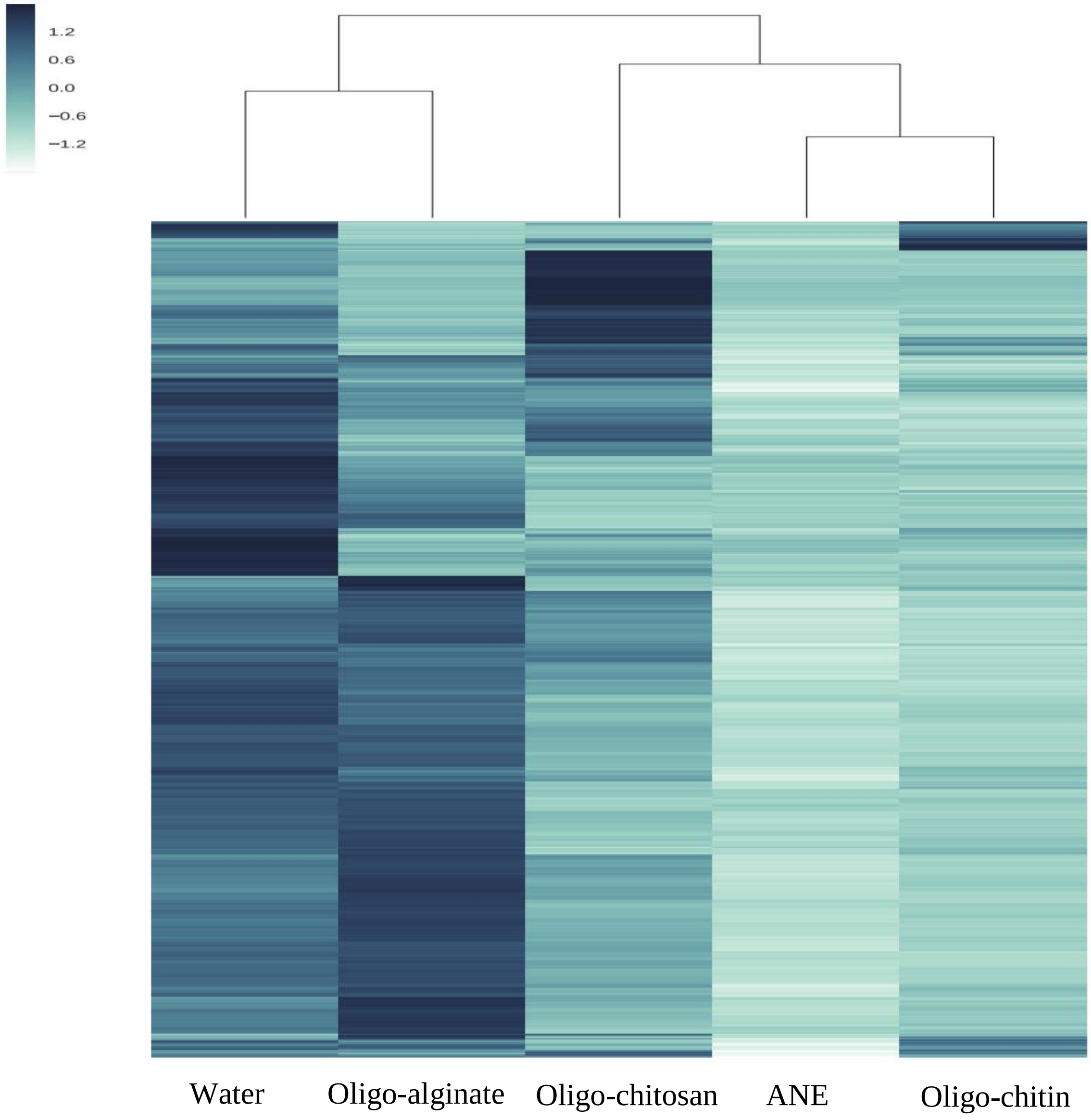


Figure 17: Relationships of different treatments based on ANE down-regulated genes.

6.6 Similarity of treatments based on the number of commonly regulated genes.

6.6.1 Commonly regulated genes between treatments with ANE and oligosaccharides.

As shown in the bar chart in Figure 18, different treatments regulated the expression of different numbers of genes; ANE, oligo-chitin, -chitosan and -alginate treatments regulated the expression of 1975, 2866, 2810 and 1004 genes, respectively. In terms of commonly regulated genes after treatment as shown in the Venn diagram in Figure 18, all treatments commonly regulated the expression of 95 genes. Oligo-chitin and ANE treatments commonly regulated the expression of 1168 genes. The expression of 718 genes were commonly regulated in oligo-chitosan and ANE treatments, whereas the expression of only 265 genes were commonly regulated in oligo-alginate and ANE treatments. In terms of the unique action of different treatments, ANE treatment uniquely regulated the expression of 578 genes and oligo-chitin, -chitosan, and -alginate uniquely regulated the expression of 1102, 1593 and 411 genes respectively.

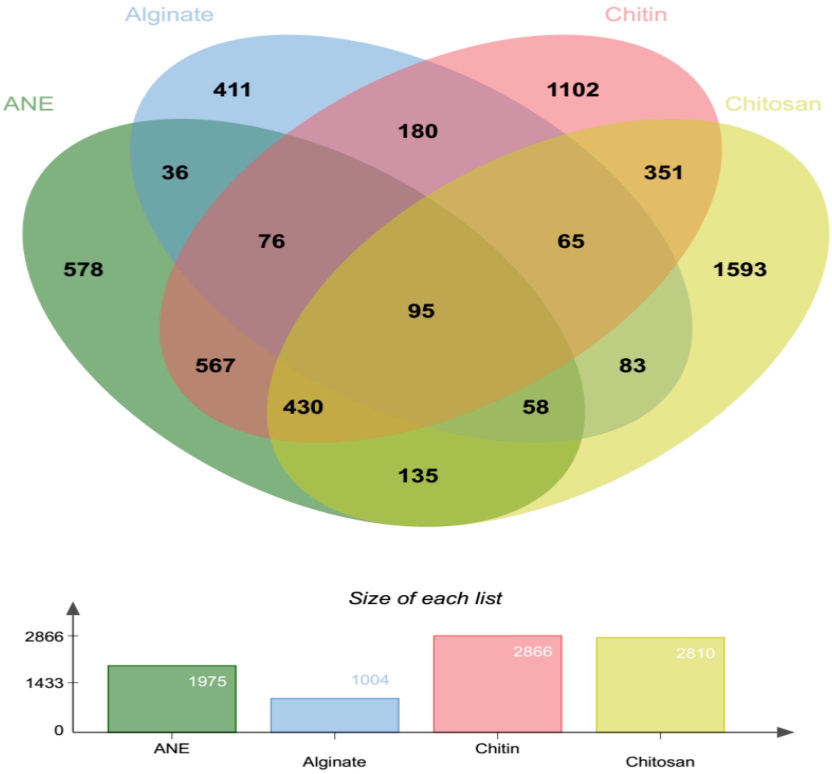


Figure 18: Commonly regulated genes between ANE and oligosaccharide treatments.

6.6.2 Commonly up-regulated genes between treatments of ANE and oligosaccharides.

As shown in the bar chart in Figure 19, ANE and oligo-alginate up-regulated similar numbers of genes, whereas oligo-chitin and -chitosan both up-regulated the expression of more than 1000 genes. Treatments of ANE and oligo-alginate only commonly up-regulated the expression of 77 genes. Oligo-chitin and ANE commonly up-regulated the expression of 247 genes, which accounts for half the number of up-regulated genes in ANE treatment. Oligo-chitosan shared 195 commonly up-regulated genes with ANE. Interestingly, among these 247 and 195 up-regulated genes, 124 overlapped. There were only 26 genes commonly up regulated under all treatments.

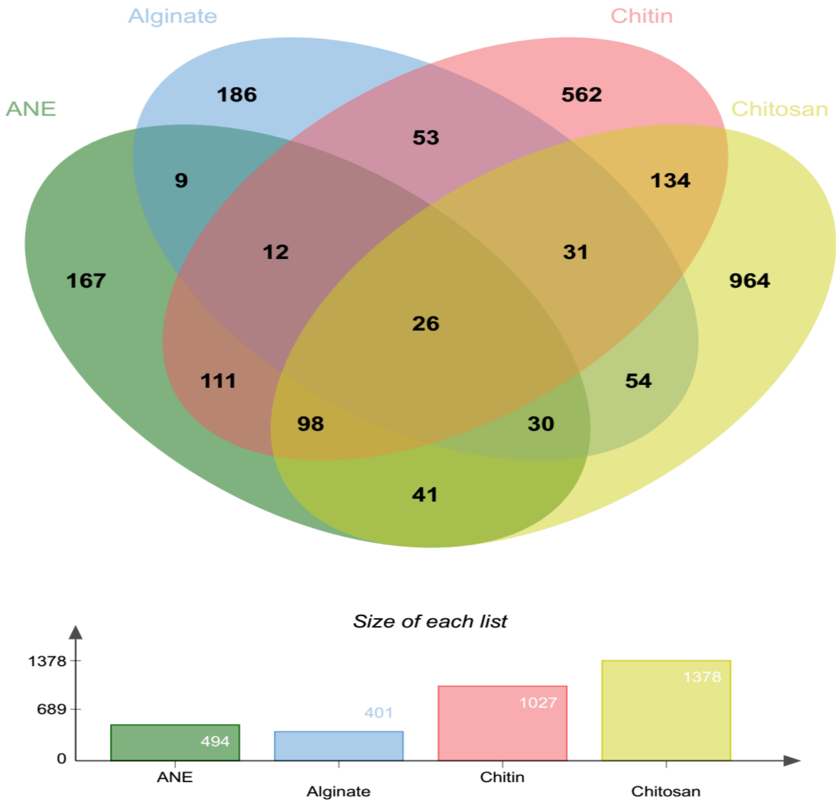


Figure 19: Commonly up-regulated genes between ANE and oligosaccharide treatments.

6.6.3 Commonly down-regulated genes between treatments of ANE and oligosaccharides.

Concerning down-regulated genes, 35 genes were commonly down-regulated by all treatments. Intriguingly, the expression of 909 genes was down-regulated in both ANE and oligo-chitin treatments, among which, 359 genes were also down-regulated in oligo-chitosan treatment. Interestingly, the expression of 467 genes was exclusively down-regulated in the treatments of ANE and oligo-chitin, whereas, expression of only 86 genes was exclusively down-regulated in ANE and oligo-chitosan treatments, and only 25 genes were exclusively down-regulated in ANE and oligo-alginate treatments. As the results suggest, oligo-chitin treatment showed a larger overlap of down-regulated genes with that of ANE. The majority of commonly down-regulated genes in treatments of oligo-chitosan and ANE were actually also down-regulated in the oligo-chitin treatment (see Figure 20).

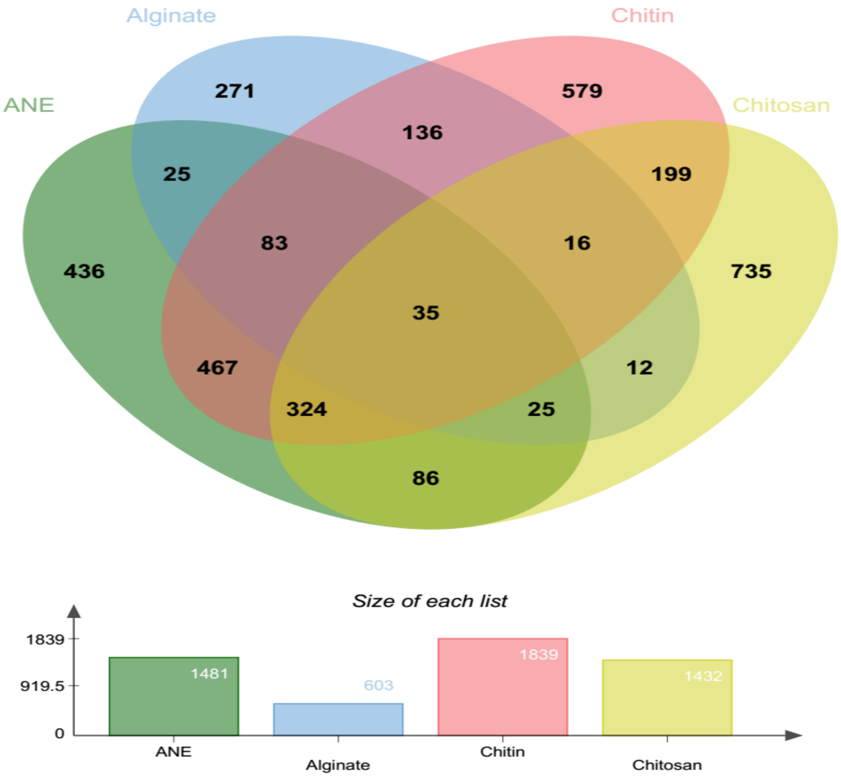


Figure 20: Commonly down-regulated genes between ANE and oligosaccharide treatments.

6.7 Function annotation of genes regulated by ANE treatment

6.7.1 Function summary of up-regulated genes in ANE treatment

ANE treatment up-regulated the expression of 494 genes (see Figure 19), 268 of which were functionally characterized. The detailed functions of specific genes up-regulated by ANE are listed in Appendix 1. The general functions of these up-regulated genes are summarized in Figure 21. Different colors represent the different treatments and the size of the colored areas is not proportional to the number of the responsive genes. As suggested, a large number of ANE up-regulated genes were also up-regulated by oligo-chitin and oligo-chitosan. These commonly up-regulated genes are mainly involved in various stress responses, transporting processes and catabolic (energy producing) processes. Furthermore, all treatments up-regulated genes involved in flavonoid and terpenoid biosynthesis and those responsible for the scavenging of reactive oxygen species (ROS).

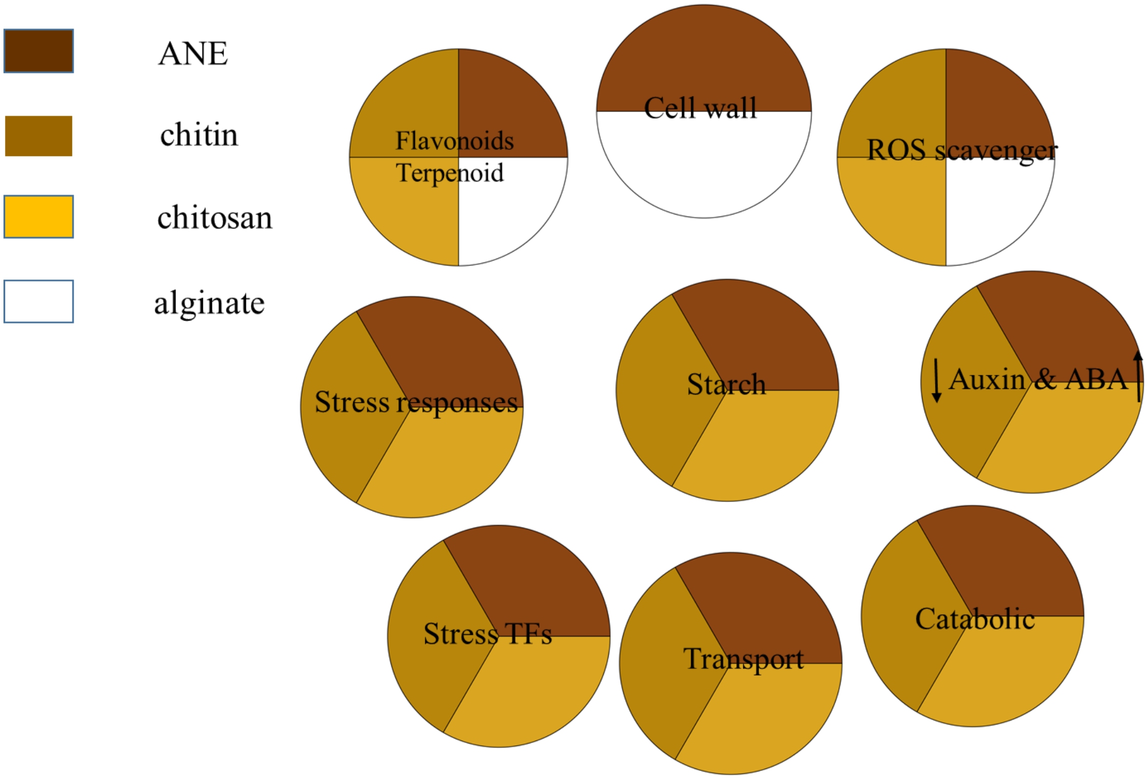


Figure 21: Functions of ANE up-regulated genes

6.7.2 Function summary of down-regulated genes by ANE treatment

ANE treatment down-regulated the expression of 1481 genes, a large number of which were also down-regulated in oligo-chitin and oligo-chitosan treatments (see Figure 20). Detailed functions of these ANE down-regulated genes are listed in the Appendix 2. Figure 22 summarize their general functions. It is clear that treatments of ANE, oligo-chitin and oligo-chitosan commonly down-regulated genes involved in protein, fatty acid and DNA synthesis and those associated with cell proliferation processes. Treatments of ANE and oligo-chitin also commonly down-regulated genes involving in cell expansion. There were few commonly down-regulated genes between oligo-alginate and ANE, the functions of which are not summarized here.

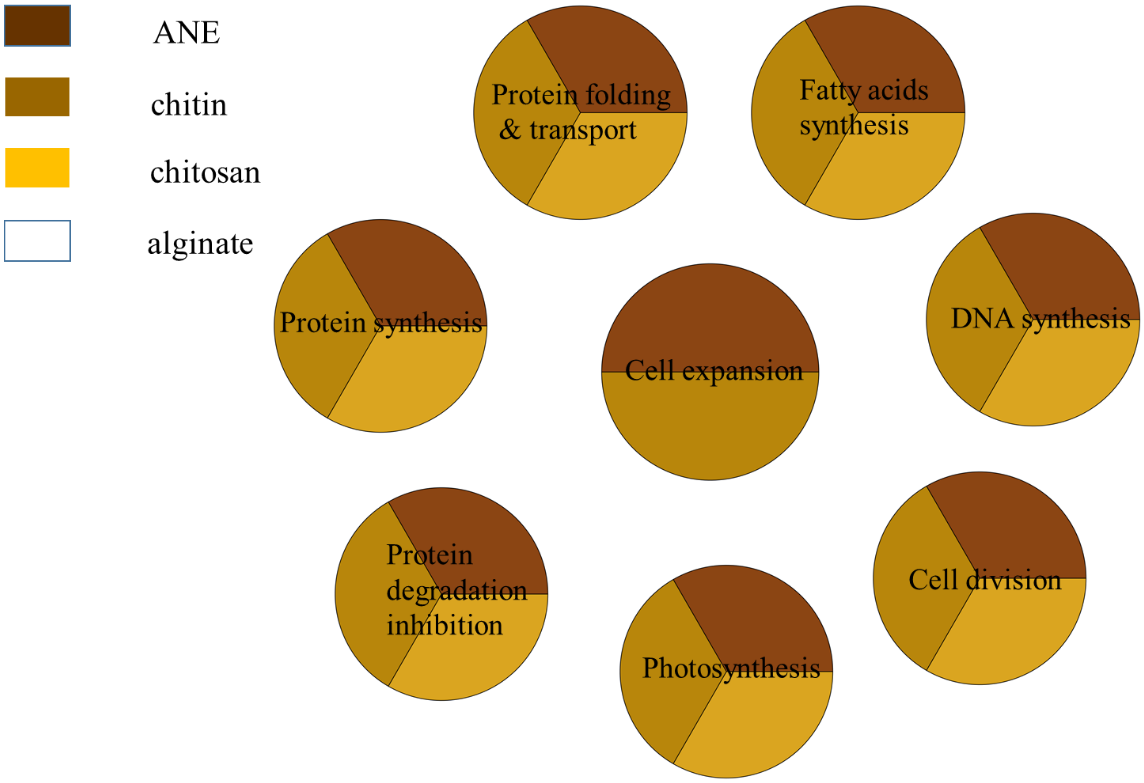


Figure 22: Functions of ANE down-regulated genes

6.8 Biological pathways regulated by all treatments

Based on corrected P values, the top 30 enriched pathways regulated by all treatments were selected and compared (see Figures. 23 through 30). The y-axis of each graph represents the various involved pathways and the x-axis represents the rich factor, with the rich factor calculated in the following way: For a specific pathway, the number of genes mapped to that pathway that were up- or down-regulated divided by the number of total genes mapped to that pathway. Thus, the rich factor represents the relative regulation of one specific pathway. The size of the circle in the legend represents the number of mapped genes for one specific pathway in a regulated gene set. The P value is represented by different colors, which acts as a combined indicator for the enrichment of one pathway.

6.8.1 Comparison of treatments up regulated biological pathways.

Figure 23 through 26 show that, all treatments up-regulated a number of genes involved in secondary metabolic pathways (as indicated by the red arrows). Secondary metabolites in plants are typically involved in plant stress and defense responses. Terpenoids and flavonoids are two important plant secondary metabolites, the biosynthetic pathways of which were also enriched in the up-regulated gene set of all treatments (as indicated by the blue and amber arrows respectively).

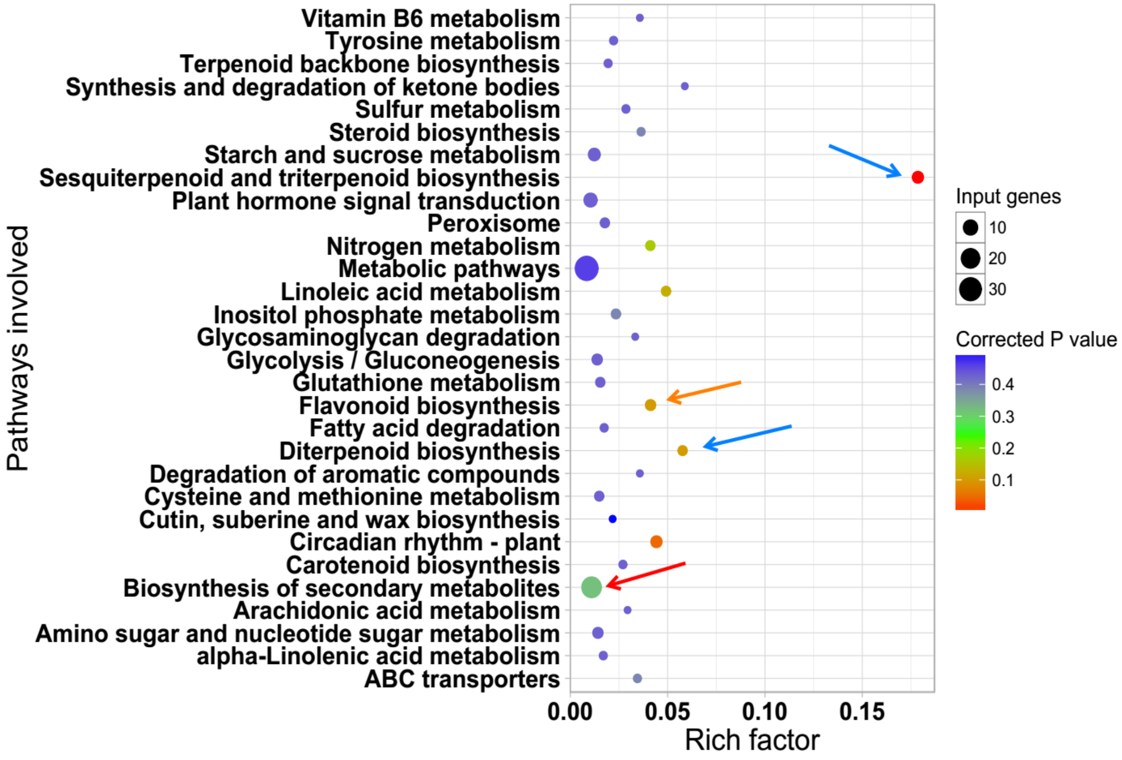


Figure 23: Top 30 ANE up-regulated genes’ involved pathways.

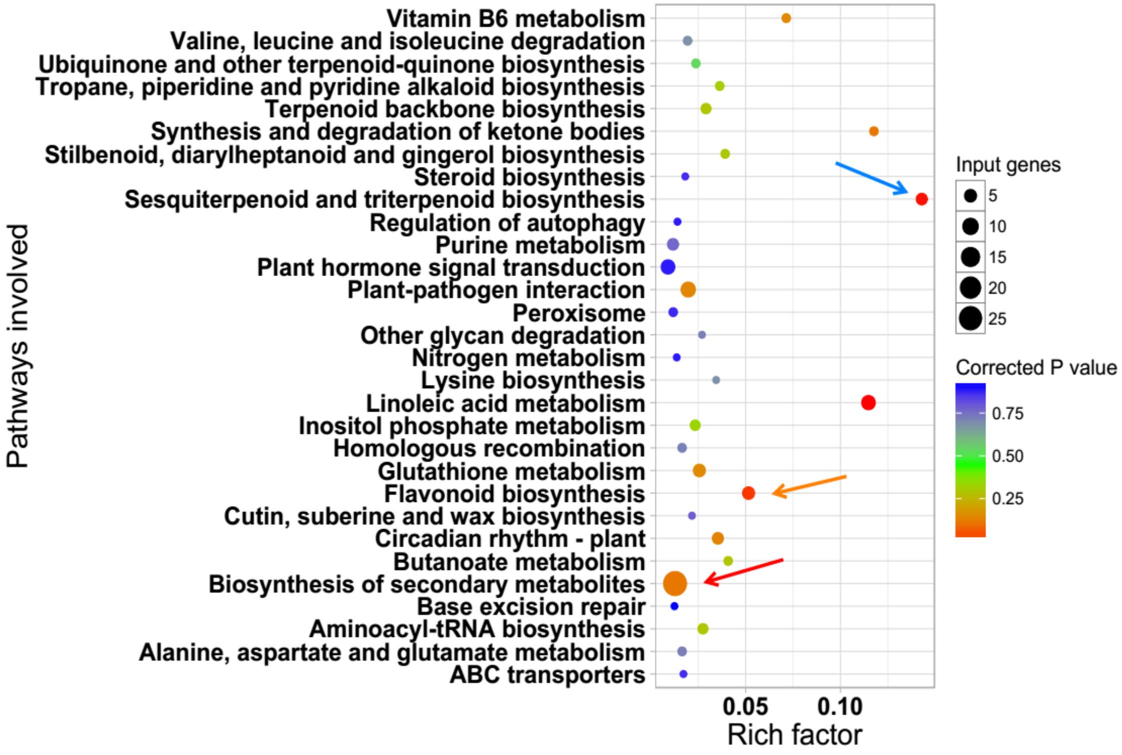


Figure 24: Top 30 oligo-alginate up-regulated genes’ involved pathways.

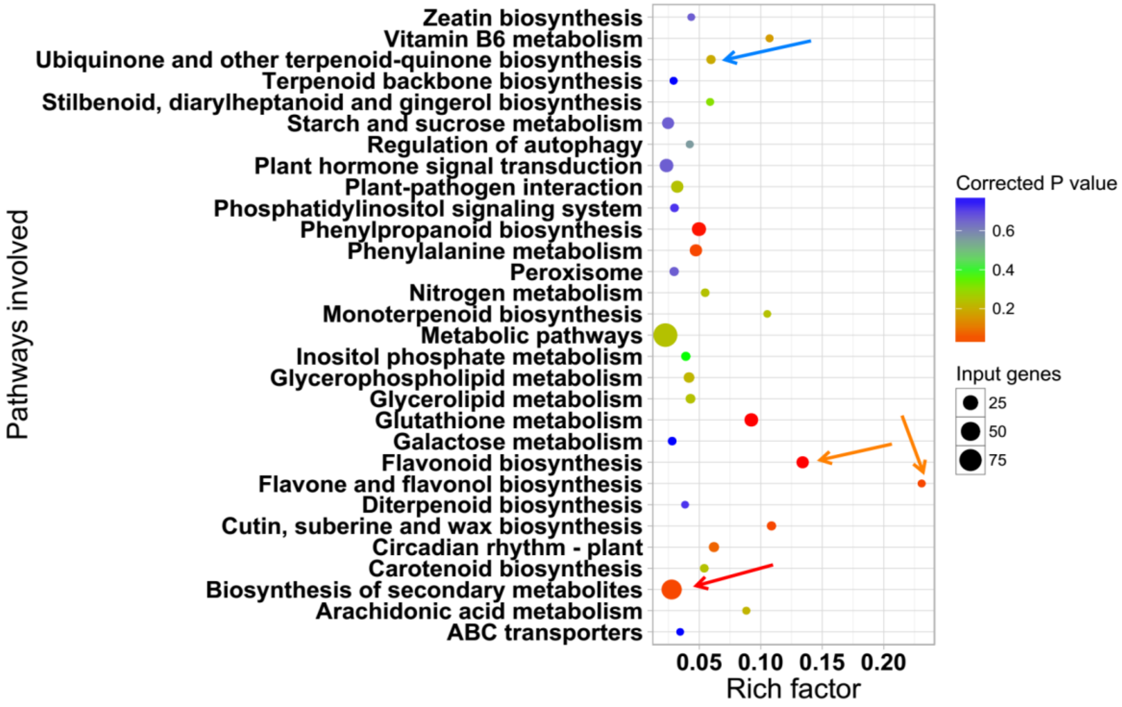


Figure 25: Top 30 oligo-chitin up-regulated genes’ involved pathways.

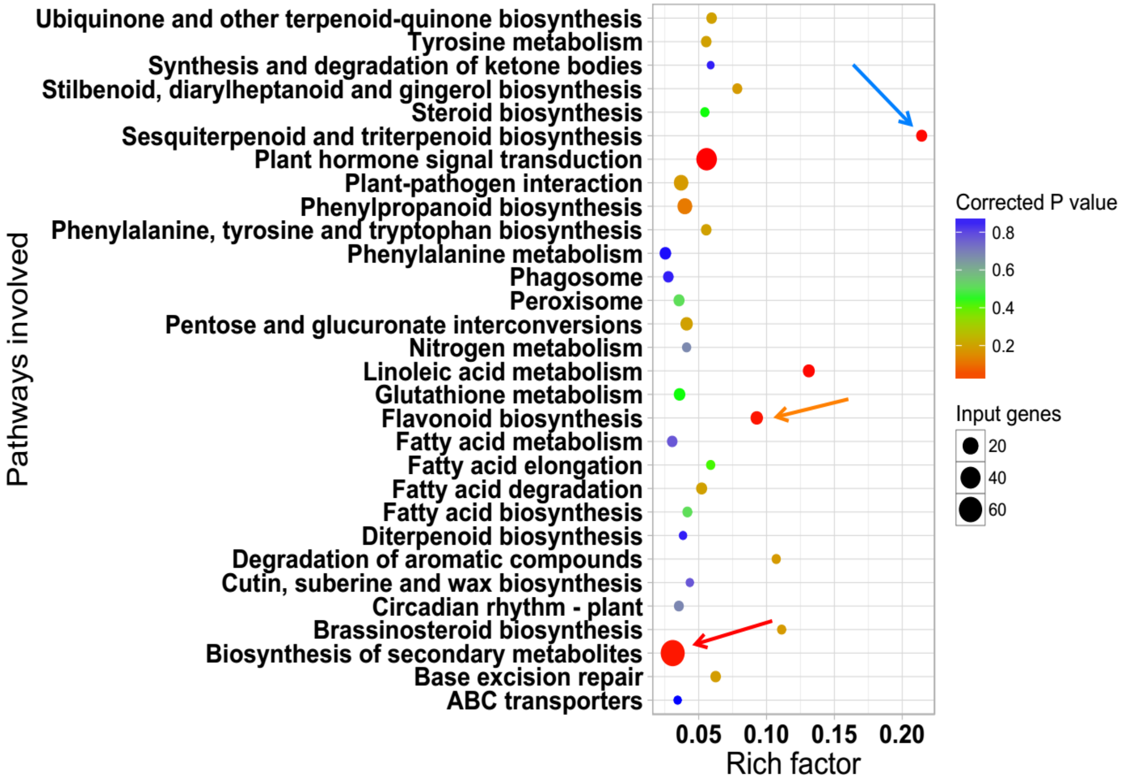


Figure 26: Top 30 oligo-chitosan up-regulated genes’ involved pathways.

6.8.2 Comparison of treatments' down-regulated biological pathways.

Figures 27 through 30 show the treatments' down-regulated genes' involved pathways. All treatments down-regulated a number of genes involved in ribosomal related pathways. The number of down-regulated genes was significantly larger following treatment with ANE, oligo-chitin and oligo-chitosan as compared to that following oligo-alginate treatment (as indicated by the green arrows). In addition, oligo-alginate and oligo-chitosan down-regulated photosynthetic pathways, whereas, ANE and oligo-chitin treatments down-regulated genes related to DNA replication. Fanilly, oligo-chitosan treatment down-regulated genes involved with amino acid and aminoacyl-tRNA synthesis.

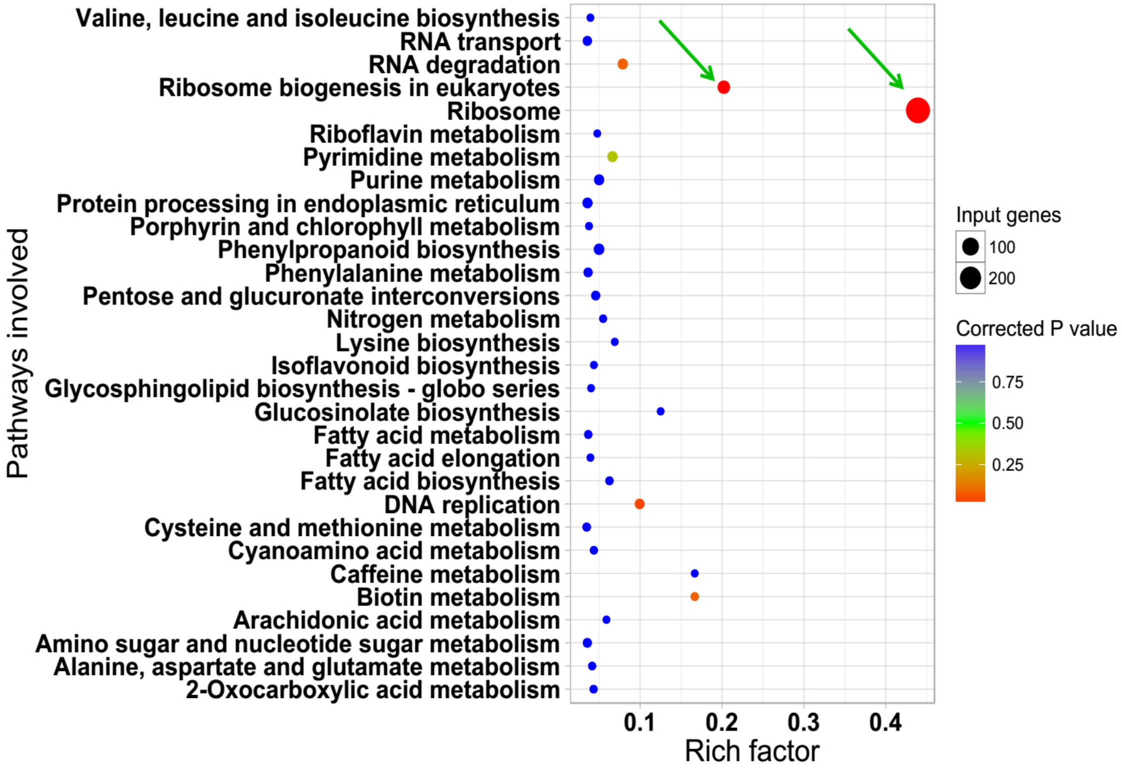


Figure 27: Top 30 ANE down-regulated genes' involved pathways.

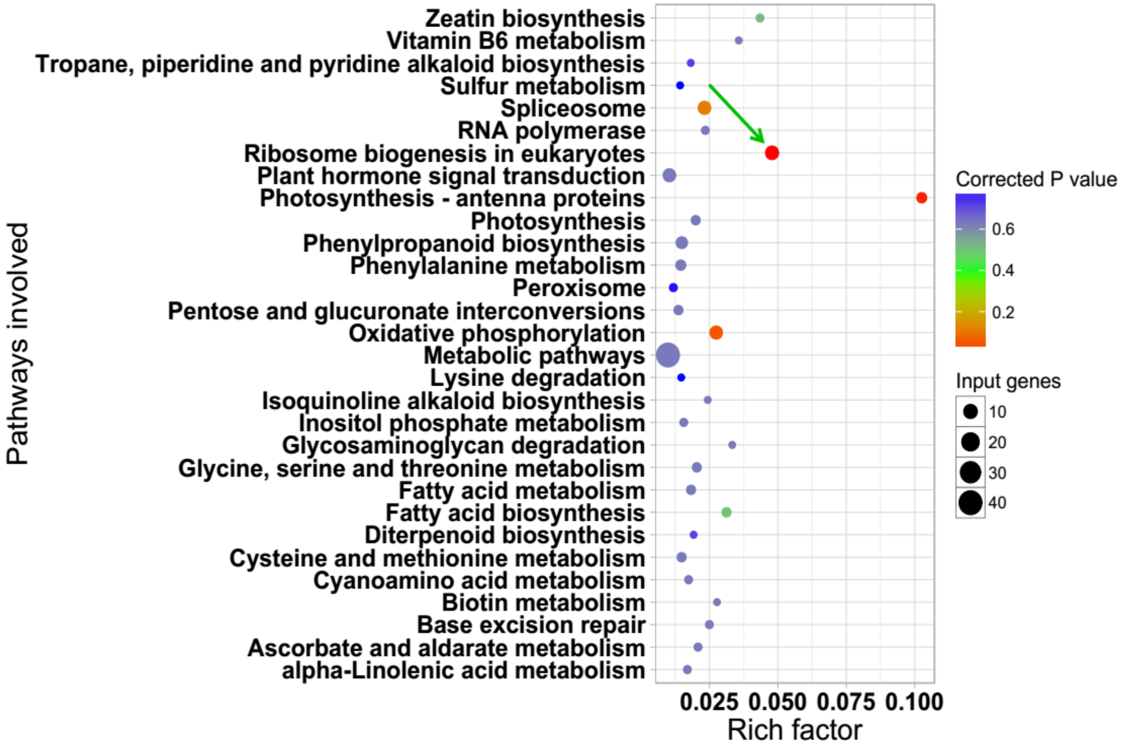


Figure 28: Top 30 oligo-alginate down-regulated genes’ involved pathways.

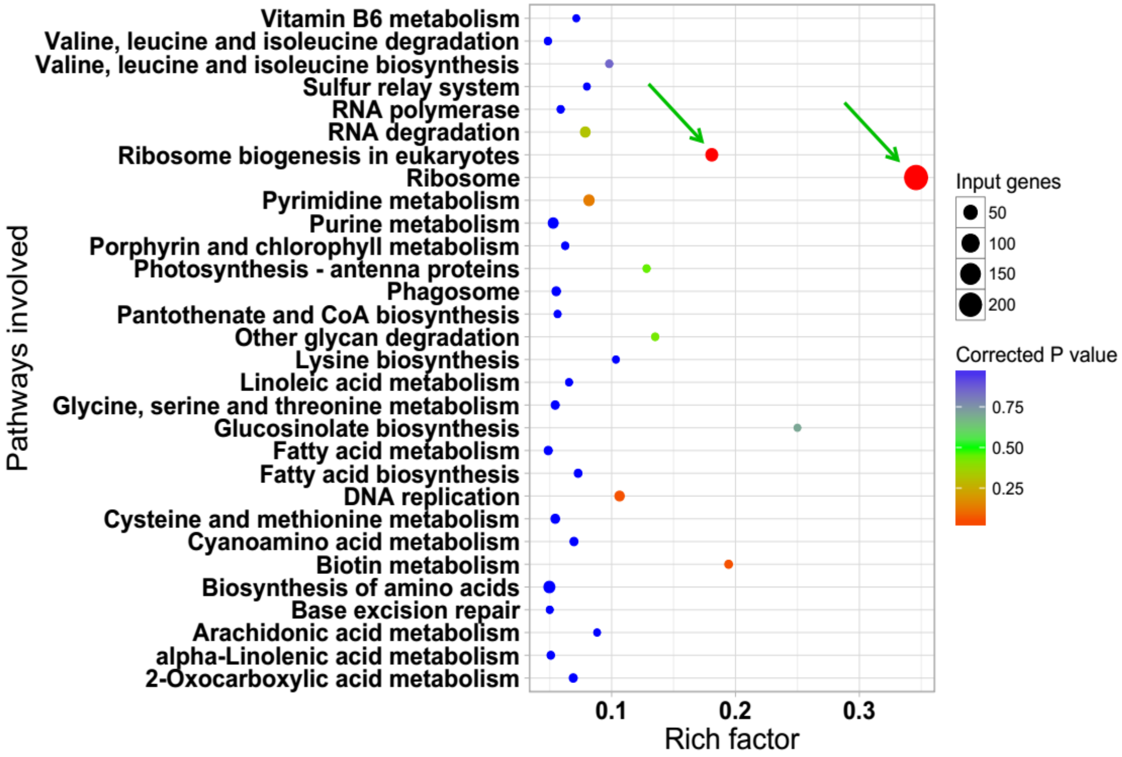


Figure 29: Top 30 oligo-chitin down-regulated genes’ involved pathways.

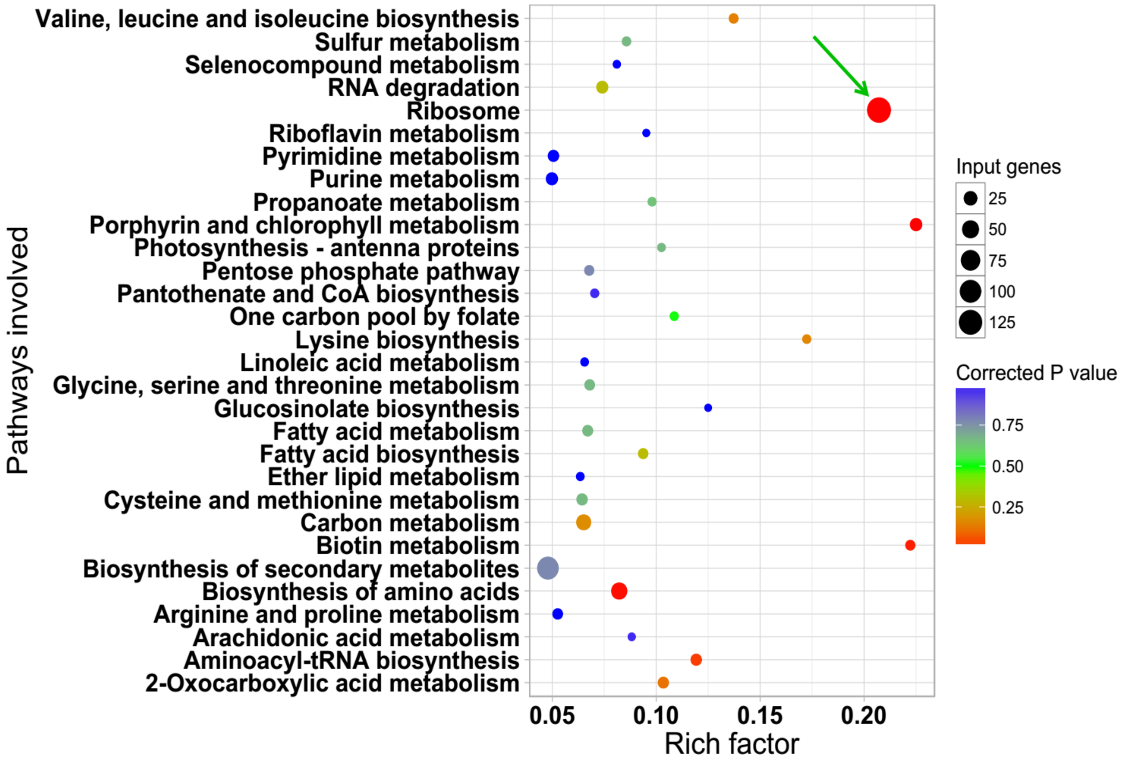


Figure 30: Top 30 oligo-chitosan down-regulated genes’ involved pathways.

6.9 Comparison of treatments’ regulated metabolic processes

ANE and oligosaccharide regulation of various biological processes is detailed in Figures 31 through 62. The legend in the top right hand corner of each figure indicates the total number of biological data points mapped by the number of regulated genes in each treatment. The number for the visible data points show the number of data points mapped to each biological process. The mapped data points are represented by small squares; the regulation of data points is indicated by color (red for up-regulation and blue for down-regulation). The scale bars in each figure represent the gfold value for each data point.

6.9.1 Comparison of treatments’ regulation of terpenoid biosynthesis.

Figures 31 through 34 show the monoterpene and diterpene biosynthetic pathways in plants. Synthesis of intermediates in these pathways is carried out independently in the cytoplasm and chloroplast (shown as blue and yellow boxes in the upper left and right sides of the figures respectively). Intermediates are then transported between the cytoplasm and chloroplast and then take their place in the final steps of the pathways (as shown in the pink box in the lower left corner). Figures 31 through 34 show regulation of the various steps of terpenoid biosynthesis under ANE, oligo-alginate, oligo-chitin and oligo-chitosan treatment respectively. For example, 1975 ANE treatment-regulated genes were mapped to 2145 biological data points (see Figure 31, legend). Among these 2145 data points, 9 were mapped to this pathways. Within these nine data points, six were mapped to up-regulated genes. As the colors of the small squares in the following figures suggest, all treatments up-regulated a number of genes involved in various steps of monoterpene and diterpene biosynthesis.

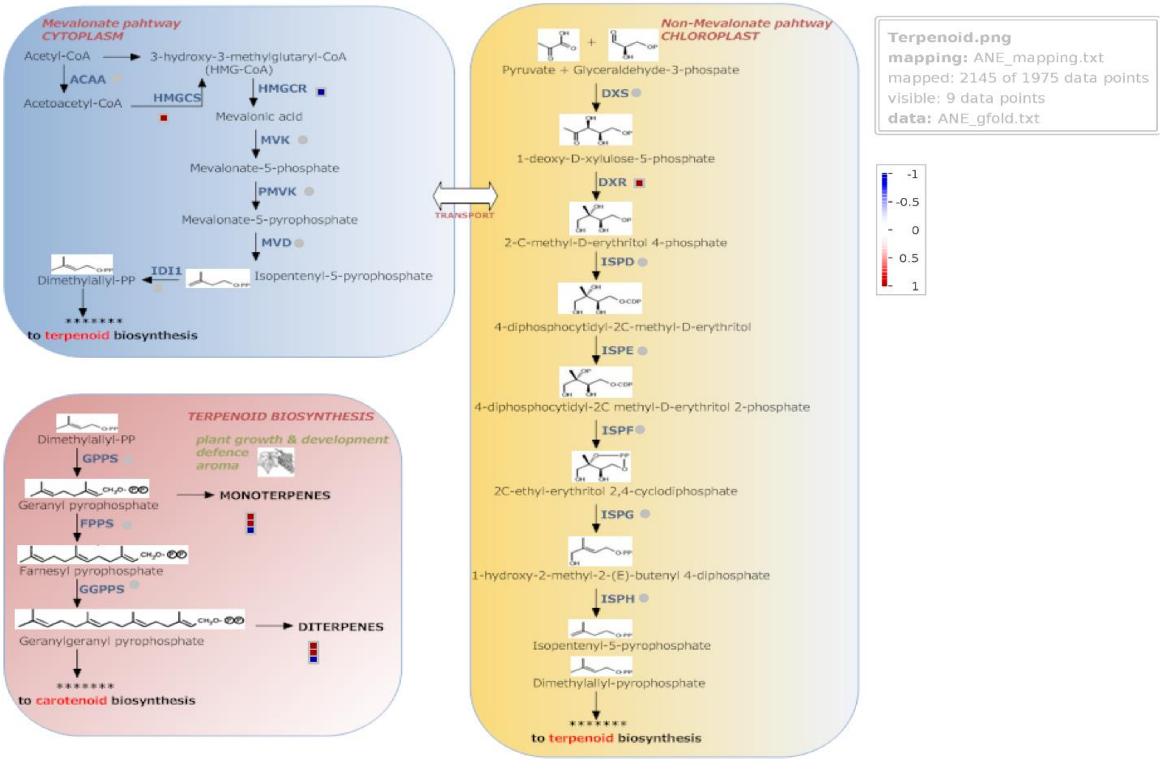


Figure 31: ANE-regulated genes involved in terpenoid biosynthesis.

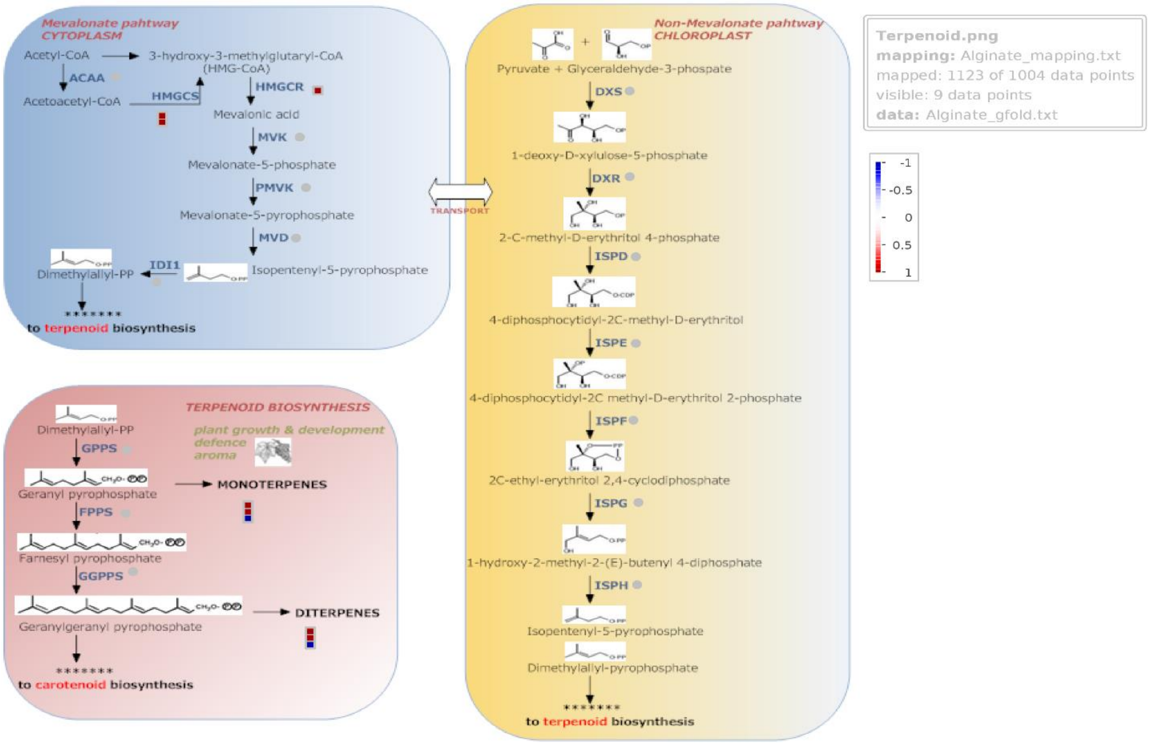


Figure 32: oligo-alginate-regulated genes involved in terpenoid biosynthesis.

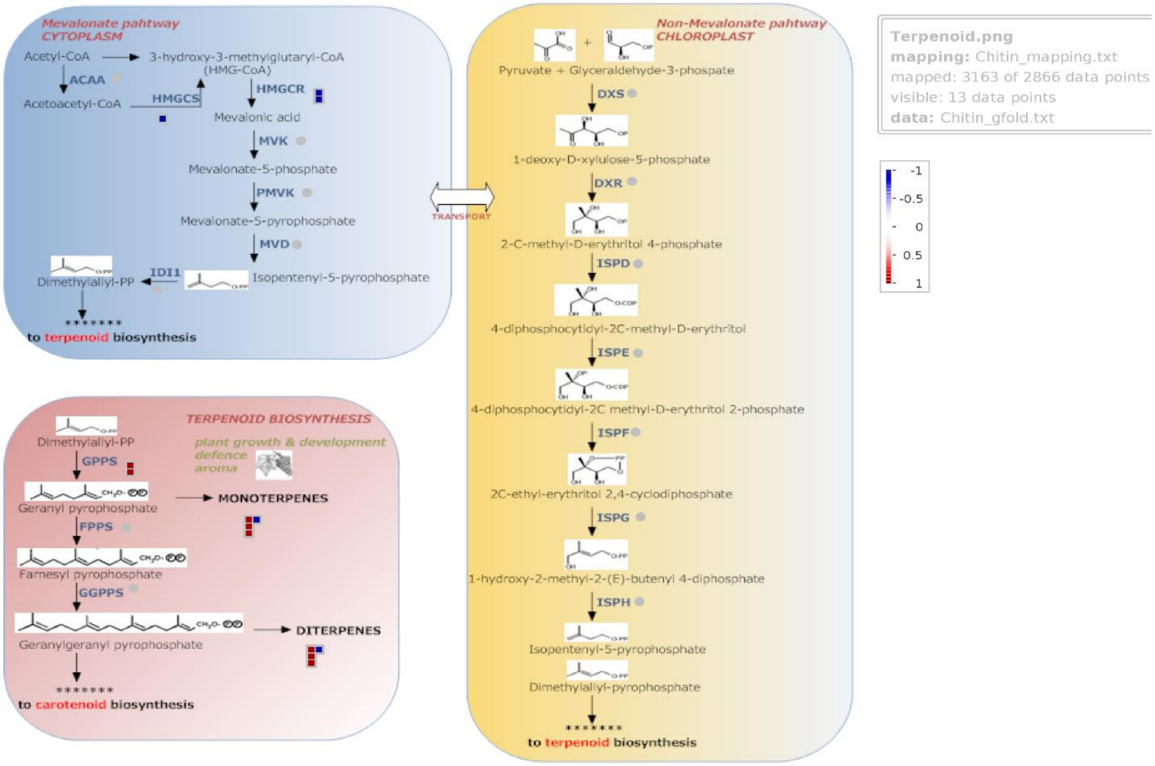


Figure 33: oligo-chitin-regulated genes involved in terpenoid biosynthesis.

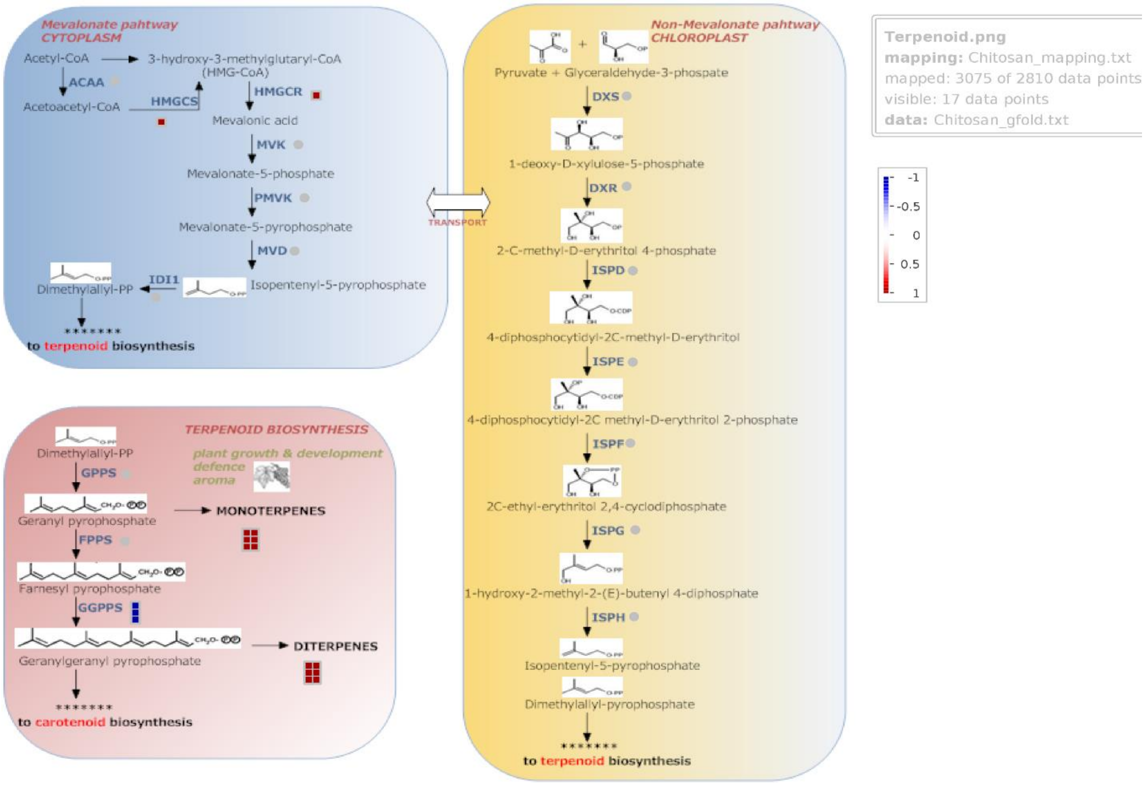


Figure 34: oligo-chitosan-regulated genes involved in terpenoid biosynthesis.

6.9.2 Comparison of treatments' regulation of flavonoid biosynthesis.

As important as the various terpenes are to plants, the versatile flavonoids are essential for plants to deal with various stresses. Figures 35 through 38 represent various flavonoid biosynthetic pathways. Plants can synthesize a suite of structuraly similar flavonoids such as catechin, resveratrol, epicatechin, quercetin, myricetin, naringenin, kaempferol, and, anthocyanins (shown as colored arrows in Figure 35). As suggested by the data points (indicated by the small squares), ANE and oligosaccharide treatments up-regulated these various flavonoid biosynthetic pathways.

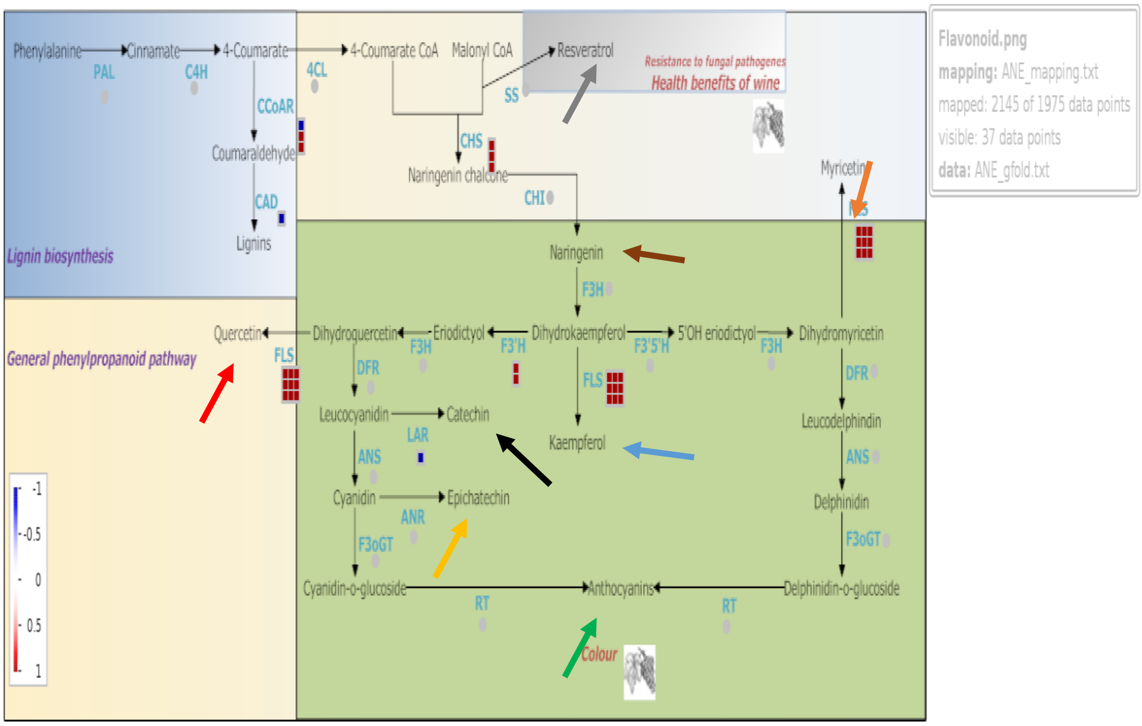


Figure 35: ANE-regulated genes involved in flavonoid biosynthesis.

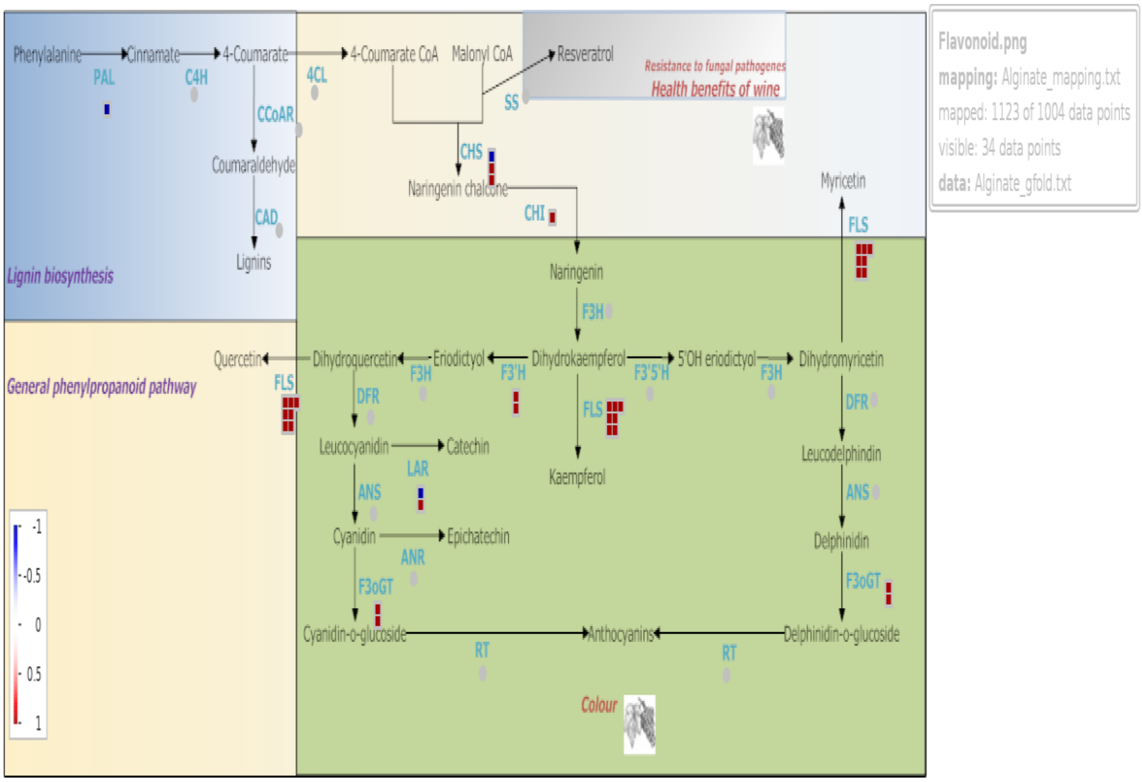


Figure 36: oligo-alginate-regulated genes involved in flavonoid biosynthesis.

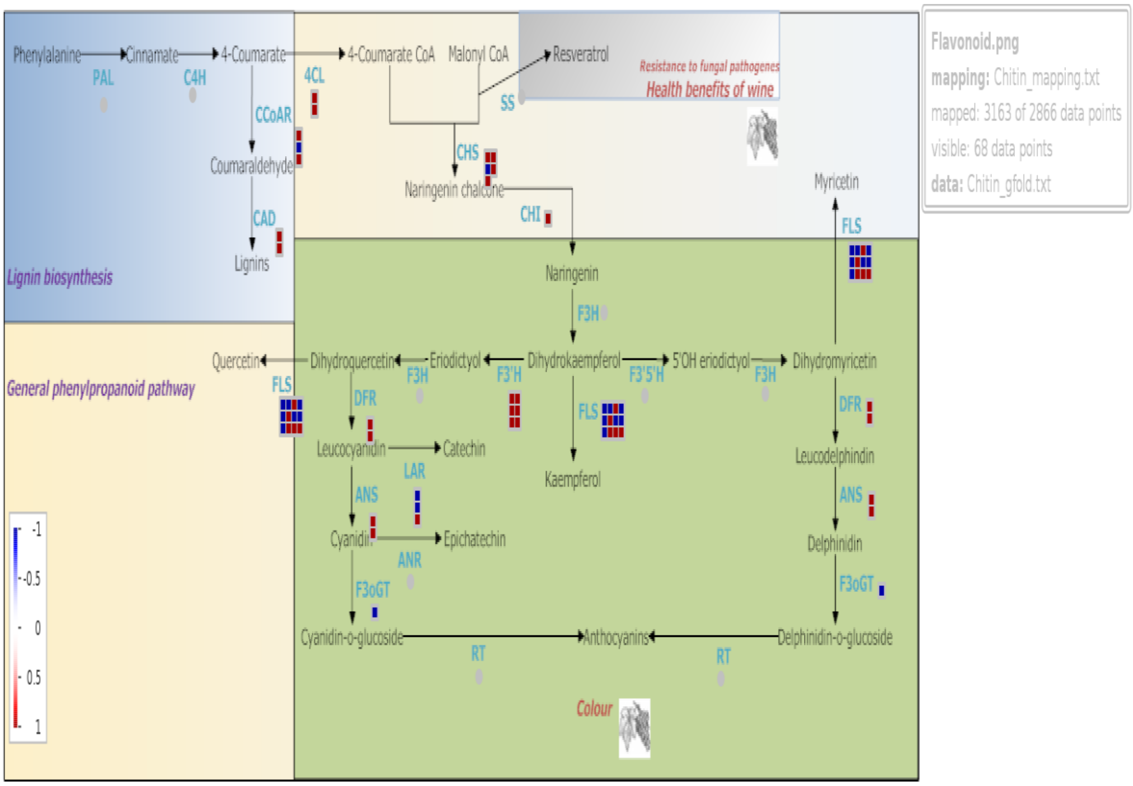


Figure 37: oligo-chitin-regulated genes involved in flavonoid biosynthesis.

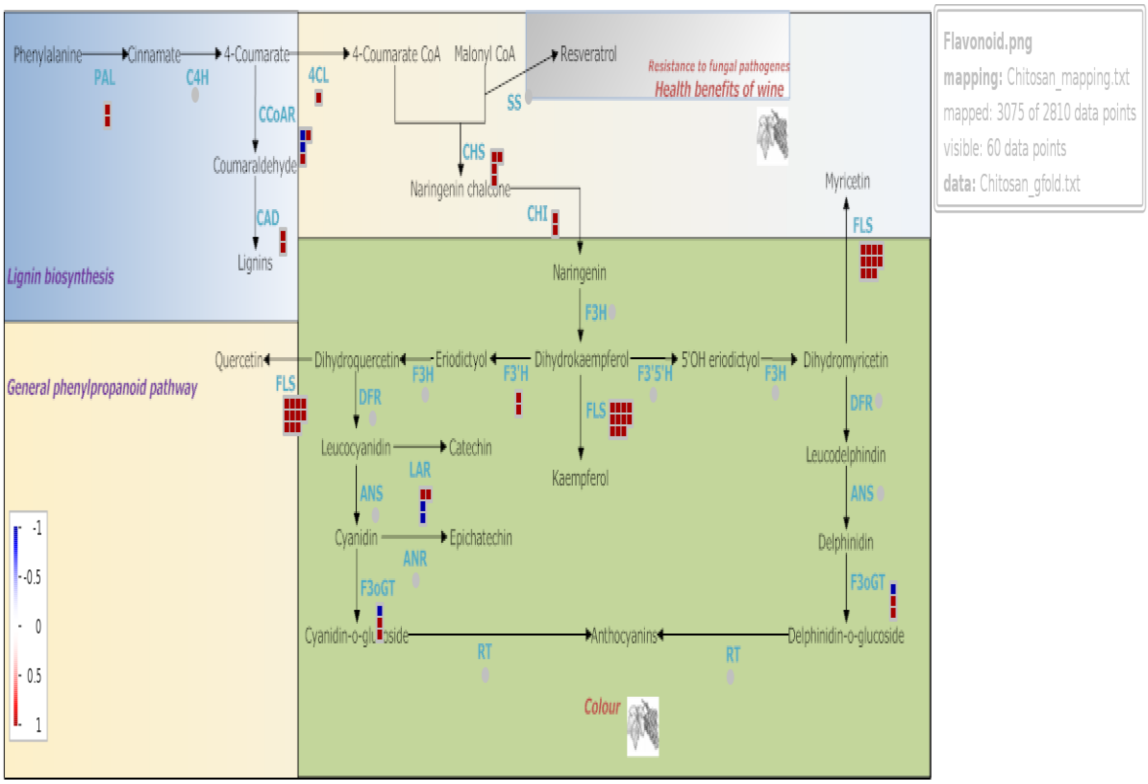


Figure 38: oligo-chitosan-regulated genes involved in flavonoid biosynthesis.

6.9.3 Comparison of general stress responses after treatments

In addition to the up-regulation of secondary metabolite biosynthesis as shown above, treatments of ANE and oligosaccharides also modulated the expression of a large number of genes related to other aspects of plant stress responses. As shown in Figures 39 through 42, all treatments, to differing extents, up-regulated genes involved in plant signaling and plant stress-related hormones such as ABA, ethylene, SA and JA (as indicated in the blue boxes). All treatments also influenced the expression of genes coding for various stress-related transcription factors such as ERF, bZIP, WRKY, MYB and DOF (as indicated in the purple boxes). Furthermore, the expression of a large number of pathogenesis-related protein (PR) encoding genes were up-regulated in all treatments (as indicated in the yellow boxes).

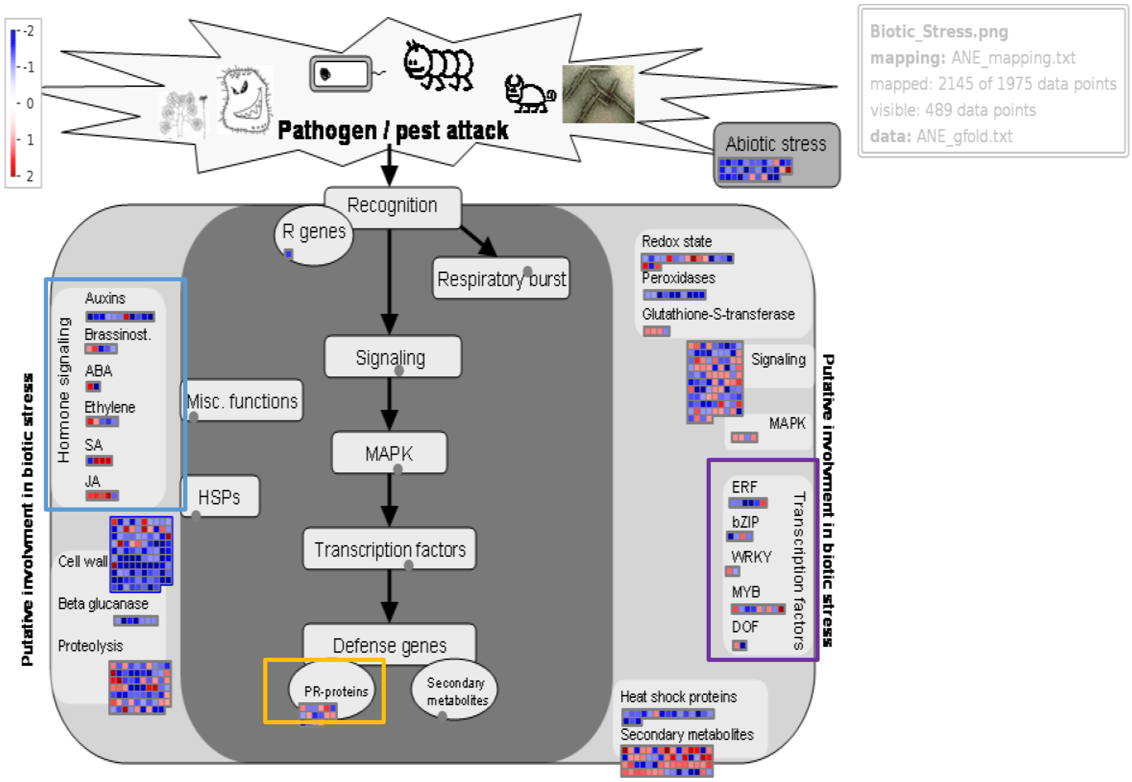
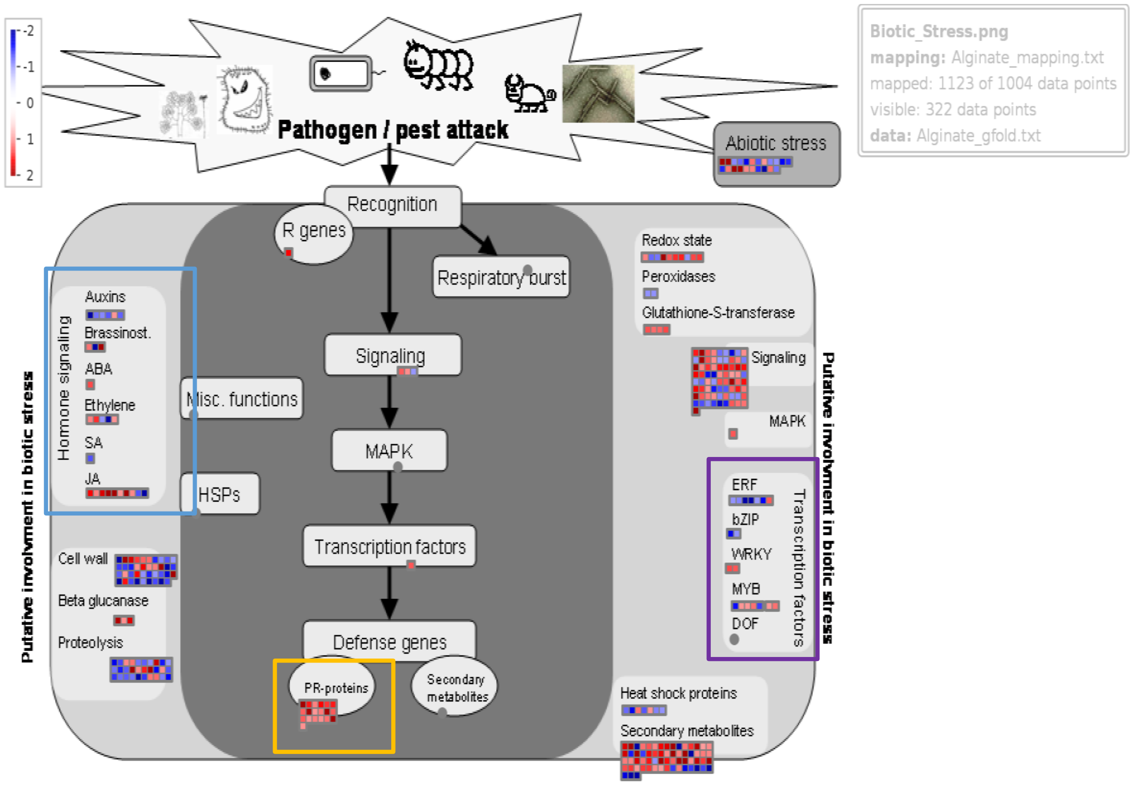
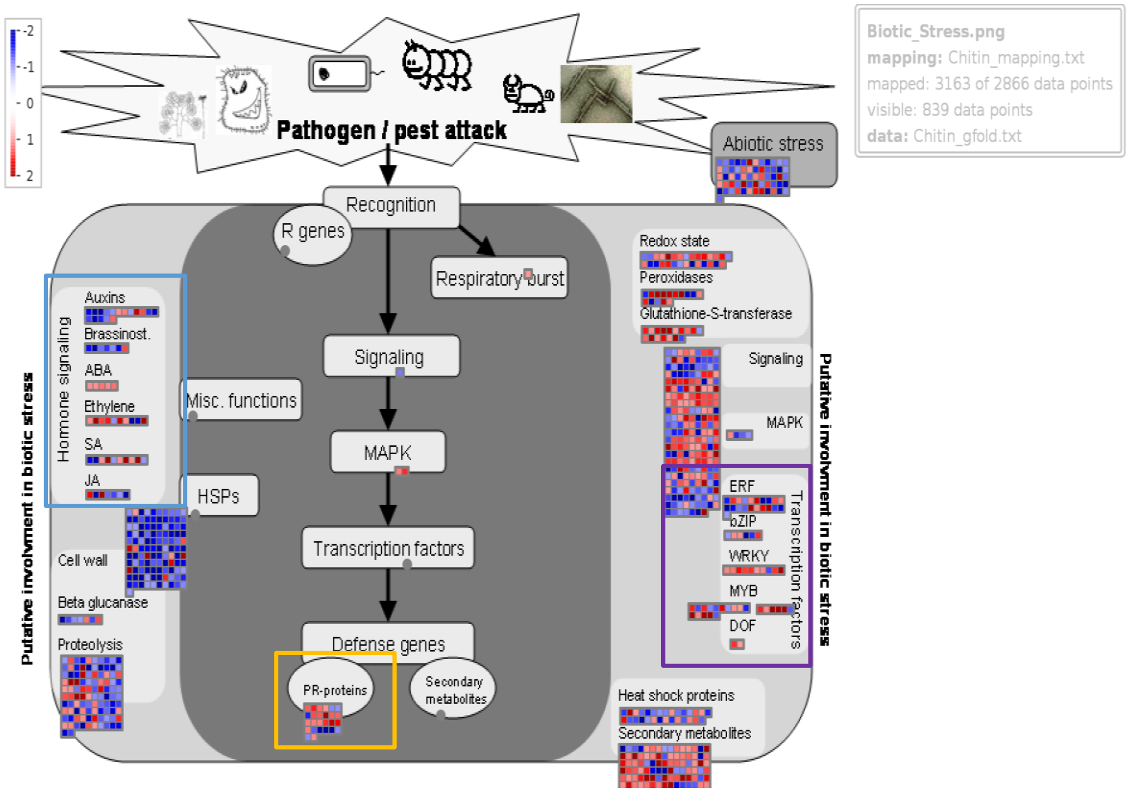


Figure 39: ANE-regulated genes involved in overall stress responses.



Biotic_Stress.png
mapping: Alginate_mapping.txt
mapped: 1123 of 1004 data points
visible: 322 data points
data: Alginate_gfold.txt

Figure 40: oligo-alginate-regulated genes involved in overall stress responses.



Biotic_Stress.png
mapping: Chitin_mapping.txt
mapped: 3163 of 2866 data points
visible: 839 data points
data: Chitin_gfold.txt

Figure 41: oligo-chitin-regulated genes involved in overall stress responses.

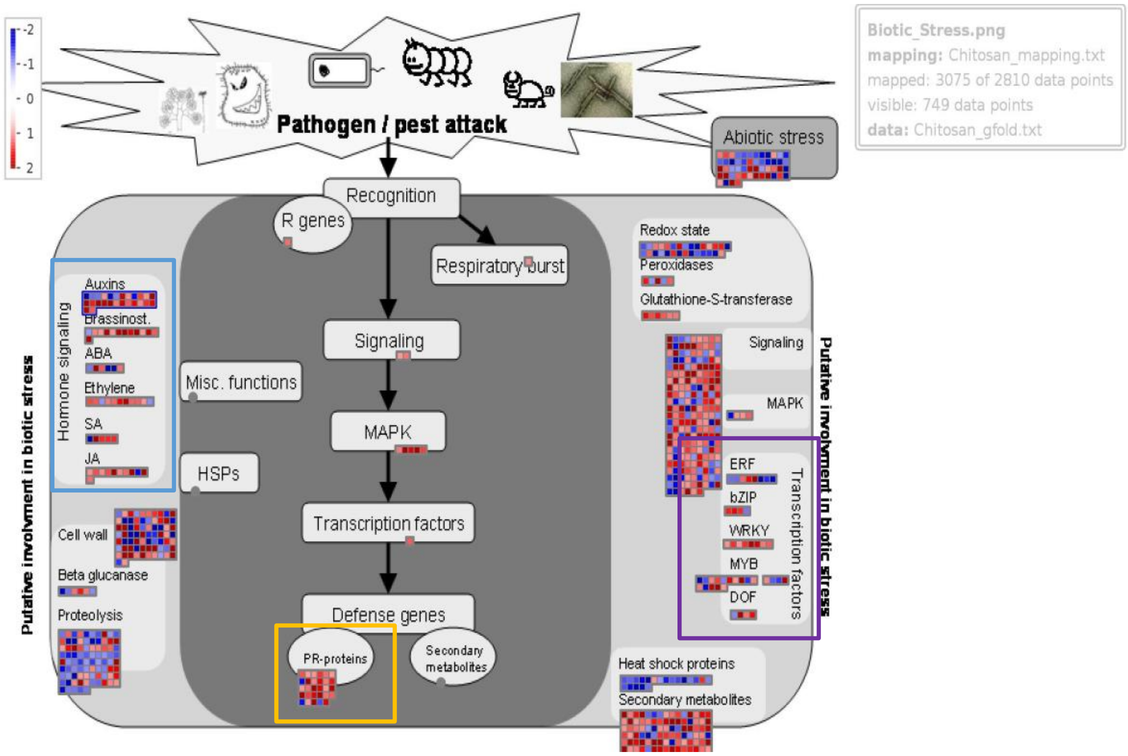


Figure 42: oligo-chitosan-regulated genes involved in overall stress responses.

6.9.4 Comparison of treatments' regulation of ribosomal protein biosynthesis.

The above results suggested a conversely large number of down-regulated genes involved in ribosomal related processes after ANE, oligo-chitin and oligo-chitosan treatments should be expected. ANE, oligo-chitin, oligo-chitosan and oligo-alginate regulation of ribosomal protein coding genes are detailed in Figures 43 through 46. As shown in Figure 43, the ribosomal proteins in plants are scattered throughout the nucleus, mitochondria and chloroplast. ANE, oligo-chitin and oligo-chitosan clearly down-regulated a large number of ribosomal protein coding genes as shown in Figures 43, 45 and 46, respectively, whereas, oligo-alginate treatment did not have a significant effect (see Figure 44).

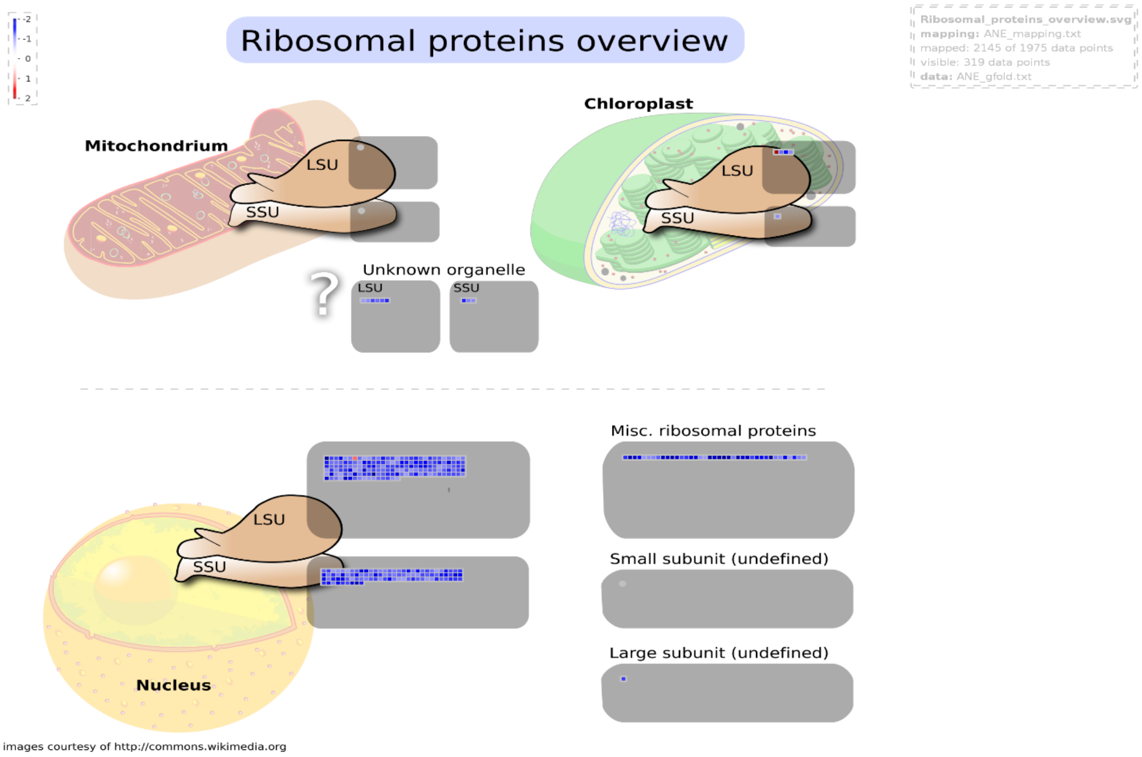


Figure 43: ANE- regulated ribosomal protein coding genes.

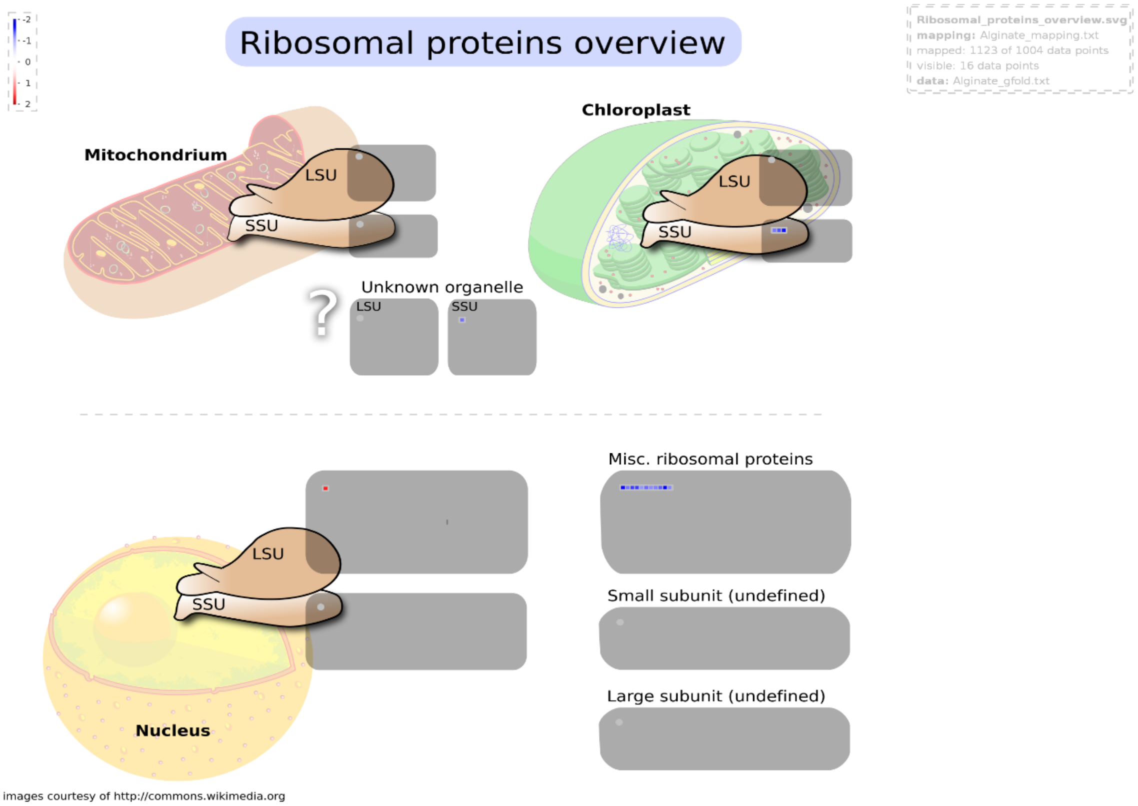


Figure 44: oligo-alginate-regulated ribosomal protein coding genes.

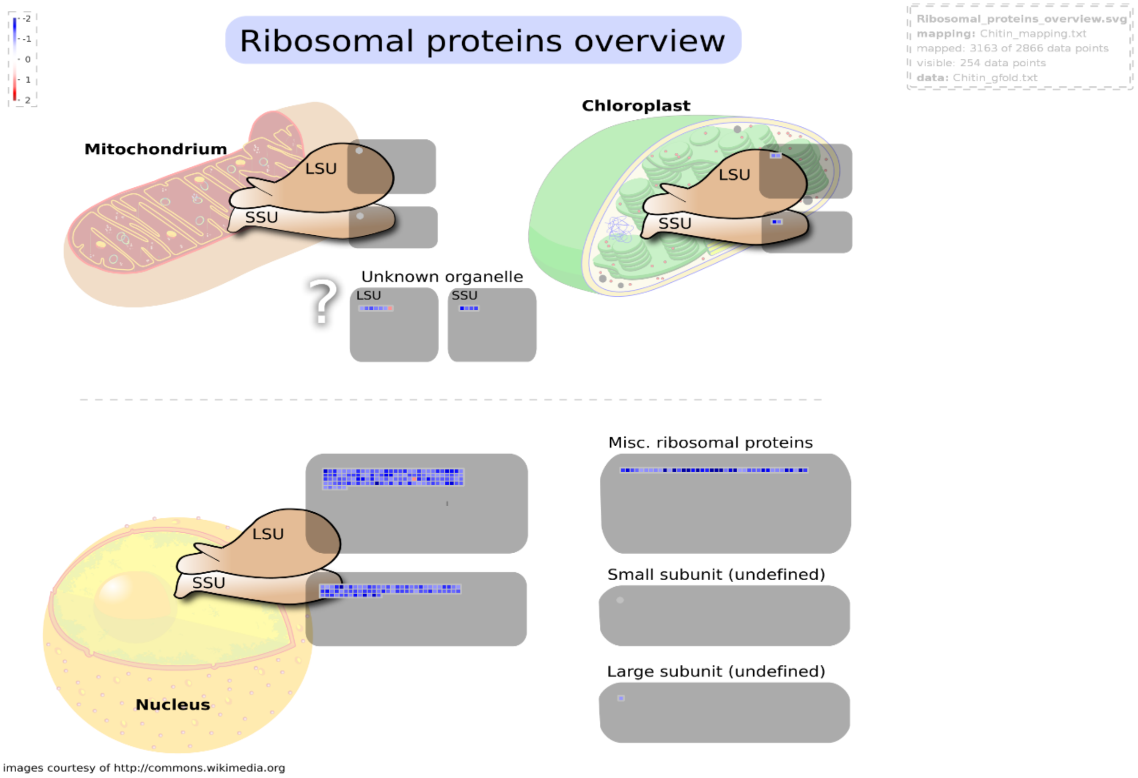
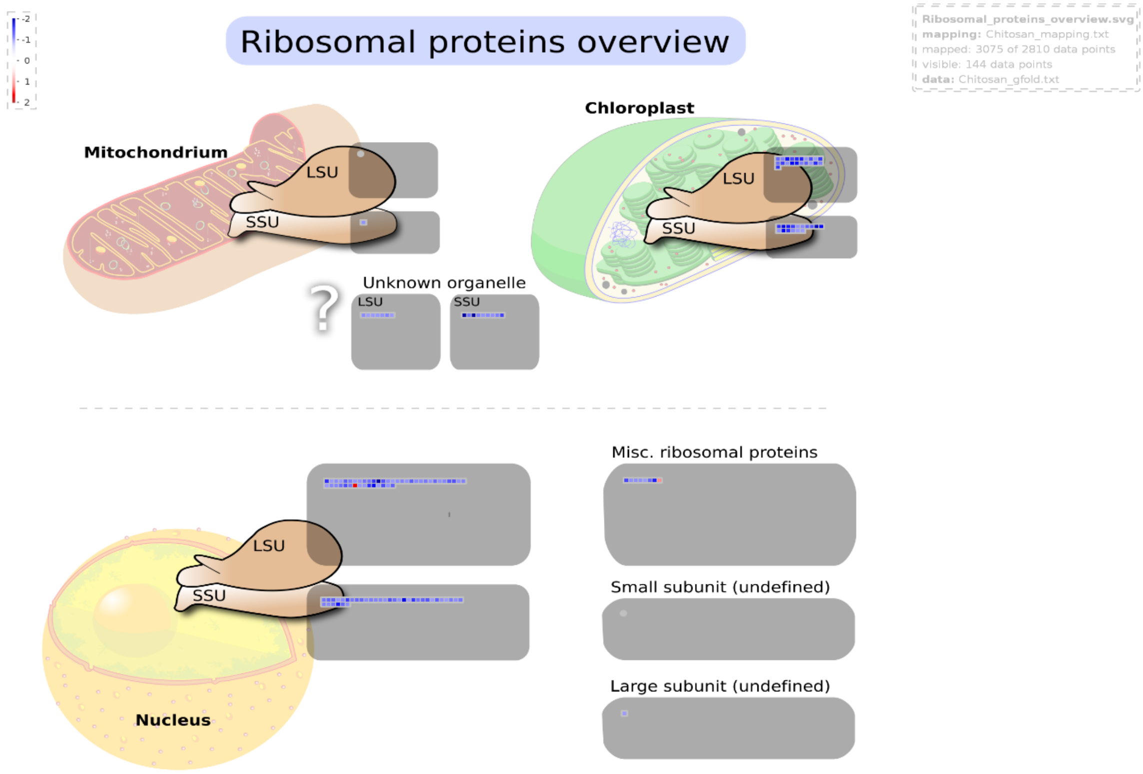


Figure 45: oligo-chitin-regulated ribosomal protein coding genes.



images courtesy of http://commons.wikimedia.org

Figure 46: oligo-chitosan-regulated ribosomal protein coding genes.

6.9.5 Comparison of treatments’ regulation of lipid biosynthesis.

Lipid biosynthesis in plants begin with the pyruvate; various enzymatic reactions result in lipid biosynthesis, as outlined in Figures 47 through 50. In the present study, treatments of ANE, oligo-chitin and oligo-chitosan were observed to inhibit the various steps of lipid synthesis, as shown in Figures 47, 49 and 50, respectively. Treatments with oligo-alginate did not significantly regulate this process (see Figure 48).

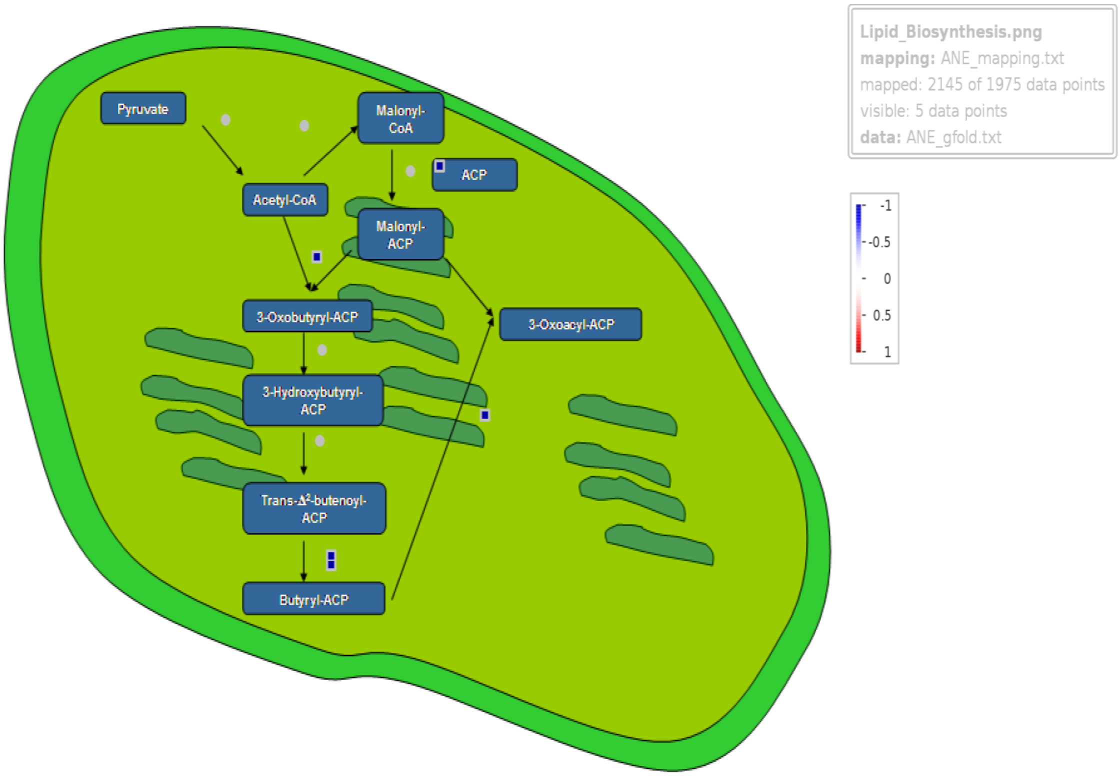


Figure 47: ANE-regulated lipid biosynthesis coding genes.

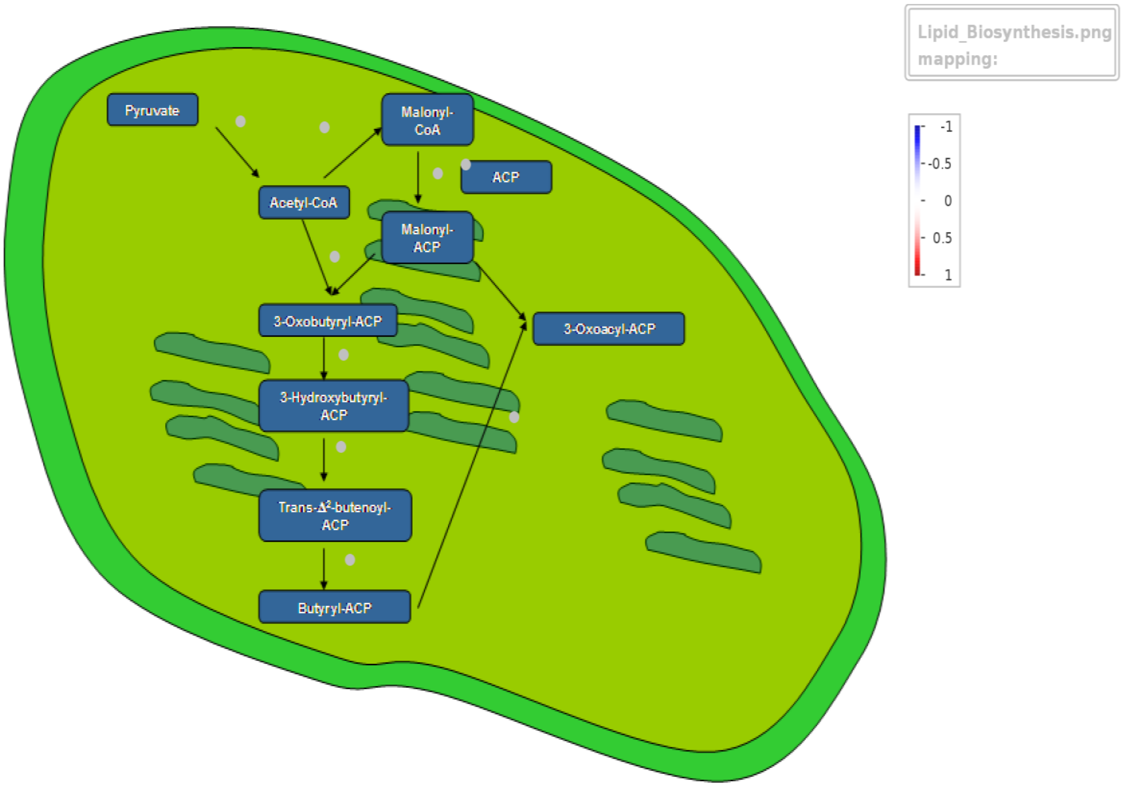


Figure 48: oligo-alginate-regulated lipid biosynthesis coding genes.

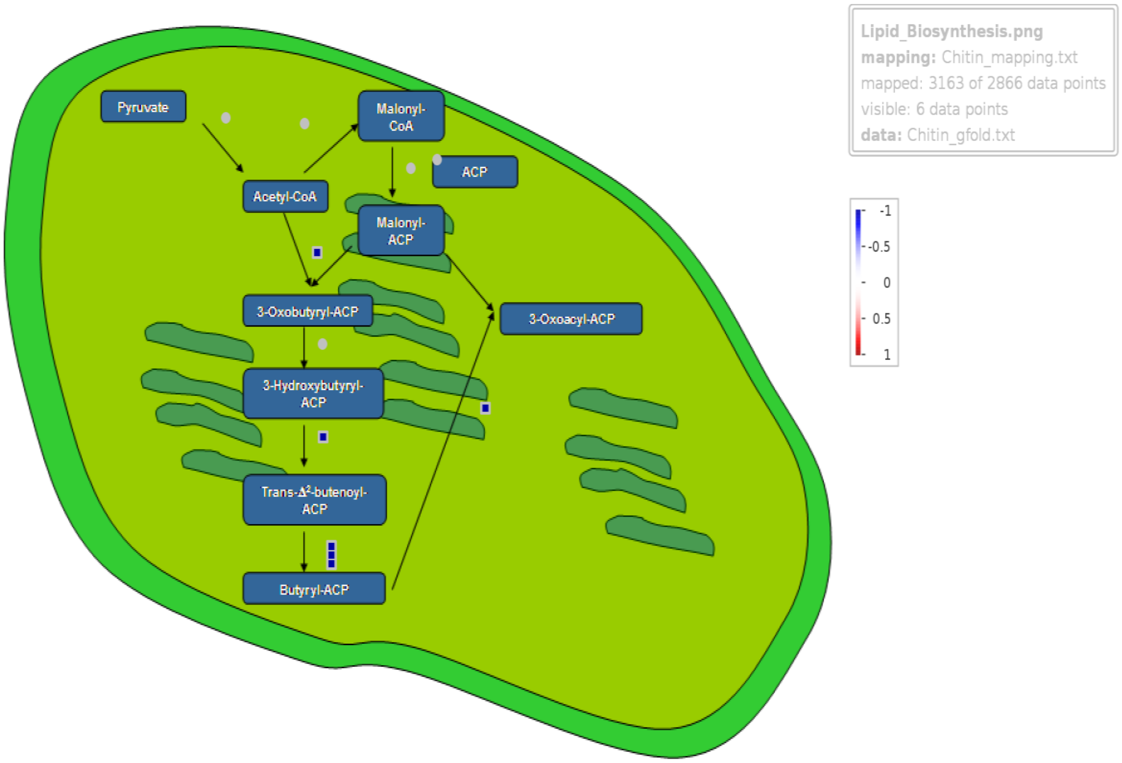


Figure 49: oligo-chitin-regulated lipid biosynthesis coding genes.

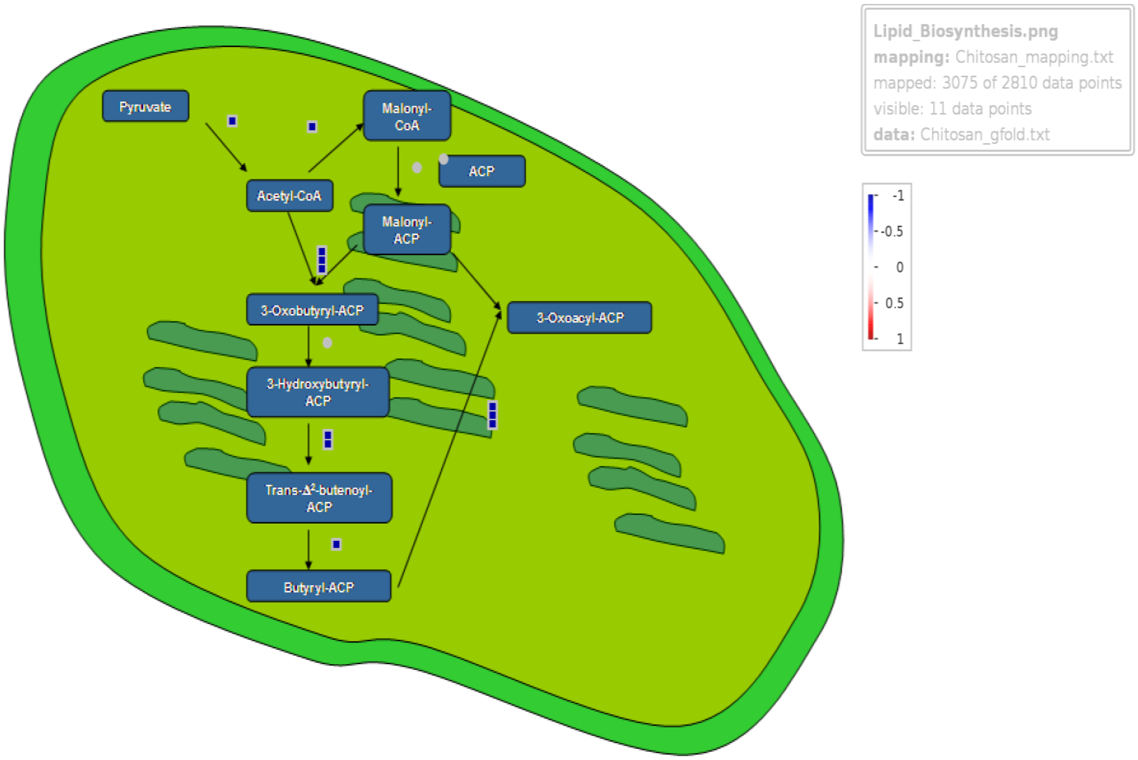


Figure 50: oligo-chitosan-regulated lipid biosynthesis coding genes.

6.9.6 Comparison of treatments’ regulation of nucleotide biosynthesis.

Similar to the observations of regulation of the ribosomal protein and lipid biosynthesis coding genes, various nucleotide synthetic processes were also seen to be modulated in this study. Figures 51 through 54 outline the steps of plant nucleotide synthesis. Each step involves an enzymatic reaction. ANE, oligo-chitin and oligo-chitosan down-regulated expression of a large number of genes involved nucleotide biosynthesis (see Figures 51, 53 and 54 respectively). Again, oligo-alginate treatment did not affect the expression of genes related to nucleotide biosynthesis (see Figure 52).

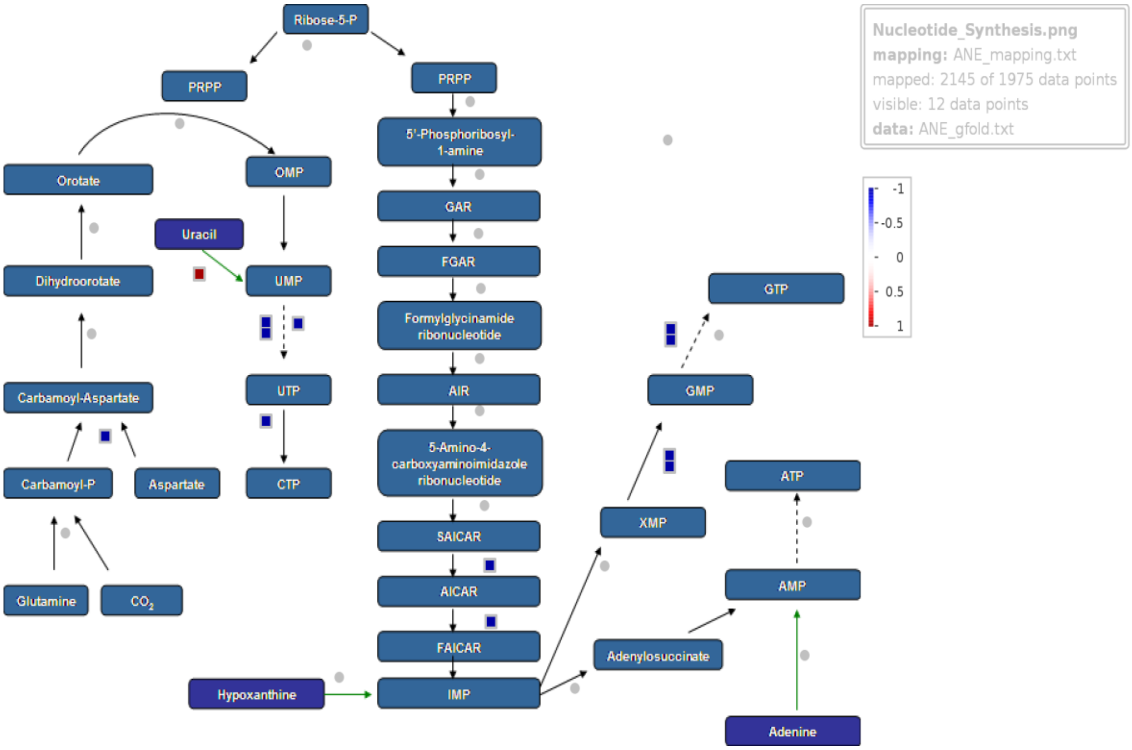


Figure 51: ANE-regulated nucleotide biosynthesis coding genes.

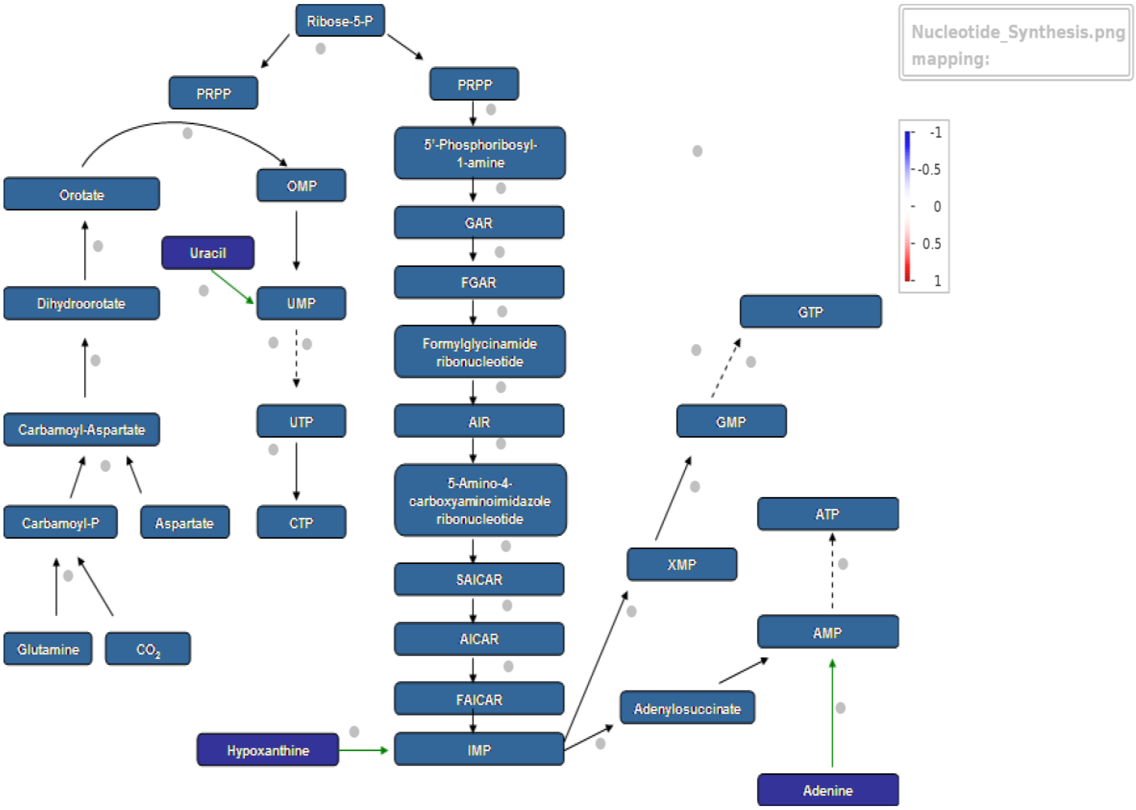


Figure 52: oligo-alginate-regulated nucleotide biosynthesis coding genes.

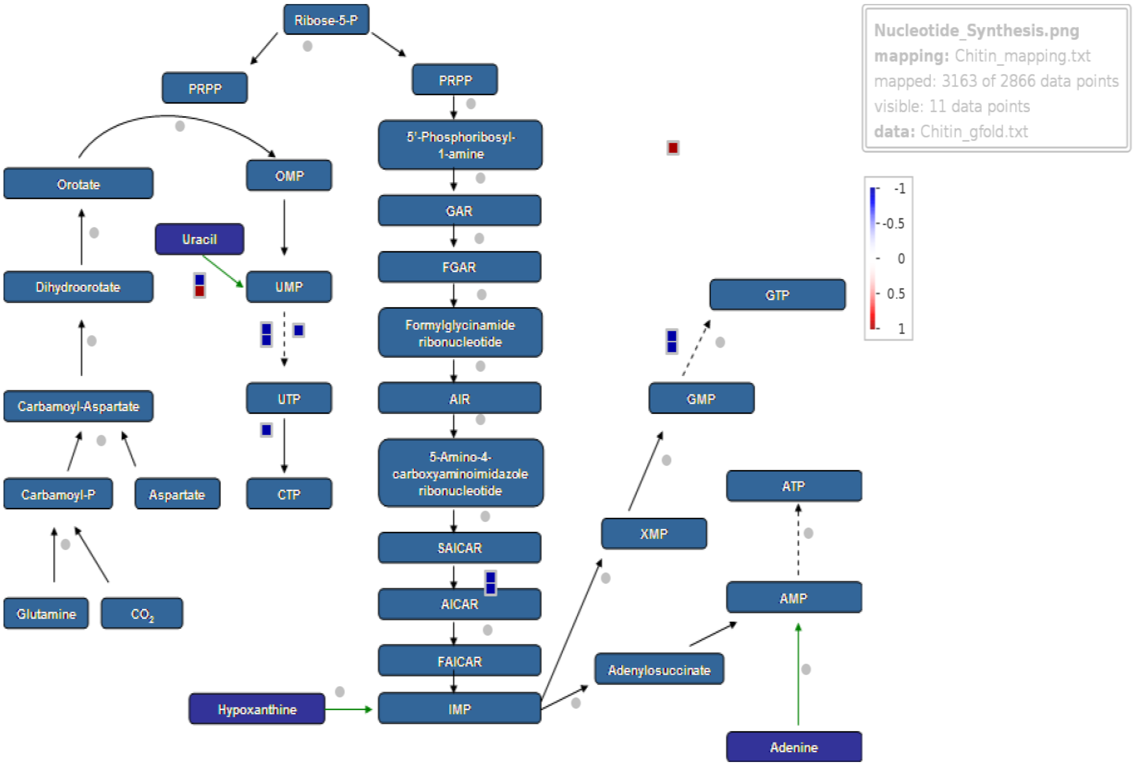


Figure 53: oligo-chitin-regulated nucleotide biosynthesis coding genes.

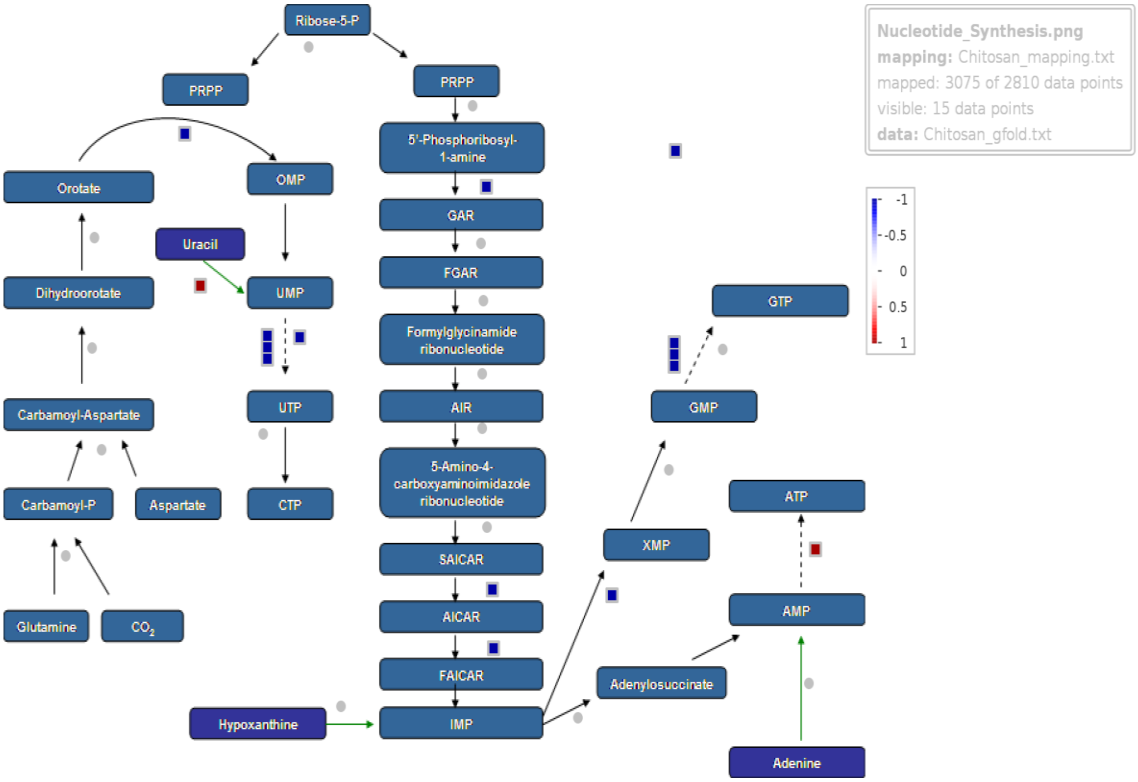
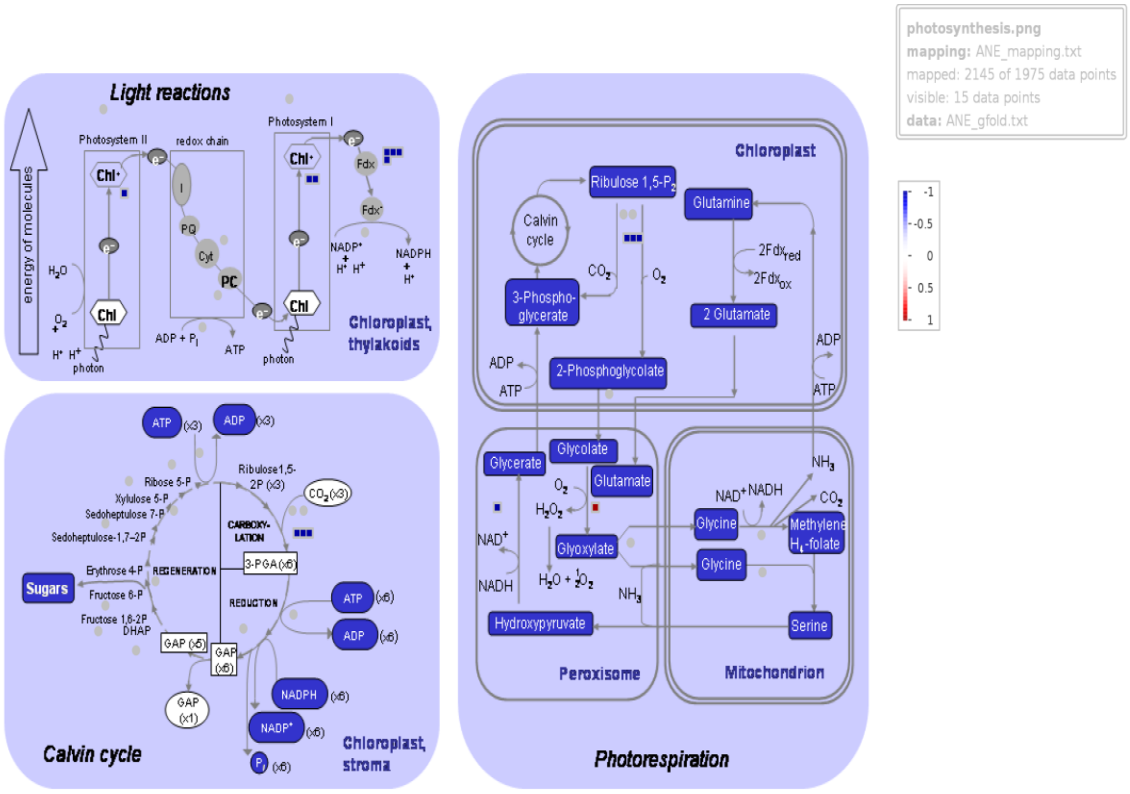


Figure 54: oligo-chitosan-regulated nucleotide biosynthesis coding genes.

6.9.7 Comparison of treatments' regulation of photosynthesis.

Photosynthesis is the ultimate energy source for plants. Photosynthesis was also found to be regulated by ANE and oligosaccharides treatments, as illustrated in Figures 55 through 58. It was observed that all treatments inhibited various steps of the light reactions, although oligo-chitin treatment did up-regulate expression of a number of genes involved in the electron transport chain. In the Calvin cycle, ANE, oligo-chitin and oligo-chitosan treatments down-regulated a larger number of genes than did oligo-alginate treatment (compare Figures 55, 57 and 58 with Figure 56).



photosynthesis.png
mapping: ANE_mapping.txt
mapped: 2145 of 1975 data points
visible: 15 data points
data: ANE_gfold.txt

Figure 55: ANE-regulated genes involved in photosynthesis.

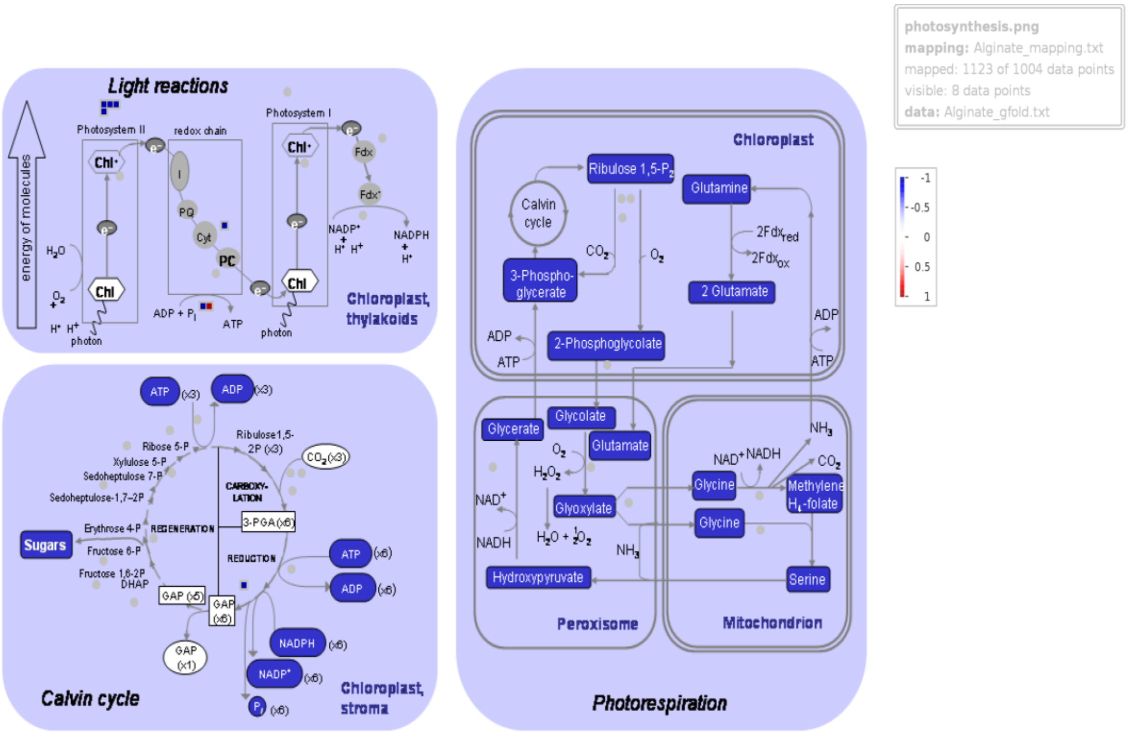


Figure 56: oligo-alginate-regulated genes involved in photosynthesis.

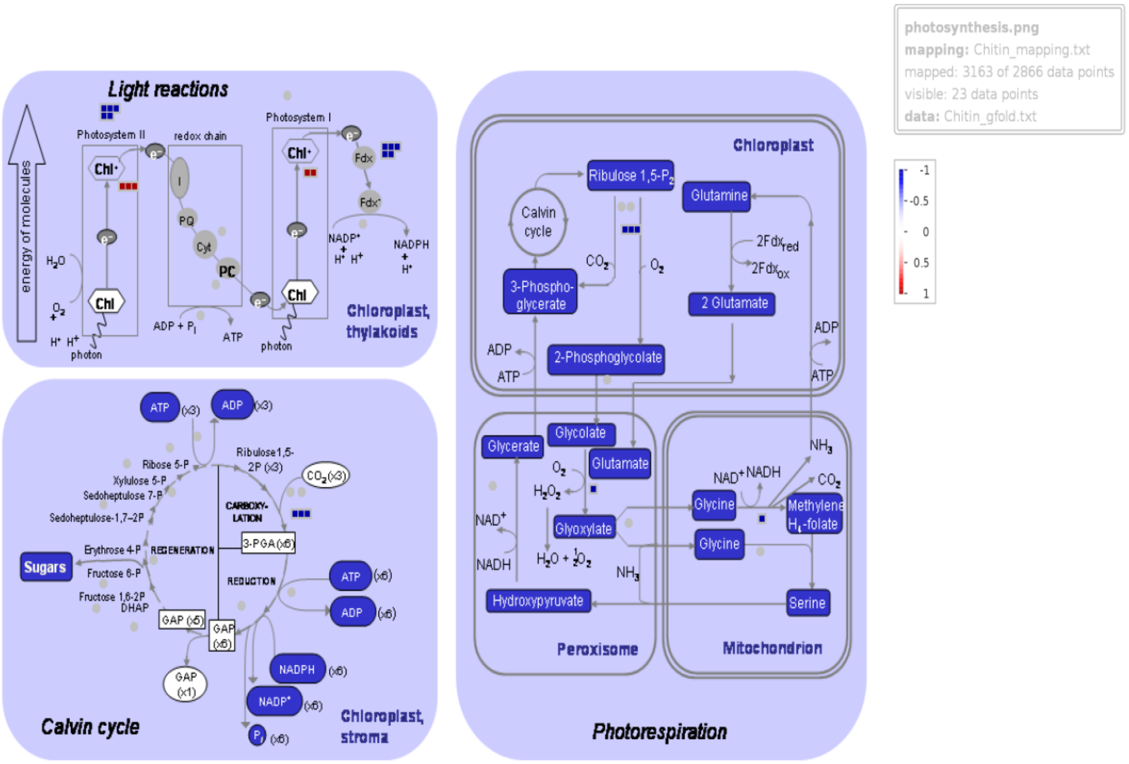


Figure 57: oligo-chitin-regulated genes involved in photosynthesis.

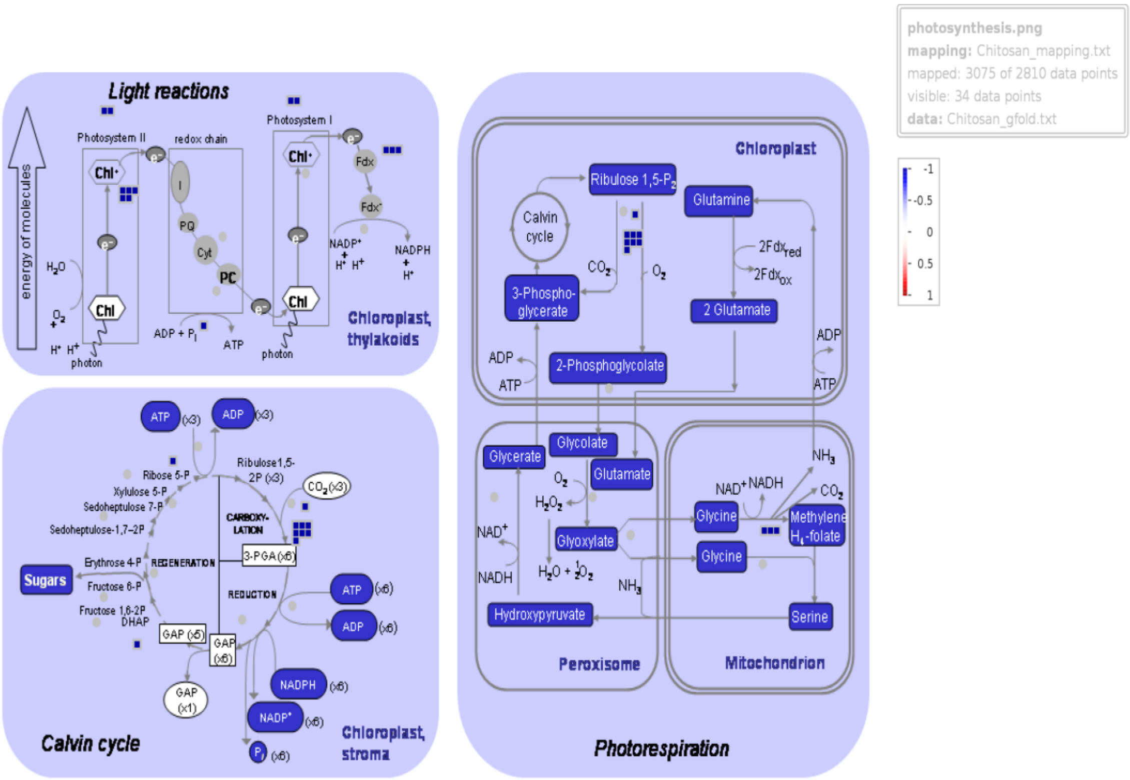


Figure 58: oligo-chitosan-regulated genes involved in photosynthesis.

6.9.8 Comparison of treatments’ regulation of chlorophyll biosynthesis.

Chlorophyll content is an important component in photosynthesis. The biosynthesis of chlorophyll in plants is carried out by a cascade of enzymatic reactions (see Figures 59 through 62). The expression of genes coding for these enzymes was modulated by ANE and oligosaccharide treatments. Similar to the inhibition of protein, lipid and nucleotide biosynthesis, treatments of ANE, oligo-chitin and oligo-chitosan also inhibited chlorophyll biosynthesis, as indicated in Figures 59, 61 and 62, respectively.

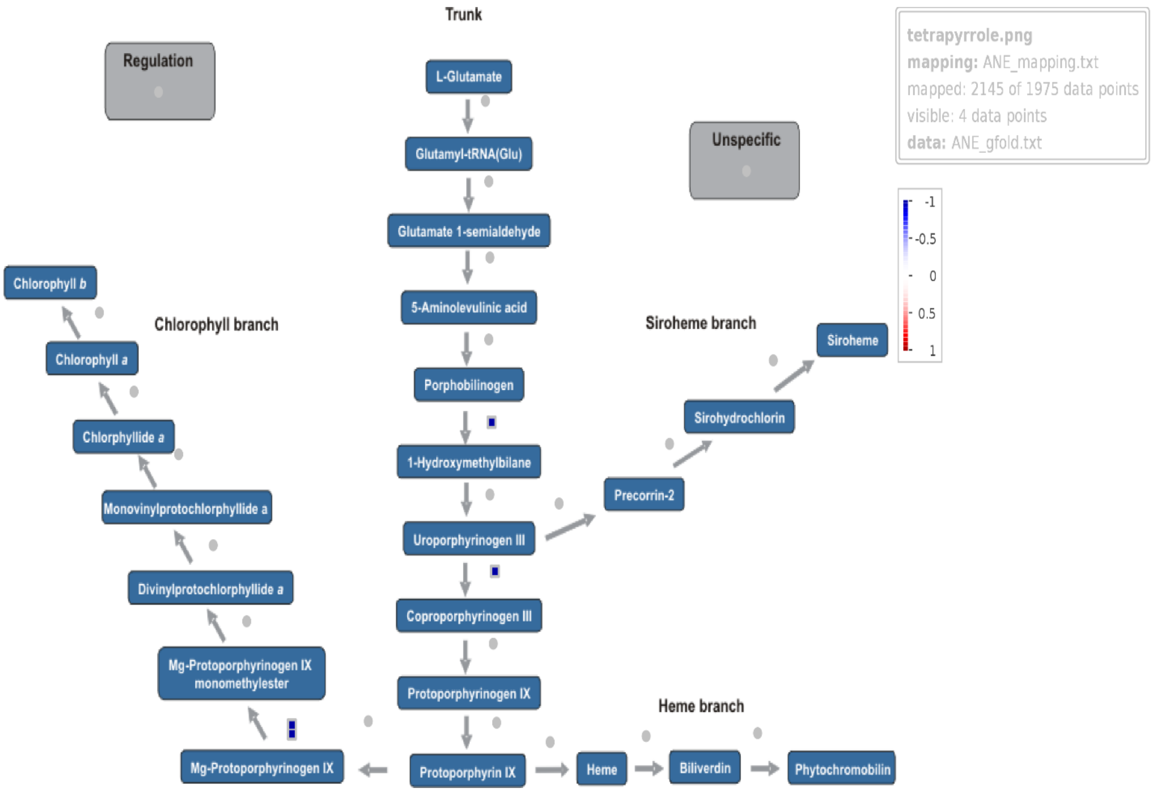


Figure 59: ANE-regulated genes involved in chlorophyll biosynthesis.

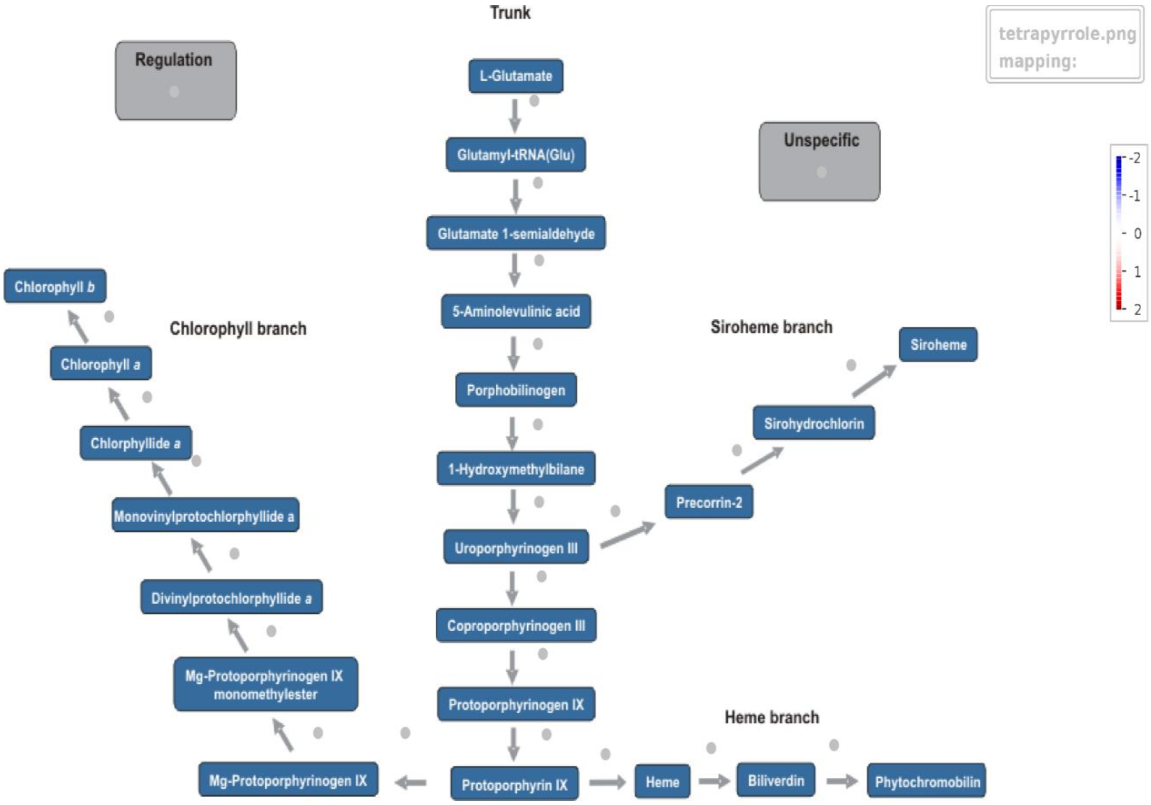


Figure 60: oligo-alginate-regulated genes involved in chlorophyll biosynthesis.

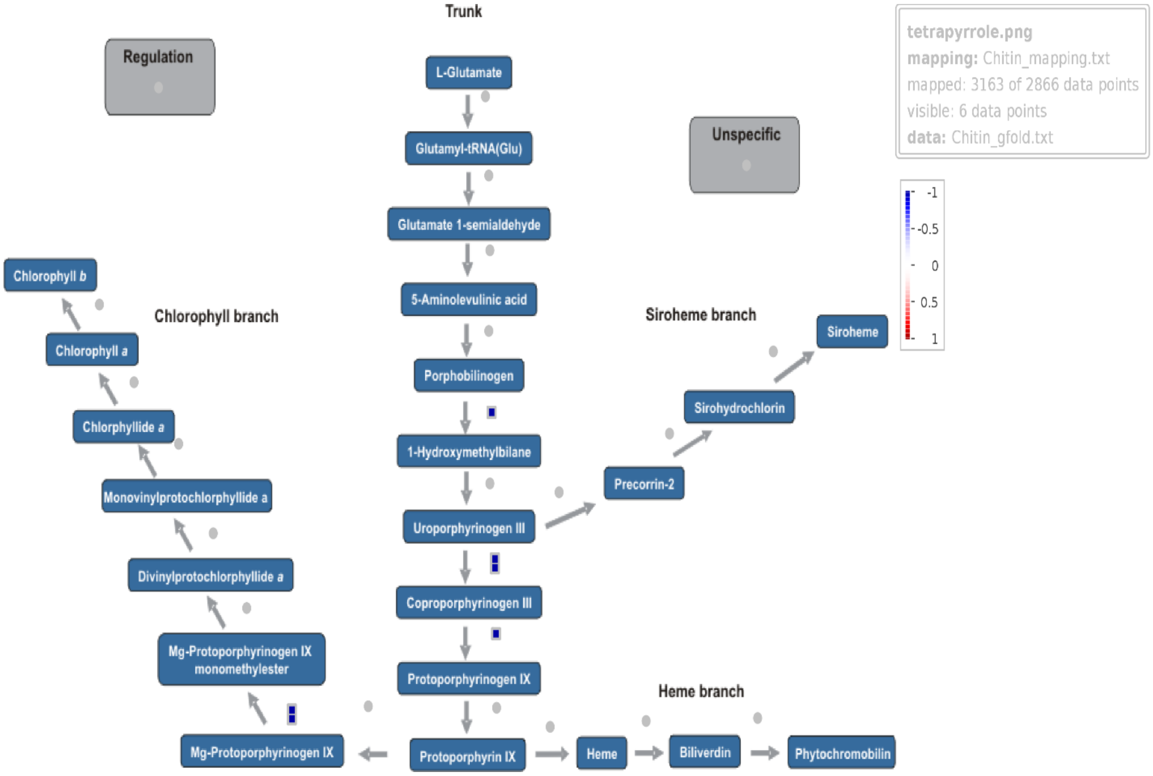


Figure 61: oligo-chitin-regulated genes involved in chlorophyll biosynthesis.

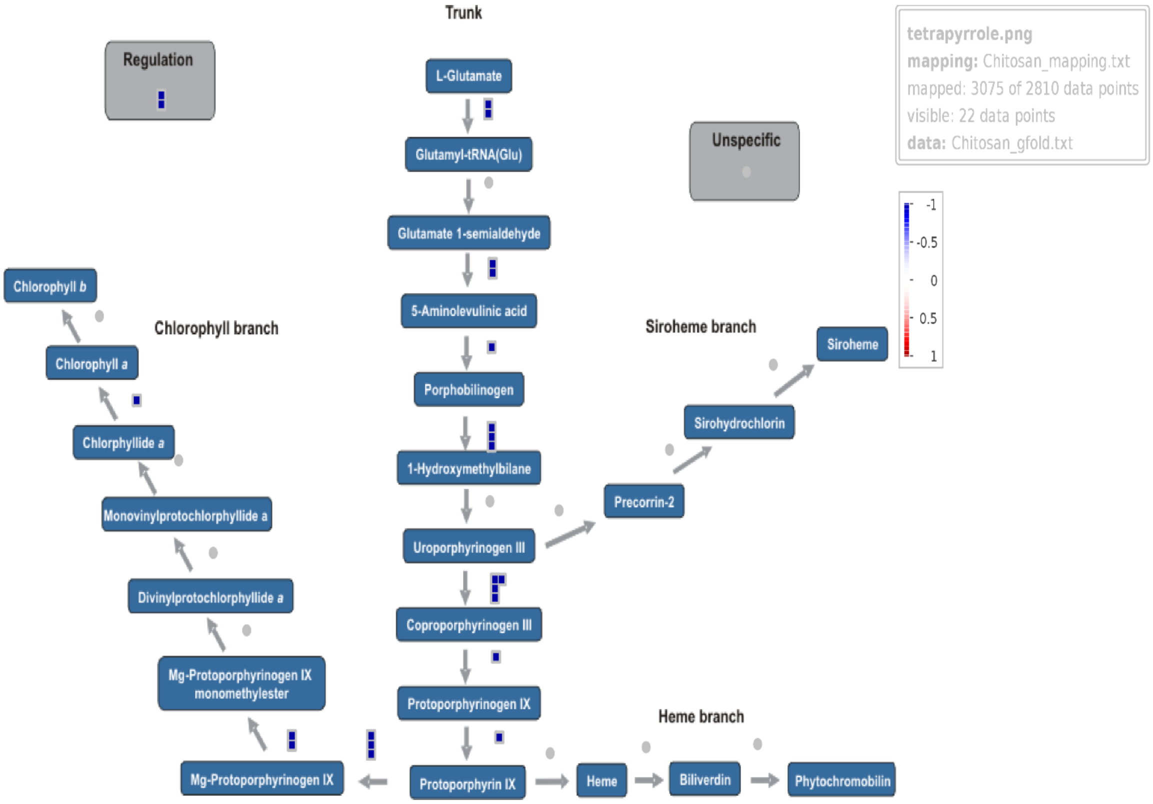


Figure 62: oligo-chitosan-regulated genes involved in chlorophyll biosynthesis.

7 Discussion

Various seaweed extracts, including ANE, have the potential to increase crop productivity. Recent reports have focused on the various beneficial effects of applications of ANE on a variety of plant systems. To efficiently use the abundant seaweed resources, it is essential to understand the bioactive components in ANE and its corresponding mechanism of action. Since ANE contains inconsistent amounts of micronutrients and very low concentrations of phytohormones, the underlying action of ANE needs to be reassessed. As ANE can be applied to large areas in very small amounts and still promote plant growth, the bioactive components most likely are derived from ANE's major components. As *Ascophyllum nodosum* is very closely associated with its symbiotic partner, the fungus *Mycophycias ascophylli*, constituents from both organisms need to be considered. The alkaline and high-pressure ANE extraction process hydrolyzes the polysaccharides of *Ascophyllum nodosum* and *Mycophycias ascophylli* into various oligosaccharides. Alginate from *Ascophyllum nodosum* becomes oligo-alginate and chitin from *Mycophycias ascophylli* becomes oligo-chitin and oligo-chitosan.

By applying these related oligosaccharides to soybean seedlings, marked changes in soybean gene expression were observed by analysis of transcriptomic data.

7.1 Quality of the data

The application of next generation sequencing (NGS) technologies are now greatly facilitating advancements in biology. NGS typically generate a large amount of data, thus quality controls are essential for obtaining accurate information. mRNA sequencing often suffers high sequencing error rates and low base quality, all depending on the quality of sequencing machines, reagent availability, and the samples. As stated above, error rate increases as the sequencing reads are extended and sequencing reagents become scarce and the first six bases have a particularly high error rate due to the random hexamers used in priming cDNA synthesis. In the present study, sequencing error rates averaged below 0.1%, ensuring the reliability of the data. More than 90 % of the bases used in this mRNA sequencing procedure possess the quality of Q30, which reduced the error in the base calling processes. After filtering out the low quality data, the clean data were then mapped onto the soybean genome. For the mRNA sequencing, exon-mapped reads should be the most abundant type of read when the reference genome is well annotated. Intron-reads may be derived from pre-mRNA contamination or intron retention events from alternative splicing. Reads map to intergenic regions mainly because of weak annotation of the reference genome. The statistics of the mapping results further ensured that most of the clean data were actually mapped to the exons of the soybean genome.

7.2 Differentially expressed gene determination

Determination of the differentially expressed genes were vital in this study, since all the bioinformatics analysis was based on the differentially expressed genes and all conclusions were drawn from it as well. Replication is very important in biological studies to account for variations and mRNA sequencing in this case is no exception. However, the costs of mRNA sequencing are currently quite high. To compensate for the lower number replications in this study, and to determine the differentially expressed genes between ANE, oligosaccharides treatments and water treatment, Bayesian approach was taken. So, instead of considering whether the differentially expressed genes were significant or not between two conditions, posterior probability of the fold changes were taken into account and generalised fold changes were calculated for determining the differentially expressed genes.

All the differentially expressed genes determined in this study have a 99% probability that the absolute fold change (RPKM in sample 2 /RPKM in sample 1) is greater than 2.

7.3 The effects of oligo-chitin and oligo-chitosan are similar to that of ANE

7.3.1 Similarity of different treatments based on the gene expression pattern and number of commonly regulated genes

The results of gene expression pattern analysis suggested that in the context of regulated gene sets (All treatment-regulated genes, ANE treatment-regulated genes, ANE treatment-up-regulated genes and ANE-treatment-down regulated genes), oligo-chitin and oligo-chitosan treatments showed a larger overlap with ANE treatments than did oligo-alginate treatments. This indicated that oligo-chitin and oligo-chitosan can potentially mimic the effects of ANE on plants.

It is worth noting that the ANE in this study was applied at 3ml / L and oligo-chitin, oligo-chitosan and oligo-alginate were applied at 30, 50 and 40 ppm, respectively. Changes in application levels of these treatments may result in slight variation in gene expression patterns. In terms of the number of genes regulated by different treatments, oligo-chitin and oligo-chitosan both regulated the expression of approximately 3000 genes, whereas ANE regulated approximately 2000 genes and oligo-alginate only modulated about 1000 genes. These marked differences in the number of genes regulated by different treatments suggest that oligo-chitin and oligo-chitosan may be the most significant bioactive component within ANE. It is also possible that oligo-chitin and/or oligo-chitosan in ANE solution were slightly diluted. Furthermore, since there were a larger number of regulated genes after oligo-chitin and oligo-chitosan treatments, it is likely that ANE regulated genes would largely overlap with those observed with oligo-chitin and chitosan treatments, which could make oligo-chitin and oligo-chitosan treatments look more similar to that of ANE when focusing on ANE-regulated genes.

Based on results of the Venn diagram, it is concluded that oligo-chitin and oligo-chitosan regulated a larger number of genes than ANE. The genes commonly up- or down-regulated by all three treatments are indicative of an overlapping of effects within the plants. Genes exclusively regulated by ANE can then account for the effects on plants unique to ANE itself.

7.3.2 Similarity of different treatments based on the functions of commonly regulated genes

7.3.2.1 Similarity of different treatments based on the functions of commonly up-regulated genes

As the results of the heat map and Venn diagram indicate, among the ANE-up regulated genes, a large number of these were also up-regulated by treatments with oligo-chitin and oligo-chitosan. The functions of these commonly up-regulated genes in part reflect the common action of ANE and oligosaccharides treatments. Gene function annotation revealed that all treatments up-regulated the expression of genes involved in terpenoid and flavonoid biosynthesis, substances typically associated with plant stress and defense responses (Fan et al., 2011; Lola-Luz et al., 2014). The stress and defense responses frequently involve the generation of various reactive oxygen species (ROS), which on the one hand could directly inhibit pathogen proliferation, yet on the other hand can be quite damaging to normal cell functions. Not surprisingly, all treatments up-regulated the expression of a number of genes that encode ROS scavenger proteins, which agrees with recent studies showing that oligo-alginate treatment increased SOD and POD generation in *Triticum aestivum* (Liu et al., 2013) and oligo-chitosan treatment increased POD generation in plants (Falcón-Rodríguez, Cabrera, Ortega & Martínez-Téllez, 2009). Chitin is an indispensable structural component of fungal cell wall. For plants, application of oligo-chitin is similar to attack from a pathogenic fungus and, so it is understandable that oligo-chitin is able to induce various plant defense responses as mentioned above. Alginate, on the other hand, is a primary constituent of brown seaweeds' cell walls. Intriguingly, land plants also perceived oligo-alginate as a danger signal as it, too, seemed to induce defense responses on the part of the treated soybean seedlings. The large number of commonly regulated genes in ANE, oligo-chitin and oligo-chitosan treatments were mainly involved in various stress responses, stress-related transcription factors and catabolic (energy producing) processes, as shown in Figure 21. Transcription factors (TFs) are vital regulatory factors in gene expression alteration. By binding to a consensus sequence motif in the regulatory region of the target genes, TFs can directly modulate the expression levels of those genes. As an important storage resource in plants, starch was often transported into roots when plants were confronted with various stresses (McKibbin et al., 2006). Since ANE, oligo-chitin and oligo-chitosan treatments generally induced plant stresses responses, it is not surprising that starch biosynthesis genes were up regulated in these treatments. Stress-related plant hormones, like abscisic acid and jasmonic acid, and their corresponding signaling pathways, were also generally up-regulated by application of ANE, oligo-chitin and oligo-chitosan. Auxin is a well known plant growth promoting hormone, involved in rapid cellular expansion, whereas, various AUX/IAA proteins act as repressors in auxin transcriptional-regulated signaling pathways. The increased expression level of IAA coding genes after ANE, oligo-chitin and oligo-chitosan treatments may, inhibit auxin’s action (Weijers & Friml, 2009). Recently, the protein Topless was found to be a corepressor of AUX/IAA in the auxin inhibitory pathway (Szemenyei, Hannon & Long, 2008). Interestingly, ANE treatment up-regulated this gene as did oligo-chitin and oligo-chitosan treatments, to a lesser extent (see Table 14 in appendix 1 for detailed information). Therefore, the up-regulation of various IAA and Topless genes following ANE, oligo-chitin, and oligo-chitosan treatments may have inhibited cell expansion and general plant growth. Decreased cell wall expansion may allow plants to save energy and at the same time reduce the possibility of pathogen invasion during cell expansion.

7.3.2.2 Similarity of different treatments based on the functions of commonly down-regulated genes

Approximately 1500 genes were down regulated after ANE treatment; not surprisingly, these genes were involved in broader biological processes. Most strikingly, a large number of ANE down-regulated genes are involved in various energy-consuming processes. Specifically, 315 ANE down-regulated genes were either involved in ribosomal protein biosynthesis, ribosomal biogenesis, ribosomal assembly, or translation regulatory processes, such as translation initiation, elongation, and termination as shown in Figure 22 (see Table 16 in Appendix 2 for detailed information). Surprisingly, the majority of these genes coding for protein synthetic machinery components were similarly down-regulated in oligo-chitin- and oligo-chitosan-treated plants. Whereas, oligo-alginate treatment appeared to exhibit no inhibitory effects. rRNA processess important steps in protein synthesis, were also inhibited by ANE, oligo-chitin and oligo-chitosan treatments, as shown in Table 17 (Appendix 2). Only 4 out of 68 related genes were down-regulated in oligo-alginate-treated plants. Amino acid biosynthesis is vital for ongoing protein biosynthesis; Nitrate reductase is involved in the assimilation of N into amino acids, which was down-regulated by treatments of ANE and oligo-chitin as shown in Table 18 (Appendix 2). In addition, various amino acid biosynthesis and transportation related genes were also down-regulated following treatments of ANE, oligo-chitin and oligo-chitosan. Also, a large number of genes encoding for elongation factors were coincidently down-regulated in ANE, oligo-chitin and oligo-chitosan-treated plants, as shown in Table 20 and 21 (Appendix 2), but not in oligo-alginate treated plants. These results strongly suggest that, treatments of ANE, oligo-chitin, and oligo-chitosan inhibit overall protein synthetic processes, which has been suggested to be quite energetically expensive (Edwards, Roberts & Atwell, 2012). Not surprisingly, the various genes responsible for protein folding and protein transportation were also down-regulated in ANE-, oligo-chitin- and oligo-chitosan-treated plants, as shown in Figure 22 (see Table 22 in Appendix 2 for detailed information).

Besides inhibiting general protein synthesis and transportation, ANE, oligo-chitin, and oligo-chitosan treatments similarly down-regulated the expression of a large number of genes involved in nucleotide and DNA synthesis, as shown in Figure 22 (see Table 23 in Appendix 2 for detailed information). Another energy consuming process is fatty acid synthesis. As shown in Figure 22, down-regulated genes involved in fatty acid biosynthesis by ANE treatment were also largely down-regulated in treatments of oligo-chitin and oligo-chitosan but not in oligo-alginate (see Table 24 in Appendix 2 for detailed information). Perhaps, to compensate for energy loss, protein degradation processes were also modulated. As shown in Figure 22, trypsin inhibitor-related genes that typically inhibit the protease trypsin, were largely down-regulated in ANE-, oligo-chitin- and oligo-chitosan-treated plants (see Table 26 in Appendix 2 for detailed information).

Treatments of ANE, oligo-chitin and oligo-chitosan commonly down-regulated various energy consuming processes such as ribosomal protein synthesis, ribosomal biogenesis, and various related RNA processes, different levels of translational processes, protein folding, transport processes and fatty acid biosynthesis. All these inhibitory effects likely resulted in decreased cell division as shown in Figure 22 (see Table 28 in Appendix 2 for detailed information). Interestingly, some of these down regulated genes related to cell division were also associated with various stress responses, which suggests the eliciting effects of ANE, oligo-chitin and oligo-chitosan were perceived by plants as stress signals and resulted in a consequent decrease in cell division.

Aside from the inhibitory effects of ANE, oligo-chitin and oligo-chitosan treatments on plant cell proliferation, the treatments also played an important role in regulation of cell expansion as shown in Figure 22 (see Table 30 in Appendix 2 for detailed information). The expression levels of 103 genes related to cell wall modification and reorganization were significantly modulated. For example, various families of expansin, xyloglucan-endo-transferase and polygalacturonase encoding genes were coordinately down-regulated by ANE, oligo-chitin and, to lesser extent, oligo-chitosan, which could potentially decrease cell expansion. There are two likely reasons for this inhibited cell expansion. Firstly, cell expansion largely relies on the actions of various enzymes, the synthesis of which is energy expensive, especially under conditions of limited efficiency of protein synthesis. Secondly, it is possible that cell expansion offers opportunities to pathogenic invaders, for instance it has been reported that nematodes have the ability to synthesize expansin, which can lossen plant cell walls. (Qin et al., 2004). Although reports are rare concerning the existence of these cell wall loosening enzymes in other plant pathogens, it is very possible that during co-evolution, some pathogens may have evolved the ability to invade the plant host either by manipulation of the synthesis of a plant cell wall loosening enzyme or directly synthesis of such an enzyme itself. For example, many microbes can manipulate plant auxin production and so increase odds of successfully invading the plant host (Kidd et al., 2011). One study has revealed that auxin binding protein, ABP1, is involved in auxin-related non-transcriptional control of cell expansion. By binding auxin, the binding protein can rapidly increase the acidity of the cell membrane, thus activating cell wall loosening enzymes like expansin and xyloglucan-endotransferase to facilitate turgor-driven cell expansion (Perrot-Rechenmann, 2010). Interestingly, as shown in Table 44 (Appendix 2), auxin binding protein encoding genes, were commonly down-regulated by ANE, oligo-chitin, and oligo-chitosan treatment. Together with up-regulated genes coding for auxin-induced proteins and the protein Topless (see Table 14 in Appendix 1 for detailed information), it seems that treatments of ANE, oligo-chitin and oligo-chitosan all decreased cell wall expansion by both decreasing the expression of cell wall loosening enzymes and down-regulating the auxin transcriptional and non-transcriptional related pathways. This inhibition of cell wall expansion may function to decrease the possibility of pathogen invasion.

Stressful conditions were often correlated with energy loss as shown in this study. It is important to understand how this energy deprivation occurred. ANE-, oligo-chitin- and oligo-chitosan-treated plants all commonly showed impaired photosynthesis, as treatments down-regulated several photosynthesis related genes as shown in Figure 22 (for detailed information see Table 38 in Appendix 2). For example, RuBisCO-related genes were significantly down-regulated in ANE, oligo-chitin and oligo-chitosan treatments, whereas it was up-regulated following oligo-alginate treatment. Further to this, various chloroplast organization-related genes and chlorophyll biosynthetic encoding genes were all commonly down-regulated in ANE-, oligo-chitin- and oligo-chitosan- treated plants.

7.3.3 Similarities between treatments' effects on biological processes.

ANE and oligosaccharide treatments all generally up-regulated synthesis of plant secondary metabolites. Enhanced secondary metabolite synthesis is essential for plants to successfully cope with stress. For example, synthesis of sesquiterpenoids, triterpenoids and diterpenoids is up-regulated during pathogen attacks (Tholl, 2015). Up-regulation of flavonoid synthesis observed in the present study is in agreement with previous reports of increases in flavonoid content following ANE treatment (Fan et al., 2011; Lola-Luz et al., 2014). ANE, oligo-chitin and oligo-chitosan all showed a tendency to down-regulate genes involved with ribosomal processes, leading to decreased protein synthesis, as well as inhibiting nucleotide and fatty acid synthesis. These activities are crucial to plant growth yet, simultaneously, they consume much of a plant's energy. The only treatment that did not inhibit these energy-consuming processes was oligo-alginate.

7.4 Possible mechanism for effects of ANE, oligo-chitin and oligo-chitosan treatments

Results indicating inhibition of plant growth responses following ANE, oligo-chitin and oligo-chitosan treatments may seem counterintuitive to reports claiming that ANE promotes plant growth. One issue may lie in the application rate of ANE in this study as compared to others. Even within our own lab, different application rates of ANE have led to results differing from the ones in the present study. Another reason may be the use of different plant species or the same species but at differing developmental stages. As a mixture of elicitors, varying concentrations of ANE may mean a different level of plant signal, which in turn induces different levels of signal transduction, consequently eliciting altered plant responses. Plant cell membranes possess various receptors; one for oligo-chitin has been determined (Kaku et al., 2006; Miya et al., 2007; Shibuya, Kaku, Kuchitsu & Maliarik, 1993). Plants of differing species, or at varying developmental stages, may possess markedly different combinations of receptors with different binding affinities and so may respond differentially to the same elicitor. In this study, compound leaves of 45-day-old soybean seedlings were used for total RNA extraction and transcriptomic analysis. Taking into consideration of both elicitor responsiveness and growth potential, these plants may have preferentially induced immune responses over growth promotion. In addition, ANE application has been extensively reported to particularly promote root growth. In this study, a number of genes regulating starch biosynthesis were up-regulated following ANE, oligo-chitin and oligo-chitosan treatments. Increased starch biosynthesis and transport to roots has been suggested as a way for plants to reallocate energy and resources when under stress (Zhang, Persson & Giavalisco, 2013). Thus, reported root growth after ANE, oligo-chitin and oligo-chitosan treatments may partly be due to induced stress responses.

ANE, oligo-chitin and oligo-chitosan treatments generally induced energy-saving and stress responses; this is quite the opposite to the induced responses of the activated TOR (target of rapamycin) pathway. The TOR pathway is highly conserved amongst eukaryotes and is found in organisms as diverse as yeasts, plants and humans (Xiong & Sheen, 2015). When conditions are favorable, the TOR pathway is activated; by phosphorylation of downstream targets, this pathway promotes cell proliferation via increases in DNA, RNA and protein synthesis and, consequently, the expression of cell cycle-related genes, as showed in Figure 63 (Xiong & Sheen, 2015). The cost of this pathway is the inhibition of many stress- and defense-related genes, as shown in Figure 64 (Xiong & Sheen, 2015). So it would appear to be at complete cross purposes with ANE, oligo-chitin and oligo-chitosan treatments, which actively promote lipid and protein degradation and other energy-saving activities as elucidated above.

A counterpart for the TOR pathway is the SnRK1 (sucrose non-fermenting related kinase1) pathway, which is also highly conserved in all eukaryotic organisms examined to date (Hulsmans, Rodriguez, Coninck & Rolland, 2016). The SnRK1 pathway is activated in response to stress, for example, when exposed to viral, bacterial or fungal infections, or when subjected to herbivory, as shown in Figure 65 (Hulsmans, Rodriguez, Coninck & Rolland, 2016). Activation of SnRK1 inhibits TOR pathway activation, as shown in Figure 66 (Hulsmans et al., 2016), thus inhibiting energy consuming processes and plant growth in general. It is possible that treatment with ANE, oligo-chitin and oligo-chitosan to some extent activates the SnRK1 pathway, thus deactivating the TOR pathway, thereby increasing stress responses and inhibiting plant growth. Intriguingly, treatment with oligo-alginate has been shown to up-regulate expression levels of SnRK2 (a fellow member of the plant SnRK gene family) in *Triticum aestivum* (Liu et al., 2013).

To test whether the action of ANE, oligo-chitin, and oligo-chitosan treatments was involved the activation of SnRK1 or deactivation of TOR pathways, the annotated genes regulated by ANE treatment were checked. Surprisingly, ribosomal small protein 6 (RPS6), a downstream substrate of TOR-S6K (RPS6 kinase) was found to be down-regulated in ANE-, oligo-chitin- and oligo-chitosan-treated plants but not in oligo-alginate-treated plants as shown in Table 16 (Appendix 2). It has been demonstrated, together with RPS6, a histone deacetylase 2B is involved in the negative regulation of rRNA transcription (Xiong & Sheen, 2015). Ribosome biogenesis has been found to be down-regulated in ANE-, oligo-chitin-, and oligo-chitosan- treated plants. However, RPS6 and several histone deacetylase 2B-related encoding genes were down-regulated as well following treatments with ANE, oligo-chitin and oligo-chitosan, as shown in Table 43 (Appendix 2); this could be due to some unknown feedback regulations to maintain the dynamic homeostasis of ribosomal biogenesis. Significantly, the potential downstream targets of SnRK1 pathway, transcription factors of the bZIP family (Hulsmans et al., 2016), were commonly up-regulated after treatments of ANE, oligo-chitin, and oligo-chitosan but not following oligo-alginate treatment.

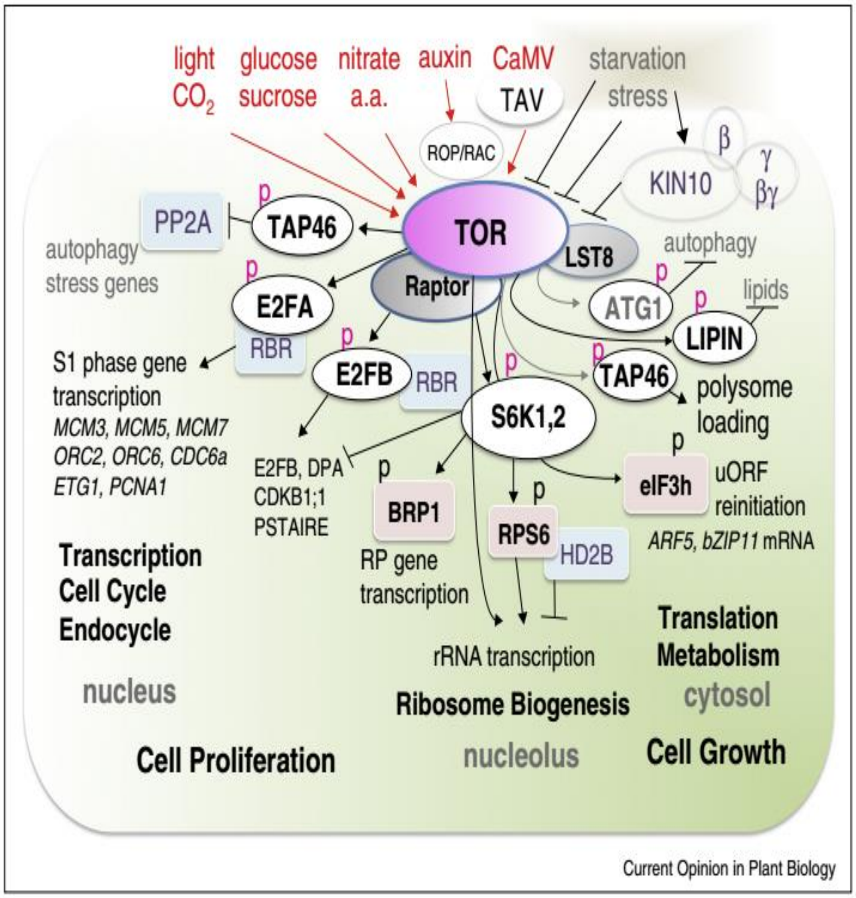
To further identify the SnRK genes in soybean and test the activation of SnRK1 in ANE-, oligo-chitin-, and oligo-chitosan- treated plants, protein sequences of the SnRK family of *Arabidopsis* were retrieved from the database (http://plantsp.genomics.purdue.edu/). SnRK gene sequences were then used to search against the soybean genome by NCBI blast software. To further infer the evolutionary relationships of these identified putative soybean SnRK gene families, *Arabidopsis* SnRK protein sequences, together with the soybean putative SnRK protein sequences, were used to construct a bootstrap phylogeny tree by using the Clustalw (Larkin et al., 2007) and Figtree software was used for the visualization. As shown in Figure 67, similar to that in *Arabidopsis*, SnRK gene families in soybeans are also divided into three distinct clades, namely SnRK1, SnRK2 and SnRK3 family members, which start with the prefix GLYMA. The majority of these genes belong to the SnRK3 clade, and only a fraction of the genes was assigned to the SnRK1 and SnRK2 clades (shown in red and green respectively). To test the involvement of the SnRK1 in this study, the expression levels of these putative genes were compared in all treatments as shown in Figure 68. The putative soybean SnRK1 gene members were underscored with red lines and the expression level for each gene was noted with an RPKM value in the heat map. The color represents the level of gene expression, with red indicating a higher level of expression and green representing a lower level of expression. Treatments of oligo-chitin and oligo-chitosan up-regulated a larger number of SnRK 2 and 3 gene families compared with those of ANE and oligo-alginate. However, the expression level of putative soybean SnRK1 genes did not show significant alterations. Nevertheless, considering the amplified cascade reaction of protein kinase and the strong evidence in terms of the responses of target genes in this study, it is possible that treatments of ANE, oligo-chitin, and oligo-chitosan all activated the SnRK1 pathway, which in turn inhibited energy consuming processes and enhanced both stress responses and secondary metabolite synthesis to ensure plant survival under stressful conditions.

ANE, oligo-chitin and oligo-chitosan treatments all inhibited photosynthesis to some degree, which likely triggered plants to increase energy-saving behaviours whilst decreasing energy-consuming activities; this, of course, inhibits plant growth, at least over the short term. Upon exposure to stress, chloroplasts, and the photosynthetic processes carried out in them, may be the most sensitive to stress of all of a plant's constituent parts. Down-regulation of photosynthesis may be the catalyst for the activation of the SnRK1 pathway and deactivation of the TOR pathway, reallocating energy and resources to mounting a successful stress response instead. This allows plants to more readily survive short-term periods of stress so they can continue to grow and thrive when the stress is removed.

To better understand the mechanism of ANE's effects on plant growth, more study is required of all its bioactive components. As laminaran and fucoidan are fundamental components of *Ascophyllum nodosum* and β-glucan and chitin are essential constituents of fungal cell walls, there may be a larger overlapping of effects using oligo-β-glucan, oligo-chitin and oligo-chitosan treatments, since plants would interpret all three as fungal invasions and react accordingly.

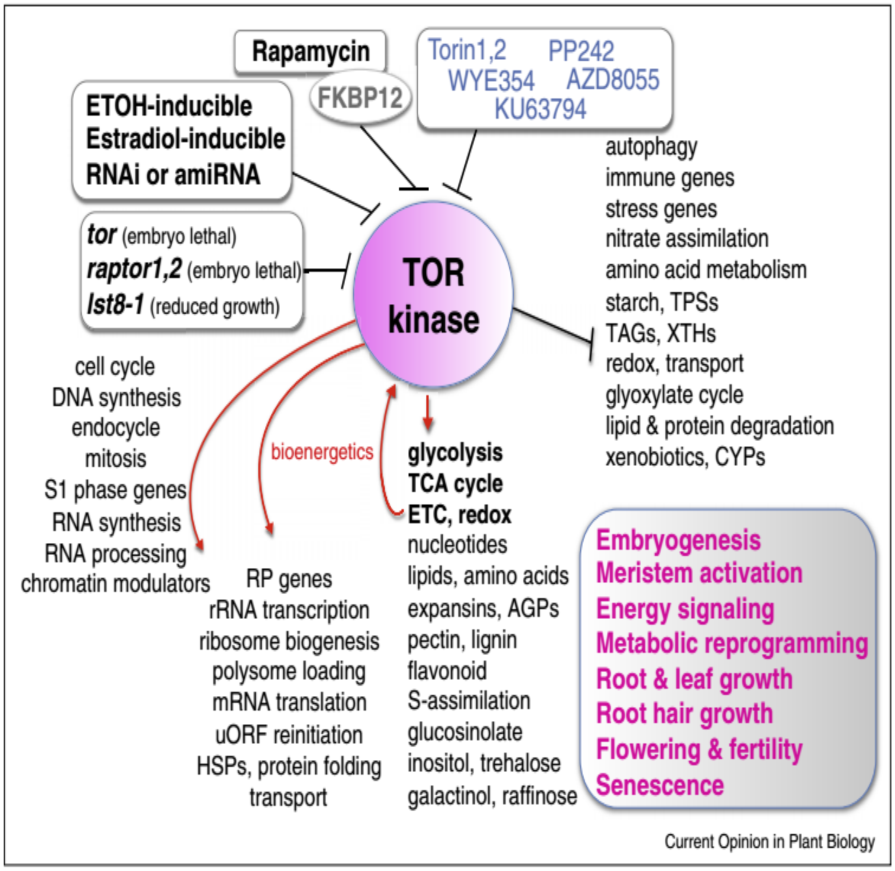
To understand the role of the SnRK1 and TOR pathways in the action of ANE, oligo-chitin and oligo-chitosan treated plants, further manipulation of the activity of SnRK1 or TOR would be essential. To further elucidate the connection between the binding of oligo-chitin to the activation of SnRK1 or the inhibition of TOR pathways, deployment of mutant plants with chitin receptors would be important. A possible mechanism explaining the effects of ANE, oligo-chitin and oligo-chitosan on plant growth is proposed and presented in Figure 69.

Results in the present study suggest oligo-chitin and oligo-chitosan can mimic the effects of ANE on plants; applications of all three, at appropriate rates for all species and developmental stages, may help prime plants so they are better prepared to withstand potential stressors and thus, in the long term, enjoy improved growth. As suggested previously, it would appear counterintuitive that ANE and oligosaccharides are said to improve plant growth when the results of this study would suggest they do precisely the opposite, but by inhibiting growth in the short-term during periods of stress, these treatments likely lead to improved plant growth over the long term life of the plant.



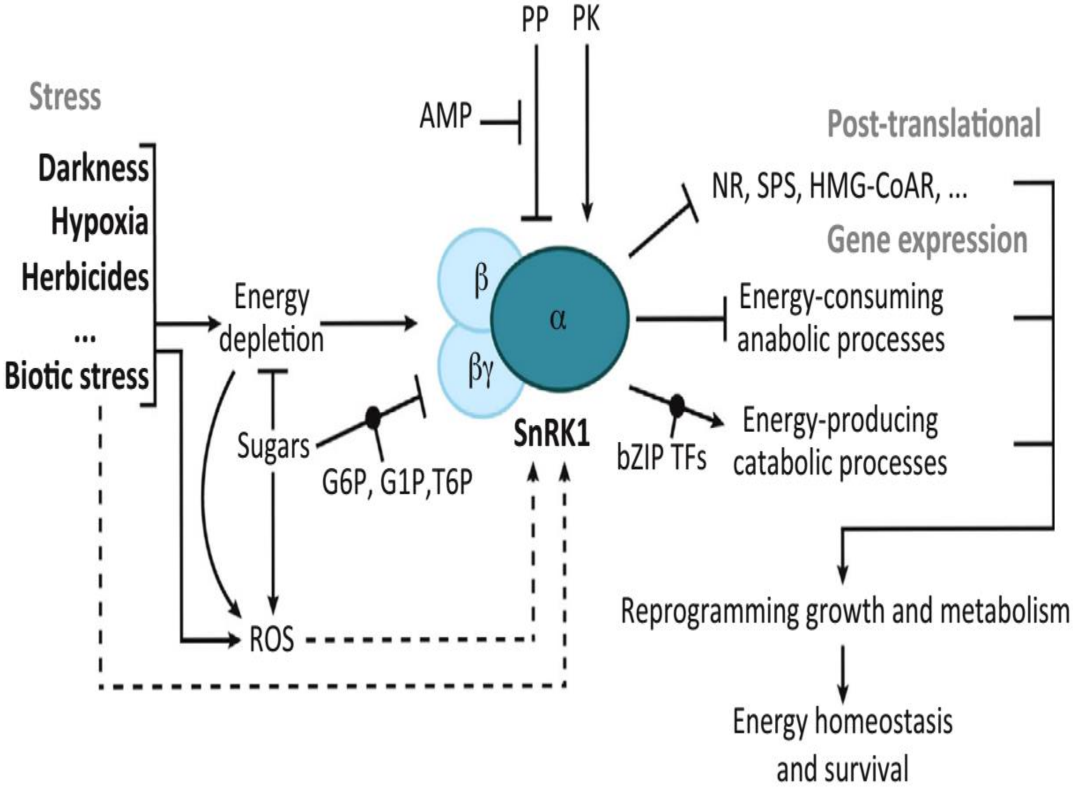
The plant TOR signaling network. *Arabidopsis* TOR kinase is modulate by diverse upstream inputs and regulatory partners (Raptor and LST8) to phosphorylate S6K1,2, TAP46, E2FA, E2FB, LIPIN and ATG1 in the nucleus, nucleolus and cytosol to control transcription, cell cycle, endocycle, rRNA transcription, ribosome biogenesis, translation and metabolism, all pivotal to cell proliferation and growth. S6K, small ribosome protein 6 (RPS6) kinase; TAP46, a regulatory subunit of PP2A; E2FA/B, transcription factors; RBR, retinoblastoma repressor; HD2B, histone deacetylase2B; eIF3h, eukaryotic translation initiation factor 3h; ATG1, autophagy related kinase1.

Figure 63: Various regulators involved in plant TOR and SnRK1 pathways (Xiong & Sheen, 2015), Current Opinion in Plant Biology, Figure 3.



The multifaceted functions of TOR kinase are uncovered by integrated chemical, genetic, genomic and metabolomics analyses. The complex TOR signaling network contributes to the regulation of plant life from embryogenesis to senescence by integrating central and secondary carbon metabolism with bioenergetics, biosynthesis, signaling, chromatin modulators, transporters, autophagy and cell cycle regulation. RNAi, RNA interference; amiRNA, artificial microRNA; RP, ribosome protein; uORF, upstream open reading frame; HSP, heat shock protein; TCA, tricarboxylic acid cycle; ETC, mitochondria electron transport chain; AGP, arabinogalactan protein; TPS, trehalose-6-phosphate synthase; TAG, triacylglycerol; XTH, xyloglucan endotransglucosylase; CYP, cytochrome P450. Torin1,2, WYE354, KU63794, PP242 and AZD8055 are specific ATP-competitive chemical inhibitors of TOR kinase.

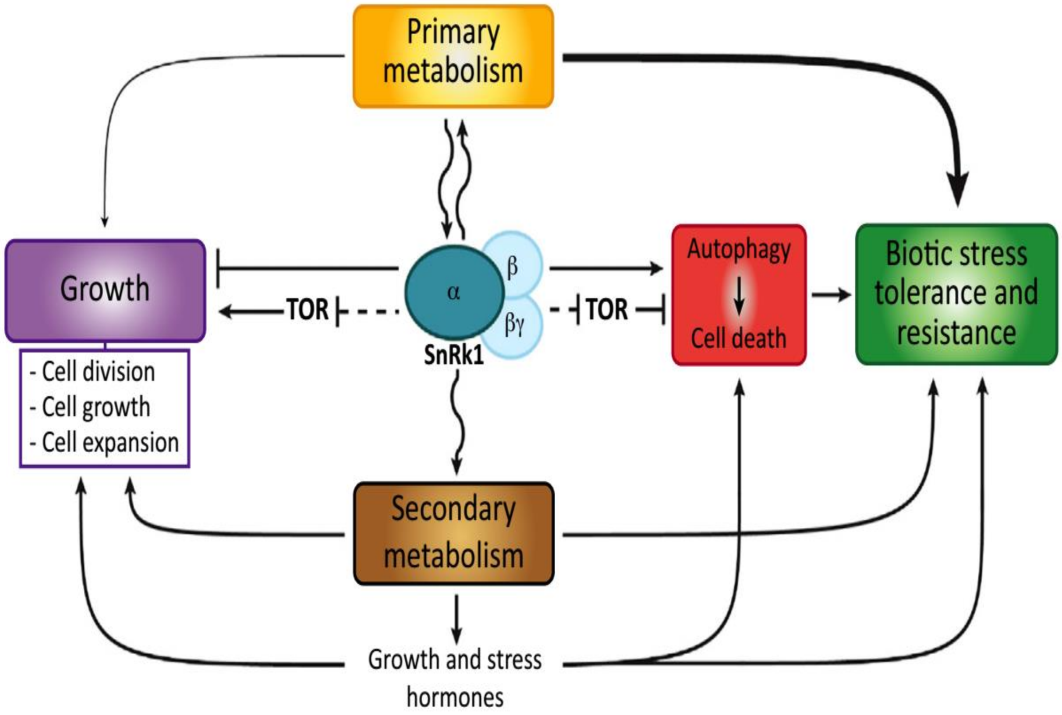
Figure 64: TOR pathways influence biological processes (Xiong & Sheen, 2015), Current Opinion in Plant Biology, Figure 2.



Trends in Plant Science

Figure 1. The Plant SnRK1 Signaling Pathway. The heterotrimeric SnRK1 complex is activated by diverse energy-depleting stress conditions and maintains energy homeostasis by post-translational modification of key metabolic enzymes such as sucrose phosphate synthase (SPS), nitrate reductase (NR), and 3-hydroxy-3-methylglutaryl-CoA reductase (HMG-CoAR), as well as by transcriptional reprogramming mediated in part by bZIP (basic leucine zipper) transcription factors (TF). In plants, a hybrid βγ protein functions as the canonical α-subunit. AMP inhibits α-subunit dephosphorylation and inactivation. Sugar phosphates including glucose-6-P (G6P), glucose-1-P (G1P), and trehalose-6-P (T6P) can act as allosteric inhibitors. Redox regulation is likely also involved in the control of SnRK1 signaling. Abbreviations: PK, protein kinase; PP, protein phosphatase; ROS, reactive oxygen species; SnRK1, SNF1-related kinase 1.

Figure 65: Various factors that influence plant SnRK1 pathways (Hulsmans et al., 2016), Trends in Plant Science, Figure 1.



Trends in Plant Science

Figure 3. Possible Mechanisms of SnRK1-Mediated Metabolic Regulation of Plant Biotic Interactions, In Addition to Direct SnRK1 Interaction with Pathogen and Plant Innate Immunity Signaling Proteins. As a key regulator of primary metabolism, SnRK1 is likely involved in shifting carbon and energy use from growth-associated processes to survival and defense upon pathogen attack (depicted by the bold arrow). In addition, it controls secondary metabolism that not only plays a key role in (chemical and physical) defense but also produces precursors for different growth and stress hormones. Metabolic (sugar) status is also known to affect hormone sensitivity and signaling. Finally, several lines of evidence suggest that SnRK1 (likely in part through inhibition of TOR, indicated by the dashed lines) more directly limits growth and can stimulate cell death mediated by autophagy. Undulating arrows indicate modification, including both positive and negative regulation of specific pathways. For more details and references, see text.

Figure 66: Plant SnRK1 pathway influenced various biological processes ((Hulsmans et al., 2016), Trends in Plant Science, Figure 3.

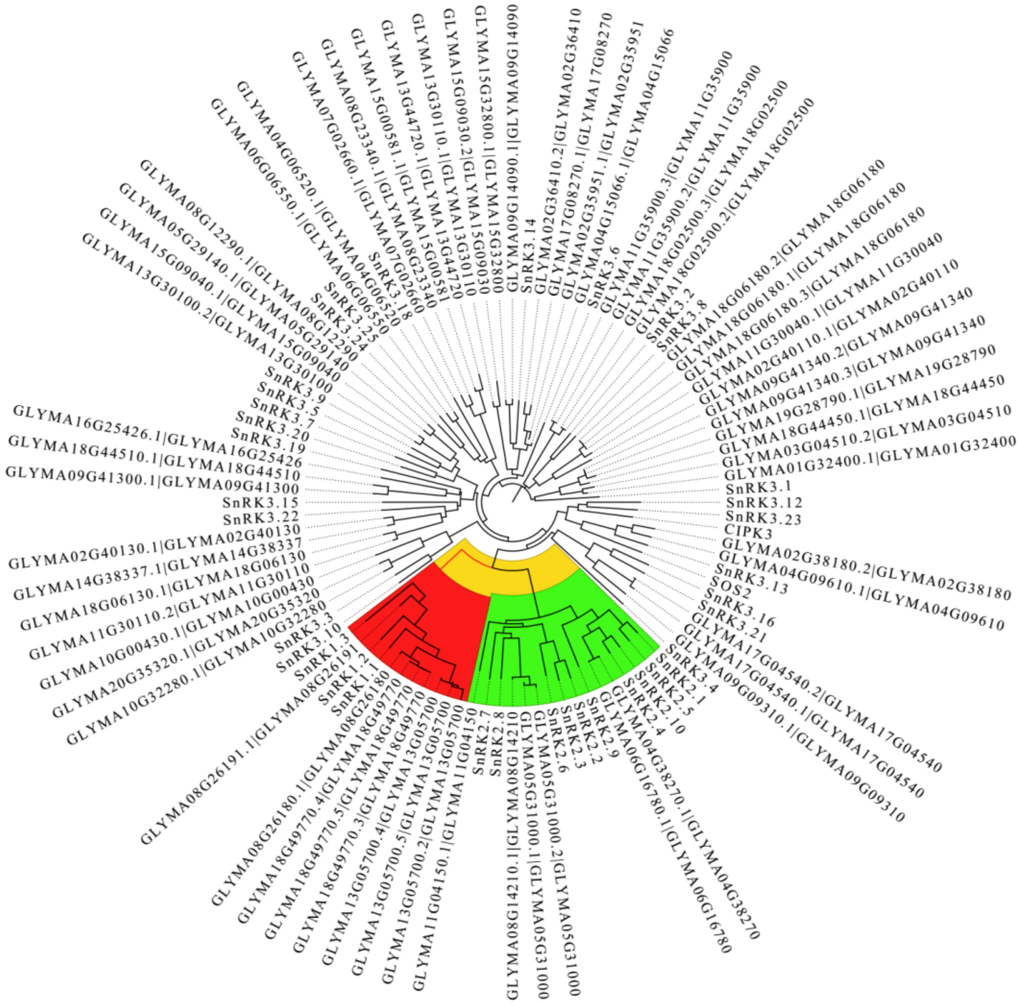
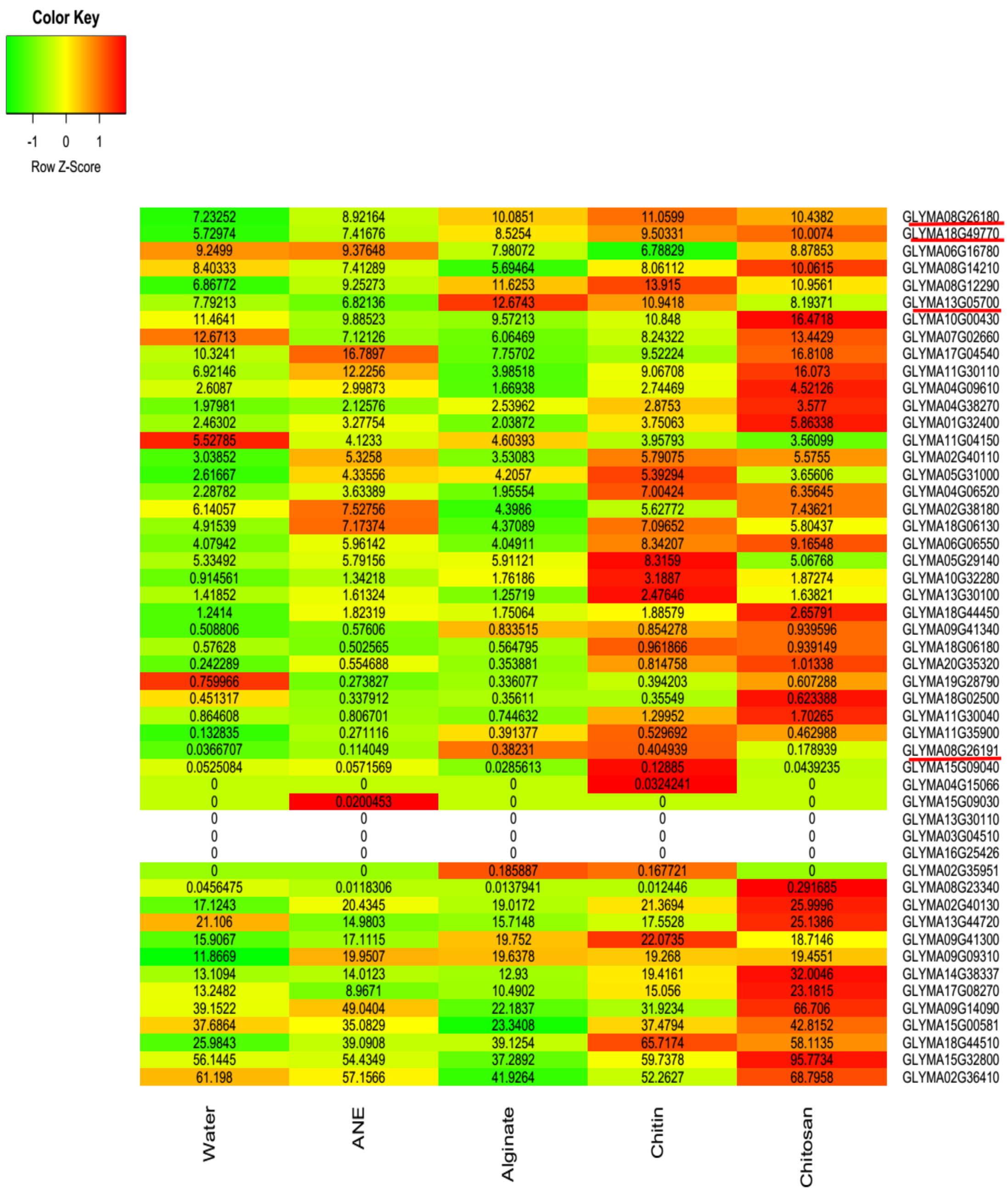


Figure 67: Putative SnRK gene family members in soybean.



Water	ANE	Alginate	Chitin	Chitosan	
7.23252	8.92164	10.0851	11.0599	10.4382	GLYMA08G26180
5.72974	7.41676	8.5254	9.50331	10.0074	GLYMA18G49770
9.2499	9.37648	7.98072	6.78829	8.87853	GLYMA06G16780
8.40333	7.41289	5.69464	8.06112	10.0615	GLYMA08G14210
6.86772	9.25273	11.6253	13.915	10.9561	GLYMA08G12290
7.79213	6.82136	12.6743	10.9418	8.19371	GLYMA13G05700
11.4641	9.88523	9.57213	10.848	16.4718	GLYMA10G00430
12.6713	7.12126	6.06469	8.24322	13.4429	GLYMA07G02660
10.3241	16.7897	7.75702	9.52224	16.8108	GLYMA17G04540
6.92146	12.2256	3.98518	9.06708	16.073	GLYMA11G30110
2.6087	2.99873	1.66938	2.74469	4.52126	GLYMA04G09610
1.97981	2.12576	2.53962	2.8753	3.577	GLYMA04G38270
2.46302	3.27754	2.03872	3.75063	5.86338	GLYMA01G32400
5.52785	4.1233	4.60393	3.95793	3.56099	GLYMA11G04150
3.03852	5.3258	3.53083	5.79075	5.5755	GLYMA02G40110
2.61667	4.33556	4.2057	5.39294	3.65606	GLYMA05G31000
2.28782	3.63389	1.95554	7.00424	6.35645	GLYMA04G06520
6.14057	7.52756	4.3986	5.62772	7.43621	GLYMA02G38180
4.91539	7.17374	4.37089	7.09652	5.80437	GLYMA18G06130
4.07942	5.96142	4.04911	8.34207	9.16548	GLYMA06G06550
5.33492	5.79156	5.91121	8.3159	5.06768	GLYMA05G29140
0.914561	1.34218	1.76186	3.1887	1.87274	GLYMA10G32280
1.41852	1.61324	1.25719	2.47646	1.63821	GLYMA13G30100
1.2414	1.82319	1.75064	1.88579	2.65791	GLYMA18G44450
0.508806	0.57606	0.833515	0.854278	0.939596	GLYMA09G41340
0.57628	0.502565	0.564795	0.961866	0.939149	GLYMA18G06180
0.242289	0.554688	0.353881	0.814758	1.01338	GLYMA20G35320
0.759966	0.273827	0.336077	0.394203	0.607288	GLYMA19G28790
0.451317	0.337912	0.35611	0.35549	0.623388	GLYMA18G02500
0.864608	0.806701	0.744632	1.29952	1.70265	GLYMA11G30040
0.132835	0.271116	0.391377	0.529692	0.462988	GLYMA11G35900
0.0366707	0.114049	0.38231	0.404939	0.178939	GLYMA08G26191
0.0525084	0.0571569	0.0285613	0.12885	0.0439235	GLYMA15G09040
0	0	0	0.0324241	0	GLYMA04G15066
0	0.0200453	0	0	0	GLYMA15G09030
0	0	0	0	0	GLYMA13G30110
0	0	0	0	0	GLYMA03G04510
0	0	0	0	0	GLYMA16G25426
0	0	0.185887	0.167721	0	GLYMA02G35951
0.0456475	0.0118306	0.0137941	0.012446	0.291685	GLYMA08G23340
17.1243	20.4345	19.0172	21.3694	25.9996	GLYMA02G40130
21.106	14.9803	15.7148	17.5528	25.1386	GLYMA13G44720
15.9067	17.1115	19.752	22.0735	18.7146	GLYMA09G41300
11.8669	19.9507	19.6378	19.268	19.4551	GLYMA09G09310
13.1094	14.0123	12.93	19.4161	32.0046	GLYMA14G38337
13.2482	8.9671	10.4902	15.056	23.1815	GLYMA17G08270
39.1522	49.0404	22.1837	31.9234	66.706	GLYMA09G14090
37.6864	35.0829	23.3408	37.4794	42.8152	GLYMA15G00581
25.9843	39.0908	39.1254	65.7174	58.1135	GLYMA18G44510
56.1445	54.4349	37.2892	59.7378	95.7734	GLYMA15G32800
61.198	57.1566	41.9264	52.2627	68.7958	GLYMA02G36410

Figure 68: Expression patterns of putative SnRK genes under different treatments.

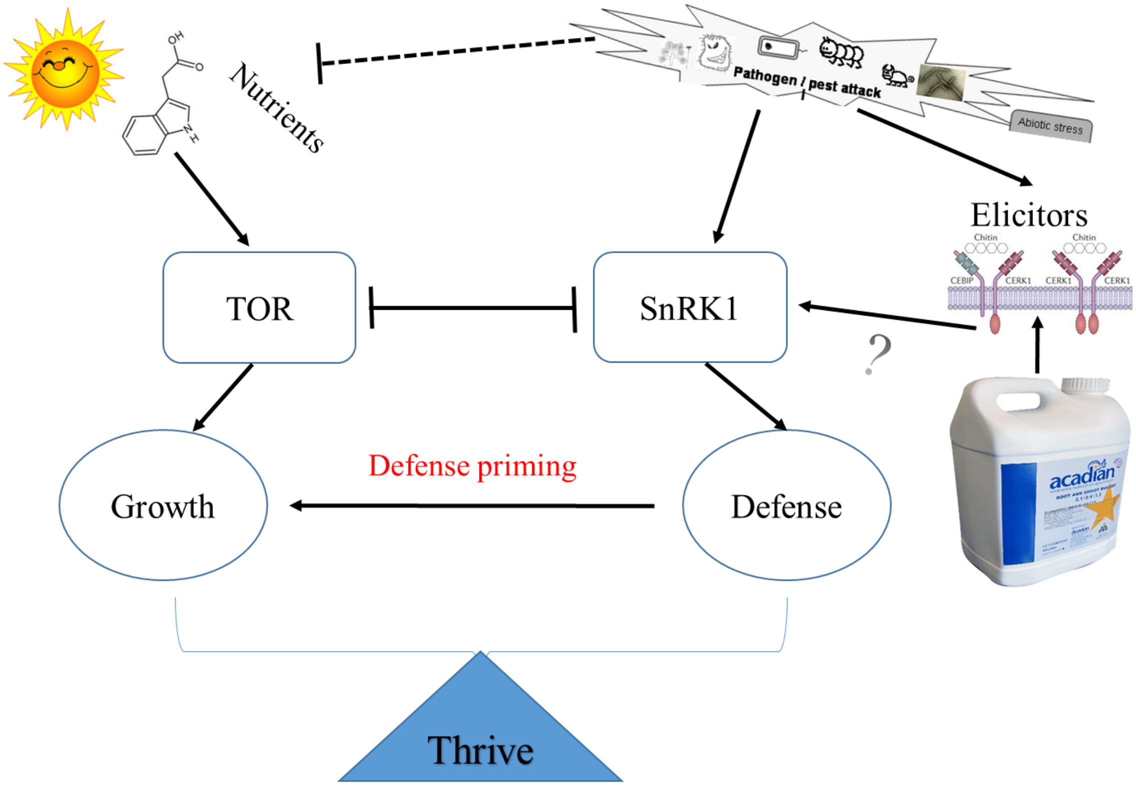


Figure 69: Possible mechanism for ANE, oligo-chitin and, chitosan treatments.

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Appendix 1

ANE up-regulated gene function annotations

Table 4: ANE up-regulated genes involved in plant defense and stress responses.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA08G24750	Q9SSK5	1.90	1.67	1.36	3.87	MLP-like protein 43	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE-Alginate-Chitin-Chitosan	GLYMA07G39900	I1KNM2	2.45	1.81	1.41	2.17	Uncharacterized protein	response to stress (GO:0006950);
ANE-Alginate-Chitin-Chitosan	GLYMA16G22488	Q6NQK2	2.03	2.98	1.77	1.22	NAC domain-containing protein 8	DNA damage checkpoint (GO:0000077); response to gamma radiation (GO:0010332);
ANE-Alginate	GLYMA15G19500	Q39012	1.53	1.12	0.00	0.00	Shaggy-related protein kinase iota	brassinosteroid-mediated signaling pathway (GO:0009742); hyperosmotic salinity response (GO:0042538);
ANE-Alginate-Chitosan	GLYMA15G30110	Q9SSK5	1.49	3.16	0.40	3.10	MLP-like protein 43	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE-Alginate-Chitosan	GLYMA07G30440	Q9SM02	1.10	1.20	-0.03	2.13	Squalene epoxidase 1	response to water deprivation (GO:0009414); sterol biosynthetic process (GO:0016126);
ANE-Alginate-Chitosan	GLYMA19G03770	Q940Z5	1.25	1.22	-0.94	1.51	Phenolic glucoside malonyltransferase 1	response to toxic substance (GO:0009636);
ANE-Chitin-Chitosan	GLYMA03G35930	Q9SJ52	1.96	0.08	1.71	1.65	Protein YLS9	defense response to virus (GO:0051607);
ANE-Chitin-Chitosan	GLYMA07G01461	O82155	1.16	0.50	1.46	1.20	Dof zinc finger protein DOF1.7	response to chitin (GO:0010200);
ANE-Chitin-Chitosan	GLYMA18G02510	Q8VZR7	1.12	0.41	1.73	1.32	Protein NRT1/ PTR FAMILY 5.1	response to nematode (GO:0009624);
ANE-Chitin-Chitosan	GLYMA17G32691	P93604	1.10	0.22	1.27	1.83	Rust resistance kinase Lr10	Novel disease-related genes; involved in regulation of cell death
ANE-Chitin-Chitosan	GLYMA17G32830	P93604	1.08	0.38	1.38	1.36	Rust resistance kinase Lr10	Novel disease-related genes; involved in regulation of cell death
ANE-Chitin-Chitosan	GLYMA02G08600	Q9M9H6	1.59	-0.07	1.13	1.20	Probable choline kinase 1	CDP-choline pathway (GO:0006657); response to wounding (GO:0009611);
ANE-Chitin-Chitosan	GLYMA11G02620	Q8LBU2	1.99	0.00	1.62	1.58	Probable glutathione peroxidase 8	cellular oxidant detoxification (GO:0098869); response to oxidative stress (GO:0006979);

ANE-Chitin-Chitosan	GLYMA19G41660	I1NBS0	1.22	0.21	1.35	1.52	Uncharacterized protein	response to stress (GO:0006950);
ANE-Chitin-Chitosan	GLYMA10G41370	Q8L731	1.17	0.00	2.07	1.52	Protein DETOXIFICATION 12	No-GOBP
ANE-Chitin-Chitosan	GLYMA01G26230	P32110	1.17	0.58	1.55	1.62	Probable glutathione S-transferase	glutathione metabolic process (GO:0006749); toxin catabolic process (GO:0009407);
ANE-Chitin-Chitosan	GLYMA10G00220	Q8GT20	2.24	0.00	2.03	1.06	Benzyl alcohol O-benzoyltransferase	Produced in damaged leaves; (D'Auria, Chen, & Pichersky, 2002)
ANE-Chitin	GLYMA14G03470	O80852	1.13	0.20	1.07	0.68	Glutathione S-transferase F9	cellular oxidant detoxification (GO:0098869); defense response (GO:0006952);
ANE-Chitin	GLYMA15G24010	Q9LMA1	1.40	0.66	1.03	0.94	Probable flavin-containing monooxygenase 1	cellular response to hypoxia (GO:0071456); defense response signaling pathway, resistance gene-dependent (GO:0009870);
ANE-Chitin	GLYMA01G36620	Q38853	1.69	0.00	1.26	0.90	Rhodanese-like domain-containing protein 15, chloroplastic	aging (GO:0007568); response to jasmonic acid (GO:0009753); response to oxidative stress (GO:0006979); response to wounding (GO:0009611);
ANE-Chitin	GLYMA01G43933	O54408	1.08	0.24	1.01	0.69	GTP pyrophosphokinase	response to starvation (GO:0042594);
ANE-Chitin	GLYMA05G33650	Q9LVJ0	1.15	0.83	1.70	0.80	UV-B-induced protein, chloroplastic	response to UV-B (GO:0010224); response to wounding (GO:0009611);
ANE-Chitin	GLYMA06G12400	O80662	1.10	0.14	1.08	0.88	Glutathione S-transferase TCHQD	response to toxic substance (GO:0009636);
ANE-Chitin	GLYMA06G17020	Q6H795	1.27	0.60	1.50	0.53	Chaperone protein ClpD1, chloroplastic	cellular response to heat (GO:0034605);
ANE-Chitin	GLYMA07G01400	Q67YC0	1.20	0.81	1.56	0.19	Inorganic pyrophosphatase 1	cellular response to phosphate starvation (GO:0016036);
ANE-Chitin	GLYMA08G06070	Q9LVJ0	1.14	0.87	1.53	0.75	UV-B-induced protein, chloroplastic	response to UV-B (GO:0010224); response to wounding (GO:0009611);
ANE-Chitin	GLYMA15G11910	P80471	1.49	0.35	1.46	0.75	Light-induced protein, chloroplastic	response to stress (GO:0006950);
ANE-Chitin	GLYMA17G13160	V7CAG1	1.10	0.00	1.23	0.00	Uncharacterized protein	response to stress (GO:0006950);
ANE-Chitin	GLYMA17G16570	Q8S7M7	1.09	0.45	1.46	0.73	Plant intracellular Ras-group-related LRR protein 5	response to light stimulus (GO:0009416);
ANE-Chitin	GLYMA15G11410	Q9LVD9	1.27	0.54	1.20	0.60	Protein DETOXIFICATION 40	No-GOBP
ANE-Chitin	GLYMA17G36420	Q9LZ25	1.06	0.84	1.14	0.66	Probable disease resistance protein	defense response to bacterium (GO:0042742);

ANE-Chitin	GLYMA13G43880	Q76CU2	1.20	0.23	1.27	0.67	Pleiotropic drug resistance protein 1	transport (GO:0006810);
ANE-Chitin	GLYMA09G02170	Q8RWS6	1.13	0.67	1.21	0.32	Ferric reduction oxidase 6	ion transport (GO:0006811); response to light stimulus (GO:0009416);
ANE-Chitosan	GLYMA09G05230	Q96262	1.35	-0.65	0.45	2.34	Plasma membrane-associated cation-binding protein 1	cellular response to molecule of bacterial origin (GO:0071219); response to water deprivation (GO:0009414); viral process (GO:0016032);
ANE-Chitosan	GLYMA10G06960	P83332	1.47	0.00	0.00	1.70	Thaumatin-like protein 1	defense response (GO:0006952);
ANE-Chitosan	GLYMA15G16560	Q96262	1.14	-0.78	0.38	1.83	Plasma membrane-associated cation-binding protein 1	cellular response to molecule of bacterial origin (GO:0071219); response to water deprivation (GO:0009414); viral process (GO:0016032);
ANE	GLYMA03G03640	O65782	1.14	0.55	0.57	0.10	Cytochrome P450 83B1	adventitious root development (GO:0048830); defense response by callose deposition in cell wall (GO:0052544);
ANE	GLYMA13G25630	Q3EAQ5	1.10	0.00	0.00	0.26	Probable peptide/nitrate transporter	response to nematode (GO:0009624); transmembrane transport (GO:0055085);
ANE	GLYMA05G27290	Q9LRR9	1.06	0.00	0.94	0.94	Peroxisomal (S)-2-hydroxy-acid oxidase GLO1	defense response to bacterium (GO:0042742);
ANE	GLYMA06G07800	Q8VZK0	1.03	0.00	0.29	0.00	E3 ubiquitin-protein ligase MIEL1	defense response (GO:0006952);
ANE	GLYMA16G03550	O81825	1.00	0.69	0.00	0.20	Probable disease resistance protein	defense response (GO:0006952);
ANE	GLYMA19G38590	Q9SJ52	1.08	0.00	0.00	0.00	Protein YLS9	defense response to virus (GO:0051607); leaf senescence (GO:0010150); response to other organism (GO:0051707); signal transduction (GO:0007165);
ANE	GLYMA01G42860	Q8LBU2	1.05	-0.78	0.07	0.63	Probable glutathione peroxidase 8	cellular oxidant detoxification (GO:0098869); response to oxidative stress (GO:0006979);
ANE	GLYMA03G36310	Q93YS4	1.15	0.00	0.70	0.33	ABC transporter G family member 22	response to water deprivation (GO:0009414); transmembrane transport (GO:0055085); transpiration (GO:0010148);
ANE	GLYMA06G03850	O49394	1.02	0.11	0.54	0.00	Cytochrome P450 82C2	cellular response to hypoxia (GO:0071456); secondary metabolite biosynthetic process (GO:0044550);
ANE	GLYMA06G44981	P34913	1.04	0.76	0.50	0.00	Bifunctional epoxide hydrolase 2	reactive oxygen species metabolic process (GO:0072593); xenobiotic metabolic process (GO:0006805);
ANE	GLYMA09G01660	Q94B74	1.08	0.00	0.55	0.68	Nudix hydrolase 2	response to oxidative stress (GO:0006979);

ANE	GLYMA10G01610	Q9LZP9	1.22	0.06	0.83	0.80	Calvin cycle protein CP12-2, chloroplastic	cellular response to anoxia (GO:0071454); cellular response to cold (GO:0070417); cellular response to heat (GO:0034605);
ANE	GLYMA10G30130	P47999	1.13	-0.17	0.00	0.13	Cysteine synthase, chloroplastic/chromoplastic	response to cadmium ion (GO:0046686); response to cytokinin (GO:0009735);
ANE	GLYMA10G36600	Q9SQI2	1.02	0.04	0.88	0.30	Protein GIGANTEA	response to cold (GO:0009409);
ANE	GLYMA11G29460	Q9S9N9	1.09	0.78	0.67	0.84	Cinnamoyl-CoA reductase 1	circadian rhythm (GO:0007623); lignin biosynthetic process (GO:0009809); response to cold (GO:0009409);
ANE	GLYMA18G06510	Q9S9N9	1.08	-0.37	0.08	0.00	Cinnamoyl-CoA reductase 1	circadian rhythm (GO:0007623); lignin biosynthetic process (GO:0009809); response to cold (GO:0009409);
ANE	GLYMA08G27342	Q91DM0	1.17	0.00	0.00	0.80	Genome polyprotein	transport of virus in host, cell to cell (GO:0046740);
ANE	GLYMA12G10620	Q9LVD9	1.34	-0.25	0.99	0.49	Protein DETOXIFICATION 40	No-GOBP
ANE	GLYMA07G36530	P22242	1.02	0.00	0.00	0.00	Desiccation-related protein PCC13-62	No-GOBP

Table 5: ANE up-regulated genes involved in flavonoids and terpenoid synthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA08G27260	Q84KK5	1.65	1.46	2.35	2.03	Isoflavone 7-O-methyltransferase	isoflavonoid biosynthetic process (GO:0009717);
ANE-Alginate-Chitin-Chitosan	GLYMA13G06151	Q9SGA8	2.61	1.07	2.14	2.04	UDP-glycosyltransferase 83A1	No-GOBP
ANE-Alginate-Chitosan	GLYMA16G03760	Q2V6J9	1.30	1.44	-0.64	1.69	UDP-glucose flavonoid 3-O-glucosyltransferase 7	No-GOBP
ANE-Alginate-Chitosan	GLYMA02G11640	Q8W491	1.35	1.07	-0.66	1.65	UDP-glycosyltransferase 73B3	defense response (GO:0006952); flavonoid biosynthetic process (GO:0009813);
ANE-Alginate-Chitosan	GLYMA18G12660	Q9SYM5	1.58	2.20	0.26	1.70	Trifunctional UDP-glucose 4,6-dehydratase/UDP-4-keto-6-Deoxy-D-glucose 3,5-epimerase/UDP-4-keto-L-rhamnose-reductase RHM1	auxin efflux (GO:0010315); flavonol biosynthetic process (GO:0051555);
ANE-Chitin-Chitosan	GLYMA06G40390	Q9C9B0	1.11	0.25	1.49	1.08	UDP-glycosyltransferase 89B1	flavonoid biosynthetic process (GO:0009813);

ANE-Chitin-Chitosan	GLYMA19G27930	P22928	1.26	0.38	1.36	1.08	Chalcone synthase J	flavonoid biosynthetic process (GO:0009813);
ANE-Chitin	GLYMA08G11620	P24826	1.09	0.89	1.31	0.91	Chalcone synthase 1	flavonoid biosynthetic process (GO:0009813);
ANE-Chitin	GLYMA10G37660	F4JTB3	1.01	0.32	1.34	0.85	Protein DETOXIFICATION 35	flavonoid biosynthetic process (GO:0009813);
ANE-Chitin	GLYMA11G01350	P23569	1.22	0.59	2.38	0.97	Chalcone synthase	flavonoid biosynthetic process (GO:0009813);
ANE-Chitin	GLYMA14G37170	D3UAG2	1.30	0.00	1.67	0.71	UDP-glycosyltransferase 71K2	No-GOBP
ANE-Chitin	GLYMA08G07130	Q9LFJ8	1.17	0.76	1.93	0.65	UDP-glycosyltransferase 78D2	flavonoid biosynthetic process (GO:0009813);
ANE-Chitosan	GLYMA15G34720	D4Q9Z4	1.16	0.80	-0.50	1.36	Soyasapogenol B glucuronide galactosyltransferase	flavonoid biosynthetic process (GO:0009813);
ANE-Chitosan	GLYMA02G01740	O22264	1.39	0.00	0.70	1.13	Transcription factor MYB12	flavonoid biosynthetic process (GO:0009813);
ANE	GLYMA03G34420	Q9ZQ99	1.10	0.27	0.95	0.11	UDP-glycosyltransferase 73C1	flavonoid biosynthetic process (GO:0009813);
ANE-Alginate-Chitosan	GLYMA20G28620	Q8VWZ7	1.39	3.23	0.01	2.53	Geraniol 8-hydroxylase	monoterpenoid biosynthetic process (GO:0016099);
ANE-Chitosan	GLYMA09G40385	Q8VWZ7	1.25	0.00	0.67	1.56	Geraniol 8-hydroxylase	monoterpenoid biosynthetic process (GO:0016099);
ANE-Chitosan	GLYMA07G00330	E2IUA6	1.09	0.90	-0.16	1.59	Taraxerol synthase	triterpenoid biosynthetic process (GO:0016104);

Table 6: ANE up-regulated genes involved in other biosynthetic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA12G12230	Q6WUC2	1.71	1.70	1.27	1.37	(R, S)-reticuline 7-O-methyltransferase	alkaloid biosynthetic process (GO:0009821);
ANE-Alginate-Chitin-Chitosan	GLYMA05G31450	Q9LW96	1.78	1.36	1.65	1.01	Inositol-3-phosphate synthase	inositol biosynthetic process (GO:0006021); phospholipid biosynthetic process (GO:0008654);
ANE-Alginate-Chitin-Chitosan	GLYMA08G14670	Q9LW96	2.85	1.41	2.25	2.10	Inositol-3-phosphate synthase	inositol biosynthetic process (GO:0006021); phospholipid biosynthetic process (GO:0008654);
ANE-Alginate-Chitin	GLYMA03G39050	Q9XGI7	1.21	1.81	3.09	0.00	Nicotianamine synthase	nicotianamine biosynthetic process (GO:0030418);
ANE-Alginate-Chitin	GLYMA19G41630	Q9XGI7	1.49	1.05	2.69	0.93	Nicotianamine synthase	nicotianamine biosynthetic process (GO:0030418);
ANE-Alginate-Chitin	GLYMA07G12741	F4HVY0	1.20	1.08	1.26	0.10	Protein ECERIFERUM 1	fatty acid biosynthetic process (GO:0006633);

ANE-Alginate-Chitosan	GLYMA01G42450	P54873	1.15	1.21	-1.13	1.03	Hydroxymethylglutaryl-CoA synthase	isopentenyl diphosphate biosynthetic process, mevalonate pathway (GO:0019287); sterol biosynthetic process (GO:0016126);
ANE-Alginate-Chitosan	GLYMA13G42330	P38417	1.42	2.36	0.51	2.88	Linoleate 9S-lipoxygenase-4	oxylipin biosynthetic process (GO:0031408);
ANE-Alginate-Chitosan	GLYMA15G03050	P38417	1.49	1.77	0.36	2.60	Linoleate 9S-lipoxygenase-4	oxylipin biosynthetic process (GO:0031408);
ANE-Alginate-Chitosan	GLYMA07G00900	P24095	1.45	1.63	0.61	1.35	Seed linoleate 9S-lipoxygenase	oxylipin biosynthetic process (GO:0031408);
ANE-Alginate-Chitosan	GLYMA18G10260	Q500U8	1.35	1.41	0.20	1.17	Tetrameter alpha-pyrone reductase 1	pollen development (GO:0009555); pollen exine formation (GO:0010584); seed development (GO:0048316); sporopollenin biosynthetic process (GO:0080110);
ANE-Alginate-Chitosan	GLYMA08G42440	O64470	1.77	1.19	0.00	1.81	Spermidine hydroxycinnamoyl transferase	pollen development (GO:0009555); pollen exine formation (GO:0010584); spermidine hydroxycinnamate conjugate biosynthetic process (GO:0080088);
ANE-Alginate-Chitosan	GLYMA08G46520	Q9XHC6	1.10	1.35	-0.32	1.55	Beta-amyrin 24-hydroxylase	secondary metabolite biosynthetic process (GO:0044550);
ANE-Alginate	GLYMA06G45043	Q6WUC2	1.19	1.64	0.84	0.42	(R, S)-reticuline 7-O-methyltransferase	alkaloid biosynthetic process (GO:0009821);
ANE-Chitin-Chitosan	GLYMA18G35220	Q9SKK4	1.58	0.62	2.07	1.37	Probable 2-oxoacid dependent dioxygenase	glucosinolate biosynthetic process (GO:0019761);
ANE-Chitin	GLYMA20G02220	Q88BD4	1.13	0.27	1.16	0.80	Phosphomannomutase/phosphoglucomutase	alginic acid biosynthetic process (GO:0042121);
ANE-Chitin	GLYMA10G37560	B3SGL0	1.34	-0.03	1.20	0.76	Beta-carotene 3-hydroxylase, chloroplastic	carotenoid biosynthetic process (GO:0016117); fatty acid biosynthetic process (GO:0006633);
ANE-Chitin	GLYMA08G19410	O48923	1.40	0.74	1.56	0.53	Cytochrome P450 71D10	secondary metabolite biosynthetic process (GO:0044550);
ANE-Chitin	GLYMA15G05580	O48923	1.13	0.33	1.19	0.25	Cytochrome P450 71D10	secondary metabolite biosynthetic process (GO:0044550);
ANE-Chitin	GLYMA06G48200	F4JLK2	1.79	0.00	2.36	0.86	Probable sucrose-phosphate synthase 4	sucrose biosynthetic process (GO:0005986);
ANE-Chitin	GLYMA20G37280	P32260	1.01	0.61	1.10	0.56	Cysteine synthase, chloroplastic/chromoplastic	cysteine biosynthetic process from serine (GO:0006535);
ANE-Chitosan	GLYMA07G21100	Q7XYS3	1.81	-0.13	0.77	4.00	Allene oxide synthase 2	oxylipin biosynthetic process (GO:0031408); sterol metabolic process (GO:0016125);
ANE-Chitosan	GLYMA08G06810	O48651	1.47	0.92	0.02	1.72	Squalene monooxygenase	sterol biosynthesis rate limiting
ANE	GLYMA01G02691	U3U992	1.06	-0.16	0.41	0.05	7-deoxyloganetic acid glucosyltransferase	(-)-secologanin biosynthetic process (GO:1900994);
ANE	GLYMA11G03070	Q9LQ10	1.05	0.00	0.58	0.59	Probable aminotransferase ACS10	biosynthetic process (GO:0009058);

ANE	GLYMA16G10880	Q9XFS9	1.15	0.39	0.87	0.19	1-Deoxy-D-xylulose 5-phosphate reductoisomerase, chloroplastic	isopentenyl diphosphate biosynthetic process, methylerythritol 4-phosphate pathway (GO:0019288);
ANE	GLYMA16G05580	Q9LW26	1.44	0.01	0.00	0.47	Acyltransferase-like protein At3g26840, chloroplastic	phytol metabolic process (GO:0033306); triglyceride biosynthetic process (GO:0019432);
ANE	GLYMA17G01710	Q8W4K1	1.08	0.00	0.17	0.31	tRNA wybutosine-synthesizing protein 2/3/4	wybutosine biosynthetic process (GO:0031591); (ensure the accuracy protein translation)
ANE	GLYMA06G00800	Q9T0I8	1.55	-0.18	0.88	0.94	5'-methylthioadenosine/S-adenosylhomocysteine nucleosidase 1	L-methionine biosynthetic process from methylthioadenosine (GO:0019509);
ANE	GLYMA06G42180	Q39580	1.02	0.04	0.52	0.34	Dynein 8 kDa light chain, flagellar outer arm	methionine biosynthetic process (GO:0009086);

Table 7: ANE up-regulated genes involved in cell wall biogenesis.

Treatments	Gene IDs	Unipart IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitosan	GLYMA04G43470	Q651X7	1.80	1.78	-0.23	1.27	Cellulose synthase-like protein E1	cellulose biosynthetic process (GO:0030244);
ANE-Alginate-Chitosan	GLYMA06G48260	Q651X6	1.12	1.42	-0.50	1.27	Cellulose synthase-like protein E6	cellulose biosynthetic process (GO:0030244);
ANE-Alginate	GLYMA11G16000	Q8LEE9	2.07	1.66	0.00	0.15	Fasciclin-like arabinogalactan protein 12	plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Alginate	GLYMA12G07430	Q8LEJ6	1.92	2.01	0.67	0.00	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Chitosan	GLYMA12G10300	O80891	1.86	-0.21	0.36	1.58	Cellulose synthase-like protein B4	cellulose biosynthetic process (GO:0030244);
ANE	GLYMA12G07450	Q8LEE9	1.30	0.61	0.00	0.00	Fasciclin-like arabinogalactan protein 12	plant-type secondary cell wall biogenesis (GO:0009834);
ANE	GLYMA14G01670	Q8VZK9	1.01	0.18	0.75	0.25	Cellulose synthase-like protein E1	cellulose biosynthetic process (GO:0030244);
ANE	GLYMA15G09250	Q8LEJ6	1.01	0.92	0.10	0.00	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE	GLYMA16G21150	Q94JQ6	1.33	0.00	0.45	0.73	Cellulose synthase A catalytic subunit 6 [UDP-forming]	cell growth (GO:0016049); cell wall organization (GO:0071555); cellulose biosynthetic process (GO:0030244);
ANE	GLYMA19G33740	Q9LDG2	1.14	0.00	0.71	0.41	Protein trichome birefringence-like 10	cell wall organization or biogenesis (GO:0071554); circadian rhythm (GO:0007623);

Table 8: ANE up-regulated genes involved in catabolic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA03G38570	H1A981	1.83	1.14	2.85	1.11	11-oxo-beta-amyrin 30-oxidase	11-oxo-beta-amyrin catabolic process (GO:1902382); glycyrrhetinate biosynthetic process (GO:1902386);
ANE-Chitin-Chitosan	GLYMA13G30463	Q9SHP6	1.49	0.00	1.28	1.18	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE-Chitin-Chitosan	GLYMA17G01990	P13917	1.06	0.06	1.19	1.15	Basic 7S globulin	protein catabolic process (GO:0030163); proteolysis (GO:0006508);
ANE-Chitin-Chitosan	GLYMA15G19580	Q8RWQ9	1.57	0.88	2.04	1.62	Thiol protease aleurain-like	No-GOBP
ANE-Chitin-Chitosan	GLYMA09G08100	Q8RWQ9	1.26	0.37	1.47	1.23	Thiol protease aleurain-like	No-GOBP
ANE-Chitin-Chitosan	GLYMA17G34900	O24325	2.95	0.00	2.70	2.32	Vacuolar-processing enzyme	No-GOBP
ANE-Chitin-Chitosan	GLYMA14G10620	O24325	2.78	0.07	2.72	1.98	Vacuolar-processing enzyme	No-GOBP
ANE-Chitin-Chitosan	GLYMA03G27500	Q8GUU2	1.39	0.03	1.07	1.01	E3 ubiquitin protein ligase RIE1	protein ubiquitination (GO:0016567);
ANE-Chitin-Chitosan	GLYMA07G08560	Q9SN27	1.79	0.00	1.52	1.25	E3 ubiquitin-protein ligase ATL59	protein ubiquitination (GO:0016567);
ANE-Chitin-Chitosan	GLYMA04G41990	P48977	1.23	0.80	2.68	2.34	Alcohol dehydrogenase	No-GOBP
ANE-Chitin	GLYMA11G16070	Q9FFZ1	1.12	0.53	1.60	0.88	Pheophytinase, chloroplastic	chlorophyll catabolic process (GO:0015996);
ANE-Chitin	GLYMA14G06470	Q8GV43	1.13	0.45	1.60	0.00	Phosphoinositide phospholipase C 6	intracellular signal transduction (GO:0035556); lipid catabolic process (GO:0016042);
ANE-Chitin	GLYMA19G43920	Q8LB81	2.28	0.22	1.70	0.60	GDSL esterase/lipase At5g33370	lipid catabolic process (GO:0016042);
ANE-Chitin	GLYMA11G10600	Q0WPR4	1.10	0.96	1.72	0.85	Serine carboxypeptidase-like 34	proteolysis involved in cellular protein catabolic process (GO:0051603);
ANE-Chitin	GLYMA06G12800	Q9FH05	1.22	0.33	1.06	0.63	Serine carboxypeptidase-like 42	proteolysis involved in cellular protein catabolic process (GO:0051603);
ANE-Chitin	GLYMA11G02980	A7VLV1	1.29	0.65	1.31	0.80	Protein STAY-GREEN, chloroplastic	regulation of chlorophyll catabolic process (GO:0010271);
ANE-Chitin	GLYMA16G23140	Q8VYV9	1.16	0.90	1.35	0.67	Aspartyl protease family protein 1	No-GOBP

ANE-Chitosan	GLYMA07G18130	A1L4Y2	1.24	0.19	0.53	1.25	Alcohol dehydrogenase-like 3	No-GOBP
ANE	GLYMA17G04911	K7MJX7	1.13	0.00	0.34	0.39	Uncharacterized protein	deoxyribonucleotide catabolic process (GO:0009264);
ANE	GLYMA11G01881	Q9FVV1	1.39	-0.59	0.01	0.08	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE	GLYMA12G32330	P10538	1.08	0.04	0.30	0.54	Beta-amylase	polysaccharide catabolic process (GO:0000272);
ANE	GLYMA14G17070	Q9SCJ9	1.03	-0.11	0.77	0.00	Ubiquitin carboxyl-terminal hydrolase 26	ubiquitin-dependent protein catabolic process (GO:0006511);
ANE	GLYMA19G34960	B5WWZ9	1.16	0.13	0.97	0.76	Long-chain-alcohol oxidase FAO2	No-GOBP
ANE	GLYMA03G01951	Q9SN27	1.03	0.00	0.58	0.22	E3 ubiquitin-protein ligase ATL59	protein ubiquitination (GO:0016567);
ANE	GLYMA01G38780	Q94A73	1.23	0.00	0.00	0.09	BTB/POZ domain-containing protein	protein ubiquitination (GO:0016567);
ANE	GLYMA17G10510	O14099	1.08	0.00	0.56	0.64	Uncharacterized RING finger protein C2F3.16	protein ubiquitination (GO:0016567);

Table 9: ANE up-regulated genes involved in transport.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA04G42960	Q9LPF1	1.18	1.30	1.36	0.03	WAT1-related protein	amino acid export (GO:0032973);
ANE-Alginate-Chitin	GLYMA06G11790	Q501F8	1.83	2.38	1.72	0.17	WAT1-related protein	amino acid export (GO:0032973); a
ANE-Alginate-Chitosan	GLYMA06G08860	Q9M817	1.97	1.84	-0.28	1.27	Protein NRT1/ PTR FAMILY 1.2	low-affinity nitrate transport (GO:0080055);
ANE-Alginate-Chitosan	GLYMA16G28910	Q9LYS2	1.98	2.01	0.00	2.90	ABC transporter C family member 10	No-GOBP
ANE-Chitin-Chitosan	GLYMA03G06420	O65100	1.10	-0.10	2.36	1.40	Ferritin-3, chloroplastic	cellular iron ion homeostasis (GO:0006879); iron ion transport (GO:0006826);
ANE-Chitin-Chitosan	GLYMA06G12840	F4I5D5	2.93	-0.07	2.90	2.83	WAT1-related protein At1g70260	transmembrane transport (GO:0055085); transport (GO:0006810);
ANE-Chitin-Chitosan	GLYMA08G04730	Q9ZU94	1.09	0.35	1.39	1.31	Cysteine-rich repeat secretory protein 12	transport (GO:0006810); viral process (GO:0016032);
ANE-Chitin-Chitosan	GLYMA12G33010	I1LUF3	1.00	0.00	1.86	1.18	Uncharacterized protein	protein transport (GO:0015031);
ANE-Chitin-Chitosan	GLYMA14G24370	Q38967	1.54	0.14	1.95	1.19	Amino acid permease 2	acidic amino acid transport (GO:0015800); neutral amino acid transport (GO:0015804);

ANE-Chitin-Chitosan	GLYMA18G02040	Q9LFT9	1.37	0.14	1.00	1.24	Ras-related protein RABH1e	protein transport (GO:0015031); small GTPase mediated signal transduction (GO:0007264);
ANE-Chitin-Chitosan	GLYMA18G20510	Q9LPV5	1.73	-0.34	2.08	2.65	High affinity nitrate transporter 2.5	nitrate assimilation (GO:0042128); transmembrane transport (GO:0055085);
ANE-Chitin-Chitosan	GLYMA18G46286	Q93ZQ5	1.64	-0.35	1.10	1.18	SPX domain-containing membrane protein	transmembrane transport (GO:0055085);
ANE-Chitin-Chitosan	GLYMA15G01390	Q9XI23	2.18	0.44	1.82	1.31	Boron transporter 4	borate transport (GO:0046713);
ANE-Chitin	GLYMA03G30550	P93051	1.22	0.00	1.08	0.67	Sugar transporter ERD6-like 7	glucose import (GO:0046323);
ANE-Chitin	GLYMA07G19060	Q94FY2	1.05	-0.96	1.77	0.19	Ferritin, chloroplastic	cellular iron ion homeostasis (GO:0006879); iron ion transport (GO:0006826);
ANE-Chitin	GLYMA08G12070	Q8W496	1.26	0.27	1.73	0.77	Protochlorophyllide-dependent translocon component 52, chloroplastic	protein transport (GO:0015031);
ANE-Chitin	GLYMA08G39140	Q9LPV5	1.22	-0.10	1.70	0.90	High affinity nitrate transporter 2.5	nitrate assimilation (GO:0042128); transmembrane transport (GO:0055085);
ANE-Chitin	GLYMA09G06620	Q9LVD4	1.26	0.00	1.22	0.73	Oxysterol-binding protein-related protein 4C	lipid transport (GO:0006869);
ANE-Chitin	GLYMA13G25890	Q9FL41	1.04	0.77	1.47	0.05	WAT1-related protein	transmembrane transport (GO:0055085); transport (GO:0006810);
ANE-Chitin	GLYMA16G08370	Q9C6W5	1.21	0.41	2.32	0.19	ABC transporter G family member 14	transmembrane transport (GO:0055085);
ANE-Chitin	GLYMA16G28340	Q9M2C0	1.07	-0.12	1.68	0.38	Vacuolar iron transporter homolog 4	ion transport (GO:0006811); iron ion homeostasis (GO:0055072);
ANE-Chitin	GLYMA17G06770	Q9SW00	1.46	0.00	1.03	0.47	Oxysterol-binding protein-related protein 4B	lipid transport (GO:0006869);
ANE-Chitin	GLYMA09G21070	Q84MA5	1.68	0.54	2.14	0.97	Cationic amino acid transporter 1	No-GOBP
ANE-Chitosan	GLYMA06G04900	Q37145	1.10	-1.10	0.52	1.25	Calcium-transporting ATPase 1, chloroplastic	No-GOBP
ANE	GLYMA01G40700	O23140	1.00	0.00	0.59	0.74	AP-2 complex subunit mu	endocytosis (GO:0006897); intracellular protein transport (GO:0006886);
ANE	GLYMA04G10050	Q8L7W1	1.03	-0.03	0.45	0.94	Mechanosensitive ion channel protein 3, chloroplastic	ion transport (GO:0006811); transmembrane transport (GO:0055085);
ANE	GLYMA06G21651	Q94JU2	2.02	0.00	0.00	0.87	WAT1-related protein	transmembrane transport (GO:0055085); transport (GO:0006810);
ANE	GLYMA09G37280	Q19KC1	1.02	0.00	0.42	0.53	Aquaporin NIP2-1	urea transport (GO:0015840);
ANE	GLYMA09G39920	Q93ZQ5	1.33	-0.07	0.83	-0.13	SPX domain-containing membrane protein	transmembrane transport (GO:0055085);
ANE	GLYMA10G34820	I1LCT5	1.11	0.38	0.98	0.42	Uncharacterized protein	transmembrane transport (GO:0055085);

ANE	GLYMA10G37610	F4I4K7	1.15	0.17	0.68	0.75	Ferric reduction oxidase 3, mitochondrial	ion transport (GO:0006811);
ANE	GLYMA11G35610	Q948P6	1.03	0.01	0.71	0.03	Ferritin-3, chloroplastic	cellular iron ion homeostasis (GO:0006879); iron ion transport (GO:0006826);
ANE	GLYMA13G28440	Q8LBI9	1.72	0.00	0.95	0.90	Sugar transporter ERD6-like 16	carbohydrate transport (GO:0008643);
ANE	GLYMA13G42121	K7M3K5	1.22	0.00	0.20	0.00	Uncharacterized protein	vesicle-mediated transport (GO:0016192);
ANE	GLYMA15G27750	Q9LH79	1.05	0.00	0.74	0.91	Bidirectional sugar transporter SWEET2	carbohydrate transmembrane transport (GO:0034219);
ANE	GLYMA15G34653	Q9FF81	2.03	0.83	0.00	0.42	Vacuolar protein sorting-associated protein 36	protein transport (GO:0015031);
ANE	GLYMA15G37621	O04619	3.92	0.00	0.00	0.00	Mitochondrial adenine nucleotide transporter ADNT1	transmembrane transport (GO:0055085);
ANE	GLYMA20G25520	P82353	1.03	0.00	0.84	0.00	Non-specific lipid-transfer protein 2	transport (GO:0006810);
ANE	GLYMA10G03460	Q94LW6	1.02	0.66	0.79	-0.10	Probable sulfate transporter 3.5	No-GOBP
ANE	GLYMA16G28900	Q9LYS2	1.13	0.24	0.00	0.94	ABC transporter C family member 10	No-GOBP
ANE	GLYMA19G39810	Q9LZJ5	1.03	0.66	0.79	0.32	ABC transporter C family member 14	No-GOBP
ANE	GLYMA18G49470	Q9LQL2	1.11	0.44	0.00	0.00	Protein NRT1/ PTR FAMILY 7.3	nitrate transport (GO:0015706);
ANE	GLYMA15G00620	Q9SAI4	1.01	0.17	0.96	0.37	Aquaporin NIP6-1	borate transmembrane transport (GO:0035445);
ANE	GLYMA13G41460	Q9SYD6	1.17	0.11	0.90	0.61	Protein DETOXIFICATION 42	citrate transport (GO:0015746); response to aluminum ion (GO:0010044);
ANE	GLYMA10G05420	Q9ZUT3	1.15	-0.05	0.00	0.60	Protein ALUMINUM SENSITIVE 3	ion transmembrane transport (GO:0034220); response to aluminum ion (GO:0010044);

Table 10: ANE up-regulated genes involved in starch metabolism.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA17G13830	Q9FKS0	1.25	0.47	2.15	1.79	Uridine kinase-like protein 1, chloroplastic	regulation of starch metabolic process (GO:2000904); UMP salvage (GO:0044206);
ANE-Chitin-Chitosan	GLYMA20G36040	P0C585	1.70	0.00	1.20	1.83	Granule-bound starch synthase 1, chloroplastic/amyloplastic	starch biosynthetic process (GO:0019252);
ANE-Chitin	GLYMA01G41190	O23553	1.75	0.21	2.17	0.60	Beta-amylase 3, chloroplastic	maltose biosynthetic process (GO:0000024); response to cold (GO:0009409); starch catabolic process (GO:0005983);

ANE	GLYMA06G09880	Q8H103	1.18	0.00	0.65	0.56	Glucose-6-phosphate isomerase 1, chloroplastic	positive regulation of flower development (GO:0009911); starch metabolic process (GO:0005982);
ANE	GLYMA11G12410	Q9SIK1	1.27	0.00	0.00	0.00	Probable glucose-1-phosphate adenylyltransferase large subunit, chloroplastic	starch biosynthetic process (GO:0019252);

Table 11: ANE up-regulated genes involved in photosynthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA02G37710	Q94CE3	1.94	0.00	2.14	1.24	Beta carbonic anhydrase 5, chloroplastic	carbon utilization (GO:0015976);
ANE-Chitin-Chitosan	GLYMA17G02371	Q39734	2.30	0.20	2.07	1.76	Pyruvate, phosphate dikinase, chloroplastic	photosynthesis (GO:0015979); pyruvate metabolic process (GO:0006090);
ANE-Chitin	GLYMA07G38390	Q39734	1.14	0.31	1.14	0.00	Pyruvate, phosphate dikinase, chloroplastic	photosynthesis (GO:0015979); pyruvate metabolic process (GO:0006090);
ANE-Chitin	GLYMA10G12410	F4J3G5	1.04	0.02	1.15	0.30	Carboxyl-terminal-processing peptidase 3, chloroplastic;	photosynthetic water oxidation

Table 12: ANE up-regulated genes encoding various receptors.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitosan	GLYMA16G28740	C0LGQ5	1.02	1.17	0.54	1.74	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE-Alginate-Chitosan	GLYMA16G28850	C0LGQ5	1.15	1.30	0.31	1.04	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE-Alginate	GLYMA03G22510	Q39202	1.07	1.48	0.97	0.93	G-type lectin S-receptor-like serine/threonine-protein kinase RLK1	No-GOBP
ANE-Chitin-Chitosan	GLYMA09G02205	Q9LT96	1.50	0.92	1.53	1.60	Probable leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE-Chitin-Chitosan	GLYMA13G09445	Q9LMN7	1.10	0.81	1.02	1.67	Wall-associated receptor kinase 5	No-GOBP
ANE-Chitin	GLYMA18G02850	Q9LYN6	1.13	0.50	1.10	0.66	Probable inactive receptor-like protein kinase	No-GOBP

ANE-Chitin	GLYMA10G08010	Q9LT96	1.14	0.36	1.51	0.49	Probable leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE-Chitin	GLYMA12G36241	Q9C9H7	1.19	0.64	1.23	0.96	Receptor-like protein 12	No-GOBP
ANE-Chitin	GLYMA06G46130	Q9SW97	1.11	0.07	1.06	0.85	Glutamate receptor 3.5	cellular response to amino acid stimulus (GO:0071230);
ANE	GLYMA11G32315	C0LGH3	1.27	0.00	0.64	0.67	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE	GLYMA20G37470	Q8RXC8	1.43	0.00	0.00	0.45	Receptor-like cytosolic serine/threonine-protein kinase RBK2	No-GOBP
ANE	GLYMA12G36220	Q9SHI3	1.06	0.09	0.71	0.38	Receptor-like protein 2	No-GOBP
ANE	GLYMA11G13120	O80462	1.54	0.00	0.78	0.00	Extra-large guanine nucleotide-binding protein 1	G-protein coupled receptor signaling pathway (GO:0007186);

Table 13: ANE up-regulated genes involved in transcriptional regulation.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA19G00730	Q58FS3	1.96	1.21	3.97	2.05	Transcription factor RADIALIS	Involved in the dorsoventral asymmetry of flowers.
ANE-Alginate-Chitin	GLYMA04G33270	Q9FLJ2	1.79	1.10	2.94	0.96	NAC domain-containing protein 100	transcription, DNA-templated (GO:0006351);
ANE-Alginate-Chitosan	GLYMA01G40620	Q9T072	1.77	2.09	0.38	1.84	Transcription factor bHLH25	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin-Chitosan	GLYMA05G01285	Q9C9F0	1.25	0.36	1.41	1.04	Probable WRKY transcription factor 9	Elicitor induced responses
ANE-Chitin-Chitosan	GLYMA10G42750	Q700D9	1.58	0.52	1.95	1.13	Putative Myb family transcription factor	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin-Chitosan	GLYMA15G12570	O23310	1.25	0.27	1.01	1.81	Nuclear transcription factor Y Subunit B-3	Promote the transcription of genes contain CCAAT motif
ANE-Chitin-Chitosan	GLYMA15G42680	Q66GR3	1.41	0.00	1.52	1.02	Transcription factor bHLH130	photoperiodism, flowering (GO:0048573);
ANE-Chitin-Chitosan	GLYMA19G34210	Q9LUH8	1.96	0.40	1.23	1.57	Heat stress transcription factor A-6b	transcription, DNA-templated (GO:0006351);
ANE-Chitin-Chitosan	GLYMA20G29250	Q6EU39	1.97	1.00	2.07	1.39	MADS-box transcription factor 6	meristem determinacy (GO:0010022); specification of floral organ identity (GO:0010093);

ANE-Chitin-Chitosan	GLYMA08G02020	Q9SW80	1.22	0.00	1.36	1.06	BEL1-like homeodomain protein 2	leaf morphogenesis (GO:0009965);
ANE-Chitin-Chitosan	GLYMA12G22790	Q9LD44	1.55	0.43	1.53	1.17	NAC transcription factor 56	regulation of embryonic development (GO:0045995); response to jasmonic acid (GO:0009753
ANE-Chitin-Chitosan	GLYMA13G35560	Q9LD44	2.12	0.59	2.26	1.64	NAC transcription factor 56	regulation of embryonic development (GO:0045995); response to jasmonic acid (GO:0009753);
ANE-Chitin-Chitosan	GLYMA12G04440	Q9LZP8	1.28	0.69	1.47	1.63	bZIP transcription factor 53	cellular response to starvation (GO:0009267); hypotonic response (GO:0006971); Activated by low energy stress;
ANE-Chitin	GLYMA06G13100	Q9ZNX9	1.19	0.15	1.21	0.44	RNA polymerase sigma factor sigE, chloroplastic/mitochondrial	chloroplast organization (GO:0009658); photosystem II assembly (GO:0010207);
ANE-Chitin	GLYMA12G13430	Q5Z5I4	1.13	0.00	1.19	0.68	Probable transcription factor GLK1	positive regulation of organelle organization (GO:0010638);
ANE-Chitin	GLYMA01G05810	Q9LXG0	1.18	0.46	1.50	0.36	Zinc-finger homeodomain protein 8	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin	GLYMA03G42221	Q6LA42	1.38	0.88	1.57	0.72	Two-component response regulator-like APRR5	rhythmic process (GO:0048511); Involved in the inhibition of leaf expansion;
ANE-Chitin	GLYMA05G04040	Q8VYJ2	1.24	0.00	1.26	0.85	AT-hook motif nuclear-localized protein 1	binds AT-rich DNA sequences;
ANE-Chitin	GLYMA11G13390	O22864	1.95	0.90	1.93	0.94	Protein NLP8	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin	GLYMA17G10970	Q9FLJ2	1.17	0.10	1.59	0.00	NAC domain-containing protein 100	transcription, DNA-templated (GO:0006351);
ANE-Chitin	GLYMA10G07940	Q9SGQ0	1.19	0.14	1.40	0.91	Transcription factor VOZ1	cold acclimation (GO:0009631); defense response to bacterium, incompatible interaction (GO:0009816); long-day photoperiodism, flowering (GO:0048574);
ANE-Chitosan	GLYMA03G25100	Q1PF16	1.96	0.53	0.60	1.83	Transcription factor bHLH19	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitosan	GLYMA07G03100	A0A0B2NW04	1.51	0.68	-0.20	1.85	Transcription factor bHLH25	No-GOBP
ANE-Chitosan	GLYMA08G23020	Q1PF16	1.36	0.00	-0.36	1.38	Transcription factor bHLH19	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitosan	GLYMA08G23035	Q1PF16	1.21	0.00	0.00	1.27	Transcription factor bHLH19	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitosan	GLYMA03G39220	I1JR72	1.13	0.00	0.94	1.11	Uncharacterized protein	cell differentiation (GO:0030154); positive regulation of transcription from RNA polymerase II promoter (GO:0045944);
ANE	GLYMA05G36290	Q8S9H7	1.13	0.00	0.13	0.00	Transcription factor DIVARICATA	flower development (GO:0009908);

ANE	GLYMA07G04050	Q93ZH2	1.28	0.24	0.86	0.25	Nuclear transcription factor Y Subunit A-3	binding to a CCAAT motif in promoters;
ANE	GLYMA07G10311	Q8GZ13	2.34	0.00	0.00	0.00	Transcription factor BEE 1	Positive regulation of shade avoidance (GO: 1902448); Positive regulator of brassinosteroid signaling.
ANE	GLYMA15G29630	Q9LT23	1.13	-0.44	0.15	0.00	Transcription factor bHLH121	transcription, DNA-templated (GO:0006351);
ANE	GLYMA20G33890	Q9SVQ0	1.41	0.63	0.43	0.22	Ethylene-responsive transcription factor ERF062	Binds to the GCC-box pathogenesis-related promoter element.
ANE	GLYMA04G02960	Q9LQZ7	1.25	0.00	0.00	0.66	B-box zinc finger protein 21	photomorphogenesis (GO:0009640); shade avoidance (GO:0009641);
ANE	GLYMA08G40420	O48686	1.26	-0.38	0.53	0.00	Paired amphipathic helix protein Sin3-like 3	Interacts with ERF7 to repress genes in abscisic acid and drought stress responses.
ANE	GLYMA11G20240	Q9SW80	1.05	0.08	0.55	0.03	BEL1-like homeodomain protein 2	leaf morphogenesis (GO:0009965);
ANE	GLYMA13G07720	Q9FIM0	1.20	0.00	0.47	0.75	Agamous-like MADS-box protein AGL82	Maintenance of the proper function of the central cell in pollen tube attraction.
ANE	GLYMA14G10370	Q8GRL4	1.30	0.00	0.15	0.00	Homeobox-leucine zipper protein HOX19	transcription, DNA-templated (GO:0006351);
ANE	GLYMA15G00880	Q0WV90	1.14	-0.05	0.58	0.53	Topless-related protein 1	Activates TIR-NB-LRR R protein-mediated immune responses through repression of negative regulators such as CNGC2/DND1.
ANE	GLYMA16G05751	Q8S4W7	1.30	-0.43	0.07	0.03	DELLA protein GAI1	gibberellic acid mediated signaling pathway (GO:0009740);
ANE	GLYMA18G32370	Q9M2Y9	1.08	0.00	0.00	0.00	Transcription factor RAX3	Positively regulates axillary meristems (AMs) formation and development, especially during inflorescence.
ANE	GLYMA04G41691	Q9ZNX9	1.24	-0.37	0.54	0.00	RNA polymerase sigma factor sigE, chloroplastic/mitochondrial	chloroplast organization (GO:0009658); photosystem II assembly (GO:0010207);

Table 14: ANE up-regulated genes involved in phytohormone related processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA03G06580	Q9M020	1.14	1.27	1.69	1.62	Lectin-domain containing receptor kinase VI.3	abscisic acid-activated signaling pathway (GO:0009738); seed germination (GO:0009845);

ANE-Alginate-Chitin-Chitosan	GLYMA16G02390	P46897	1.81	1.00	1.83	1.40	Homeobox-leucine zipper protein ATHB-7	response to abscisic acid (GO:0009737); response to water deprivation (GO:0009414);
ANE-Alginate-Chitin-Chitosan	GLYMA09G30716	Q9LVH4	2.30	1.59	1.82	1.71	Protein C2-DOMAIN ABA-RELATED 4	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952);
ANE-Chitin-Chitosan	GLYMA13G18910	Q38829	1.00	0.29	1.29	1.29	Auxin-responsive protein IAA11	auxin-activated signaling pathway (GO:0009734);
ANE-Chitin-Chitosan	GLYMA13G43800	P93830	1.34	0.57	1.90	1.42	Auxin-responsive protein IAA17	auxin-activated signaling pathway (GO:0009734); positive regulation of leaf senescence (GO:1900057); response to auxin (GO:0009733);
ANE-Chitin-Chitosan	GLYMA15G01550	Q5VRD1	1.35	0.47	1.95	1.09	Auxin-responsive protein IAA1	auxin-activated signaling pathway (GO:0009734); response to auxin (GO:0009733);
ANE-Chitin-Chitosan	GLYMA13G43810	A0A0B2SIQ2	1.17	0.00	1.44	1.38	Auxin-induced protein	auxin-activated signaling pathway (GO:0009734);
ANE-Chitin-Chitosan	GLYMA19G03010	Q9SYK9	1.85	0.55	2.01	1.49	UDP-glycosyltransferase 74E2	cellular hyperosmotic salinity response (GO:0071475); cellular response to abscisic acid stimulus (GO:0071215); flavonoid biosynthetic process (GO:0009813);
ANE-Chitin-Chitosan	GLYMA07G05800	P46897	1.22	0.00	1.54	1.27	Homeobox-leucine zipper protein ATHB-7	response to abscisic acid (GO:0009737); response to water deprivation (GO:0009414);
ANE-Chitin-Chitosan	GLYMA02G17010	Q9LYC1	1.15	0.55	1.45	1.28	Gibberellin receptor GID1B	floral organ morphogenesis (GO:0048444); response to gibberellin (GO:0009739);
ANE-Chitin	GLYMA04G18730	Q64FQ2	1.50	0.38	1.06	0.82	Protein kinase PINOID 2	auxin-activated signaling pathway (GO:0009734
ANE-Chitin	GLYMA01G40090	Q9FJX5	1.73	0.59	1.68	0.98	BTB/POZ and TAZ domain-containing protein 4	response to auxin (GO:0009733); response to wounding (GO:0009611);
ANE-Chitin	GLYMA17G32051	Q9FY48	1.33	0.16	1.34	0.49	E3 ubiquitin-protein ligase KEG	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952);
ANE-Chitin	GLYMA08G20030	Q38898	1.26	0.59	1.84	0.63	Potassium channel AKT2/3	regulation of membrane potential (GO:0042391); response to abscisic acid (GO:0009737);
ANE-Chitin	GLYMA09G41960	O81077	1.46	0.34	1.45	0.66	Abscisic acid 8'-hydroxylase 2	abscisic acid catabolic process (GO:0046345);
ANE-Chitin	GLYMA15G03930	Q9SFB0	1.09	0.00	1.18	0.66	Protein DETOXIFICATION 43	citrate transport (GO:0015746); drug transmembrane transport (GO:0006855); metal ion transport (GO:0030001); response to abscisic acid (GO:0009737);
ANE-Chitin	GLYMA15G10070	Q9XG83	1.13	0.44	1.05	0.59	Gibberellin 2-beta-dioxygenase	gibberellin biosynthetic process (GO:0009686);
ANE-Chitin	GLYMA13G33300	Q9SQ80	2.70	0.00	1.27	0.87	Gibberellin 2-beta-dioxygenase 1	gibberellin biosynthetic process (GO:0009686);

ANE-Chitin	GLYMA12G34175	O49561	1.56	0.68	1.80	0.69	Gibberellin 2-beta-dioxygenase 8	gibberellin biosynthetic process (GO:0009686);
ANE-Chitosan	GLYMA13G33290	Q9XG83	2.02	0.00	0.58	1.22	Gibberellin 2-beta-dioxygenase	gibberellin biosynthetic process (GO:0009686);
ANE-Chitosan	GLYMA07G34480	Q9C6B7	1.37	0.00	0.60	1.90	Protein C2-DOMAIN ABA-RELATED 3	abscisic acid-activated signaling pathway (GO:0009738);
ANE	GLYMA20G29770	Q04980	1.01	0.27	0.09	0.67	Low-temperature-induced 65 kDa protein	abscisic acid-activated signaling pathway (GO:0009738);
ANE	GLYMA14G14320	Q9FY48	1.33	0.00	0.95	0.85	E3 ubiquitin-protein ligase KEG	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952);
ANE	GLYMA04G11230	Q9MAH1	1.00	0.00	0.31	0.13	TPR repeat-containing thioredoxin TTL1	abscisic acid-activated signaling pathway (GO:0009738); cell redox homeostasis (GO:0045454); response to salt stress (GO:0009651);
ANE	GLYMA10G43030	O81360	1.24	-0.17	0.78	0.12	Zeaxanthin epoxidase, chloroplastic	abscisic acid biosynthetic process (GO:0009688);
ANE	GLYMA08G18470	Q84TE6	1.13	0.43	0.52	0.24	NAC domain-containing protein 21/22	auxin-activated signaling pathway (GO:0009734); lateral root development (GO:0048527);
ANE	GLYMA06G37401	Q6I581	1.73	0.00	0.00	0.00	Jasmonic acid-amido synthetase JAR1	induced systemic resistance, jasmonic acid mediated signaling pathway (GO:0009864); response to auxin (GO:0009733);
ANE	GLYMA15G00885	Q94AI7	1.28	-0.20	0.55	0.75	Protein TOPLESS	response to auxin (GO:0009733);
ANE	GLYMA04G02750	Q84M85	1.04	0.00	0.99	0.38	Probable cytokinin riboside 5'-monophosphate phosphoribohydrolase LOGL5	cytokinin biosynthetic process (GO:0009691);

Table 15: ANE up-regulated genes encoding SOD.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA20G33880	P28759	1.40	1.42	1.30	1.09	Superoxide dismutase [Fe], chloroplastic	Destroys superoxide anion radicals which are normally produced within the cells and which are toxic to biological systems.
ANE-Alginate-Chitin	GLYMA10G33710	P28759	1.63	1.38	1.42	0.99	Superoxide dismutase [Fe], chloroplastic	Destroys superoxide anion radicals which are normally produced within the cells and which are toxic to biological systems.

Appendix 2

ANE down-regulated gene function annotations

Table 16: ANE down-regulated genes involved in ribosomal protein synthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA01G03570	P47905	-1.28	0.12	-1.31	-1.17	Ubiquitin-40S ribosomal protein S27a	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA01G37250	Q42064	-1.27	-0.28	-1.32	-1.20	60S ribosomal protein L8-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA01G41620	O65050	-1.54	-0.37	-1.59	-1.28	60S ribosomal protein L15-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA02G05370	P46299	-1.78	0.00	-1.50	-1.11	40S ribosomal protein S4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA02G05540	Q42064	-1.29	-0.21	-1.26	-1.21	60S ribosomal protein L8-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA02G09370	Q9SCM3	-1.29	0.00	-1.12	-1.19	40S ribosomal protein S2-4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA02G43080	P49204	-1.72	-0.17	-1.97	-1.41	40S ribosomal protein S4-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA03G21710	O81361	-1.57	0.09	-1.41	-1.17	40S ribosomal protein S8	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA03G29801	P49200	-1.51	0.26	-1.21	-1.00	40S ribosomal protein S20-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA03G29810	P49200	-1.69	-0.01	-1.51	-1.27	40S ribosomal protein S20-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA03G37340	P42791	-1.61	0.00	-1.53	-1.24	60S ribosomal protein L18-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA04G40470	P30707	-1.28	0.06	-1.43	-2.00	60S ribosomal protein L9	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA05G01180	Q9FLF0	-1.45	0.00	-1.26	-1.34	40S ribosomal protein S9-2	translation (GO:0006412);

ANE-Chitin-Chitosan	GLYMA05G02570	P49637	-1.74	0.02	-1.56	-1.27	60S ribosomal protein L27a-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA05G04870	O23515	-1.39	-0.32	-1.56	-1.10	60S ribosomal protein L15-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA05G34350	P46293	-1.09	-0.55	-1.48	-1.72	40S ribosomal protein S16	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA06G14350	P30707	-1.44	0.00	-1.42	-1.32	60S ribosomal protein L9	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA07G01540	Q9M0E2	-1.62	-0.06	-1.62	-1.22	60S ribosomal protein L28-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA07G06590	Q6UNT2	-1.61	0.11	-1.50	-1.02	60S ribosomal protein L5	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA07G34440	Q9LZ17	-1.28	-0.32	-1.09	-1.26	40S ribosomal protein S17-4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA08G05290	P46293	-1.38	-0.79	-1.57	-1.44	40S ribosomal protein S16	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA08G09230	Q05462	-1.34	-0.24	-1.30	-1.11	60S ribosomal protein L27	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA08G20960	Q9M0E2	-1.59	0.00	-1.61	-1.36	60S ribosomal protein L28-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA08G43690	O81361	-1.36	-0.01	-1.27	-1.13	40S ribosomal protein S8	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA09G02790	Q9FKC0	-1.68	0.08	-1.49	-1.35	60S ribosomal protein L13a-4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA09G03660	P29344	-1.51	-0.24	-1.59	-2.32	30S ribosomal protein S1, chloroplastic	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA10G02060	P49693	-1.49	-0.15	-1.40	-1.33	60S ribosomal protein L19-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA10G05580	Q9SF40	-1.45	0.10	-1.22	-1.61	60S ribosomal protein L4-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA10G29170	P42791	-1.42	-0.13	-1.41	-1.06	60S ribosomal protein L18-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA10G39050	Q8GTE3	-1.60	0.34	-1.40	-1.19	40S ribosomal protein S3a	translation (GO:0006412);

ANE-Chitin-Chitosan	GLYMA11G00590	P42798	-1.93	0.00	-1.84	-1.08	40S ribosomal protein S15a-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA11G00600	P42798	-1.77	-0.08	-1.78	-1.26	40S ribosomal protein S15a-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA11G03755	O82528	-1.47	0.00	-1.14	-1.10	60S ribosomal protein L15	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA12G07160	O22644	-1.35	0.13	-1.40	-1.06	60S ribosomal protein L23A	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G18830	Q07760	-1.64	0.18	-1.35	-1.13	60S ribosomal protein L23	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G19930	Q9SF40	-1.46	0.22	-1.28	-1.46	60S ribosomal protein L4-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G31130	Q9M3V8	-1.60	0.21	-1.35	-1.25	40S ribosomal protein S6	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G31650	P51414	-1.43	-0.40	-1.20	-1.02	60S ribosomal protein L26-1	response to cold (GO:0009409); translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G31900	Q9FJX2	-1.52	-0.33	-1.28	-1.11	60S ribosomal protein L26-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA13G42830	Q9M0E2	-1.45	0.00	-1.53	-1.19	60S ribosomal protein L28-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA14G06170	P46299	-1.30	-0.22	-1.47	-1.25	40S ribosomal protein S4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA15G08210	Q9M3V8	-1.63	0.23	-1.23	-1.09	40S ribosomal protein S6	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA15G13650	P47905	-1.50	0.32	-1.33	-1.07	Ubiquitin-40S ribosomal protein S27a	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA15G14550	P29344	-1.14	-0.16	-1.34	-2.03	30S ribosomal protein S1, chloroplastic	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA15G42620	Q9FZ76	-1.55	0.09	-1.47	-1.16	60S ribosomal protein L6-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA16G08090	P41127	-1.49	0.00	-1.51	-1.13	60S ribosomal protein L13-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA16G10700	O81361	-1.45	0.06	-1.19	-1.12	40S ribosomal protein S8	translation (GO:0006412);

ANE-Chitin-Chitosan	GLYMA16G23730	P46299	-1.21	0.00	-1.11	-1.18	40S ribosomal protein S4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA16G24120	Q42064	-1.49	-0.36	-1.55	-1.43	60S ribosomal protein L8-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA17G10710	Q9FLF0	-1.66	0.00	-1.48	-1.23	40S ribosomal protein S9-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA17G15230	O23515	-1.45	-0.21	-1.44	-1.17	60S ribosomal protein L15-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA18G01110	P35266	-1.51	0.08	-1.49	-1.06	60S ribosomal protein L17-1	hematopoietic stem cell homeostasis (GO:0061484); translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA18G02970	Q8VZB9	-1.50	0.47	-1.35	-1.01	60S ribosomal protein L10a-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA19G32690	P49200	-1.30	-0.09	-1.43	-1.07	40S ribosomal protein S20-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA19G39240	Q43291	-1.28	0.00	-1.22	-1.06	60S ribosomal protein L21-1	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA19G39940	Q940B0	-1.73	0.00	-1.65	-1.29	60S ribosomal protein L18-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA19G43190	Q8LC83	-1.62	0.19	-1.49	-1.01	40S ribosomal protein S24-2	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA20G02170	Q9LZ17	-1.27	-0.21	-1.30	-1.31	40S ribosomal protein S17-4	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA20G26871	Q5I7K3	-1.17	-0.11	-1.42	-1.11	40S ribosomal protein S29	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA20G28780	Q8GTE3	-1.66	0.04	-1.58	-1.56	40S ribosomal protein S3a	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA20G38130	Q940B0	-1.54	-0.31	-1.67	-1.11	60S ribosomal protein L18-3	translation (GO:0006412);
ANE-Chitin-Chitosan	GLYMA20G38480	Q9M5L0	-1.39	0.20	-1.20	-1.03	60S ribosomal protein L35	translation (GO:0006412);
ANE-Chitin	GLYMA01G00740	Q9M573	-1.22	0.00	-1.32	-0.66	60S ribosomal protein L31	translation (GO:0006412);
ANE-Chitin	GLYMA01G03180	P49637	-1.33	0.00	-1.29	-0.67	60S ribosomal protein L27a-3	translation (GO:0006412);
ANE-Chitin	GLYMA01G31270	P46299	-1.27	0.32	-1.13	-0.76	40S ribosomal protein S4	translation (GO:0006412);
ANE-Chitin	GLYMA01G44700	Q8GTE3	-1.50	0.07	-1.36	-0.98	40S ribosomal protein S3a	translation (GO:0006412);

ANE-Chitin	GLYMA01G45060	P42798	-1.47	0.08	-1.19	-0.82	40S ribosomal protein S15a-1	translation (GO:0006412);
ANE-Chitin	GLYMA02G00490	O49884	-1.11	0.21	-1.14	-0.59	60S ribosomal protein L30	translation (GO:0006412);
ANE-Chitin	GLYMA02G00680	Q9T043	-1.26	0.05	-1.24	-0.59	60S ribosomal protein L14-2	translation (GO:0006412);
ANE-Chitin	GLYMA02G01950	P49693	-1.35	0.07	-1.16	-0.76	60S ribosomal protein L19-3	translation (GO:0006412);
ANE-Chitin	GLYMA02G02140	Q9M9W1	-1.30	0.08	-1.07	-0.64	60S ribosomal protein L22-2	translation (GO:0006412);
ANE-Chitin	GLYMA02G04090	P47905	-1.27	0.28	-1.22	-0.96	Ubiquitin-40S ribosomal protein S27a	translation (GO:0006412);
ANE-Chitin	GLYMA02G40660	Q9LYK9	-1.29	0.00	-1.29	-0.65	40S ribosomal protein S26-3	translation (GO:0006412);
ANE-Chitin	GLYMA02G42081	Q9LMK0	-1.85	-0.07	-1.78	-0.94	60S ribosomal protein L35a-1	translation (GO:0006412);
ANE-Chitin	GLYMA02G45430	P51418	-1.33	0.00	-1.45	-0.99	60S ribosomal protein L18a-2	translation (GO:0006412);
ANE-Chitin	GLYMA03G33460	Q9XF97	-1.23	0.01	-1.16	-0.75	60S ribosomal protein L4	translation (GO:0006412);
ANE-Chitin	GLYMA03G33530	Q9M5Z9	-1.22	0.00	-1.38	-0.94	40S ribosomal protein S23	translation (GO:0006412);
ANE-Chitin	GLYMA03G36420	P46302	-1.32	0.19	-1.21	-0.54	40S ribosomal protein S28	translation (GO:0006412);
ANE-Chitin	GLYMA03G36560	Q9FDZ9	-1.52	0.23	-1.40	-0.96	60S ribosomal protein L21-2	translation (GO:0006412);
ANE-Chitin	GLYMA03G39420	Q940B0	-1.33	0.00	-1.16	-0.59	60S ribosomal protein L18-3	translation (GO:0006412);
ANE-Chitin	GLYMA03G39480	Q9M9W1	-1.40	0.34	-1.12	-0.50	60S ribosomal protein L22-2	translation (GO:0006412);
ANE-Chitin	GLYMA03G40110	O22584	-1.28	0.37	-1.12	-0.54	40S ribosomal protein S14	translation (GO:0006412);
ANE-Chitin	GLYMA03G40130	O22584	-1.25	0.33	-1.15	-0.76	40S ribosomal protein S14	translation (GO:0006412);
ANE-Chitin	GLYMA04G06700	P51425	-1.37	0.14	-1.50	-0.96	60S ribosomal protein L39	translation (GO:0006412);
ANE-Chitin	GLYMA04G12320	P46287	-1.15	0.00	-1.39	-0.98	60S ribosomal protein L11	translation (GO:0006412);
ANE-Chitin	GLYMA04G16660	Q9SGA6	-1.66	0.37	-1.41	-0.79	40S ribosomal protein S19-1	translation (GO:0006412);
ANE-Chitin	GLYMA04G36140	P49637	-1.27	0.00	-1.07	-0.83	60S ribosomal protein L27a-3	translation (GO:0006412);
ANE-Chitin	GLYMA05G04670	P49689	-1.12	-0.03	-1.41	-0.52	40S ribosomal protein S30	translation (GO:0006412);
ANE-Chitin	GLYMA05G26290	Q9C912	-1.68	0.38	-1.34	-0.83	60S ribosomal protein L35a-3	translation (GO:0006412);
ANE-Chitin	GLYMA05G27940	O48557	-1.61	0.17	-1.39	-0.98	60S ribosomal protein L17	translation (GO:0006412);
ANE-Chitin	GLYMA05G31760	Q9FZ76	-1.22	0.00	-1.11	-0.77	60S ribosomal protein L6-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin	GLYMA05G34570	Q9SCM3	-1.42	0.20	-1.01	-0.92	40S ribosomal protein S2-4	translation (GO:0006412);
ANE-Chitin	GLYMA05G34680	Q9ZNS1	-1.22	0.27	-1.13	-0.76	40S ribosomal protein S7	translation (GO:0006412);

ANE-Chitin	GLYMA06G48130	P46287	-1.17	-0.20	-1.50	-0.68	60S ribosomal protein L11	translation (GO:0006412);
ANE-Chitin	GLYMA07G15320	Q9M573	-1.31	0.01	-1.08	-0.74	60S ribosomal protein L31	translation (GO:0006412);
ANE-Chitin	GLYMA07G31840	Q9M3V8	-1.39	0.00	-1.44	-0.89	40S ribosomal protein S6	translation (GO:0006412);
ANE-Chitin	GLYMA07G37060	Q9M7X7	-1.02	0.38	-1.13	-0.49	60S ribosomal protein L29-1	translation (GO:0006412);
ANE-Chitin	GLYMA07G38520	P42798	-1.40	0.19	-1.12	-0.22	40S ribosomal protein S15a-1	translation (GO:0006412);
ANE-Chitin	GLYMA07G39980	P31674	-1.31	0.05	-1.04	-0.81	40S ribosomal protein S15	translation (GO:0006412);
ANE-Chitin	GLYMA08G03480	P46290	-1.44	0.11	-1.25	-0.59	60S ribosomal protein L31	translation (GO:0006412);
ANE-Chitin	GLYMA08G04690	Q945U1	-1.24	0.42	-1.18	-0.54	40S ribosomal protein S15	translation (GO:0006412);
ANE-Chitin	GLYMA08G04990	Q9ZNS1	-1.40	0.06	-1.36	-0.84	40S ribosomal protein S7	translation (GO:0006412);
ANE-Chitin	GLYMA08G08030	Q96499	-1.08	0.06	-1.07	-0.44	60S ribosomal protein L44	translation (GO:0006412);
ANE-Chitin	GLYMA08G10910	O48557	-1.57	0.09	-1.41	-0.79	60S ribosomal protein L17	translation (GO:0006412);
ANE-Chitin	GLYMA08G14995	Q9FZ76	-1.18	0.00	-1.06	-0.82	60S ribosomal protein L6-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin	GLYMA08G21960	Q8LEM8	-1.05	-0.12	-1.27	-0.79	60S ribosomal protein L37-3	translation (GO:0006412);
ANE-Chitin	GLYMA08G23260	Q9XHS0	-1.21	0.17	-1.07	-0.80	40S ribosomal protein S12	translation (GO:0006412);
ANE-Chitin	GLYMA08G28800	O49884	-1.20	0.40	-1.08	-0.47	60S ribosomal protein L30	translation (GO:0006412);
ANE-Chitin	GLYMA08G46860	Q9FKC0	-1.46	0.23	-1.04	-0.86	60S ribosomal protein L13a-4	translation (GO:0006412);
ANE-Chitin	GLYMA09G05720	Q05462	-1.64	0.47	-1.17	-0.83	60S ribosomal protein L27	translation (GO:0006412);
ANE-Chitin	GLYMA09G12500	Q5SMI4	-1.14	0.14	-1.07	-0.50	60S ribosomal protein L39-3	translation (GO:0006412);
ANE-Chitin	GLYMA09G31560	Q9C514	-1.17	0.28	-1.03	-0.52	40S ribosomal protein S7-1	translation (GO:0006412);
ANE-Chitin	GLYMA09G34760	P41127	-1.38	0.12	-1.22	-0.92	60S ribosomal protein L13-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin	GLYMA10G00500	O49884	-1.36	0.22	-1.33	-0.56	60S ribosomal protein L30	translation (GO:0006412);
ANE-Chitin	GLYMA10G00680	Q9T043	-1.20	0.31	-1.04	-0.49	60S ribosomal protein L14-2	translation (GO:0006412);
ANE-Chitin	GLYMA10G02270	Q9M9W1	-1.29	0.33	-1.04	-0.45	60S ribosomal protein L22-2	translation (GO:0006412);
ANE-Chitin	GLYMA10G04560	P49690	-1.54	0.04	-1.23	-0.84	60S ribosomal protein L23	translation (GO:0006412);
ANE-Chitin	GLYMA10G05270	Q9M5L0	-1.11	-0.87	-1.53	-0.26	60S ribosomal protein L35	translation (GO:0006412);
ANE-Chitin	GLYMA10G05660	P46297	-1.31	0.21	-1.25	-0.90	40S ribosomal protein S23	translation (GO:0006412);
ANE-Chitin	GLYMA10G06040	O50003	-1.55	0.16	-1.43	-0.78	60S ribosomal protein L12	translation (GO:0006412);

ANE-Chitin	GLYMA10G29200	Q9M9W1	-1.23	0.00	-1.08	-0.76	60S ribosomal protein L22-2	translation (GO:0006412);
ANE-Chitin	GLYMA10G30200	Q8LC83	-1.38	0.36	-1.30	-0.95	40S ribosomal protein S24-2	translation (GO:0006412);
ANE-Chitin	GLYMA10G36610	P34788	-1.65	0.28	-1.41	-0.73	40S ribosomal protein S18	translational initiation (GO:0006413);
ANE-Chitin	GLYMA10G36880	P34788	-1.55	0.00	-1.38	-0.97	40S ribosomal protein S18	translational initiation (GO:0006413);
ANE-Chitin	GLYMA10G37840	Q9LZ57	-1.26	0.13	-1.32	-0.67	60S ribosomal protein L36-3	translation (GO:0006412);
ANE-Chitin	GLYMA11G00360	Q5I7K3	-1.19	0.15	-1.01	-0.47	40S ribosomal protein S29	translation (GO:0006412);
ANE-Chitin	GLYMA11G00450	Q9STY6	-1.67	0.29	-1.24	-0.48	40S ribosomal protein S20-2	translation (GO:0006412);
ANE-Chitin	GLYMA11G00890	Q8GTE3	-1.18	0.20	-1.14	-0.81	40S ribosomal protein S3a	translation (GO:0006412);
ANE-Chitin	GLYMA11G05160	P51414	-1.48	0.00	-1.45	-0.95	60S ribosomal protein L26-1	response to cold (GO:0009409); translation (GO:0006412);
ANE-Chitin	GLYMA11G08050	Q42064	-1.42	-0.12	-1.12	-0.96	60S ribosomal protein L8-3	translation (GO:0006412);
ANE-Chitin	GLYMA11G15230	O22644	-1.18	0.20	-1.11	-0.76	60S ribosomal protein L23A	translation (GO:0006412);
ANE-Chitin	GLYMA11G20570	O50003	-1.20	0.37	-1.12	-0.74	60S ribosomal protein L12	translation (GO:0006412);
ANE-Chitin	GLYMA11G25910	Q9LF30	-1.65	0.04	-1.39	-0.66	40S ribosomal protein S19-2	translation (GO:0006412);
ANE-Chitin	GLYMA11G34890	P49211	-1.42	0.06	-1.30	-0.64	60S ribosomal protein L32-1	translation (GO:0006412);
ANE-Chitin	GLYMA11G37160	O48557	-1.27	0.19	-1.05	-0.77	60S ribosomal protein L17	translation (GO:0006412);
ANE-Chitin	GLYMA12G08050	O50003	-1.27	0.16	-1.14	-0.98	60S ribosomal protein L12	translation (GO:0006412);
ANE-Chitin	GLYMA12G32850	O65731	-1.30	0.14	-1.14	-0.54	40S ribosomal protein S5	translation (GO:0006412);
ANE-Chitin	GLYMA13G06920	P49211	-1.67	0.26	-1.26	-0.66	60S ribosomal protein L32-1	translation (GO:0006412);
ANE-Chitin	GLYMA13G19650	Q9M5L0	-1.74	0.04	-1.60	-0.62	60S ribosomal protein L35	translation (GO:0006412);
ANE-Chitin	GLYMA13G20010	Q9M5Z9	-1.15	0.14	-1.01	-0.74	40S ribosomal protein S23	translation (GO:0006412);
ANE-Chitin	GLYMA13G23400	P17093	-1.60	0.58	-1.04	-0.84	40S ribosomal protein S11	translation (GO:0006412);
ANE-Chitin	GLYMA13G35150	Q9LRN8	-1.23	0.00	-1.18	-0.16	50S ribosomal protein L3-2, chloroplastic	translation (GO:0006412);
ANE-Chitin	GLYMA13G35590	Q6UNT2	-1.15	0.00	-1.13	-0.90	60S ribosomal protein L5	translation (GO:0006412);
ANE-Chitin	GLYMA13G37610	O65731	-1.16	0.10	-1.11	-0.78	40S ribosomal protein S5	translation (GO:0006412);
ANE-Chitin	GLYMA13G39270	Q8VZB9	-1.29	0.27	-1.16	-0.83	60S ribosomal protein L10a-1	translation (GO:0006412);
ANE-Chitin	GLYMA13G40780	O22644	-1.34	0.36	-1.23	-0.63	60S ribosomal protein L23A	translation (GO:0006412);
ANE-Chitin	GLYMA13G43450	Q8LEM8	-1.31	0.00	-1.07	-0.45	60S ribosomal protein L37-3	translation (GO:0006412);
ANE-Chitin	GLYMA13G44690	Q9XHS0	-1.43	0.16	-1.48	-0.70	40S ribosomal protein S12	translation (GO:0006412);

ANE-Chitin	GLYMA14G03380	Q8L7K0	-1.55	-0.02	-1.53	-0.92	60S ribosomal protein L18a-1	translation (GO:0006412);
ANE-Chitin	GLYMA14G06820	Q9C912	-1.15	0.00	-1.28	-0.24	60S ribosomal protein L35a-3	translation (GO:0006412);
ANE-Chitin	GLYMA14G38950	Q9LYK9	-1.48	0.56	-1.27	-0.59	40S ribosomal protein S26-3	translation (GO:0006412);
ANE-Chitin	GLYMA15G00610	Q9XHS0	-1.74	0.15	-1.46	-0.59	40S ribosomal protein S12	translation (GO:0006412);
ANE-Chitin	GLYMA15G02610	O82204	-1.32	0.02	-1.16	-0.84	60S ribosomal protein L28-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE-Chitin	GLYMA15G04670	O22644	-1.26	0.22	-1.09	-0.81	60S ribosomal protein L23A	translation (GO:0006412);
ANE-Chitin	GLYMA15G05671	Q96499	-1.25	0.11	-1.23	-0.80	60S ribosomal protein L44	translation (GO:0006412);
ANE-Chitin	GLYMA15G07420	Q9FJX2	-1.40	-0.10	-1.37	-0.86	60S ribosomal protein L26-2	translation (GO:0006412);
ANE-Chitin	GLYMA15G10220	Q8L7K0	-1.40	0.26	-1.08	-0.81	60S ribosomal protein L18a-1	translation (GO:0006412);
ANE-Chitin	GLYMA15G15800	P35687	-1.31	0.16	-1.40	-0.58	40S ribosomal protein S21	endonucleolytic cleavage to generate mature 3'-end of SSU-rRNA from (SSU-rRNA, 5.8S rRNA, LSU-rRNA) (GO:0000461); translation (GO:0006412);
ANE-Chitin	GLYMA15G17010	Q05462	-1.40	0.47	-1.06	-0.94	60S ribosomal protein L27	translation (GO:0006412);
ANE-Chitin	GLYMA15G23220	P49211	-1.46	0.24	-1.10	-0.62	60S ribosomal protein L32-1	translation (GO:0006412);
ANE-Chitin	GLYMA15G24650	P51425	-1.40	0.00	-1.21	-0.52	60S ribosomal protein L39	translation (GO:0006412);
ANE-Chitin	GLYMA15G40520	Q5RCE6	-1.73	-0.17	-1.45	-0.48	Ribosomal L1 domain-containing protein 1	regulation of apoptotic process (GO:0042981); regulation of cellular senescence (GO:2000772);
ANE-Chitin	GLYMA16G03170	Q6UNT2	-1.09	0.16	-1.03	-0.84	60S ribosomal protein L5	translation (GO:0006412);
ANE-Chitin	GLYMA16G17370	P41129	-1.24	0.00	-1.39	-0.93	60S ribosomal protein L13-2	translation (GO:0006412);
ANE-Chitin	GLYMA16G24590	A5VR16	-1.64	0.01	-1.06	-0.92	50S ribosomal protein L7/L12	translation (GO:0006412);
ANE-Chitin	GLYMA16G28550	P51413	-1.60	0.46	-1.02	-0.98	60S ribosomal protein L17-2	translation (GO:0006412);
ANE-Chitin	GLYMA17G02200	P42798	-1.53	0.09	-1.31	-0.88	40S ribosomal protein S15a-1	translation (GO:0006412);
ANE-Chitin	GLYMA18G32680	Q08069	-1.58	0.12	-1.42	-1.00	40S ribosomal protein S8	translation (GO:0006412);
ANE-Chitin	GLYMA18G51660	O49884	-1.46	0.53	-1.25	-0.39	60S ribosomal protein L30	translation (GO:0006412);
ANE-Chitin	GLYMA18G52310	O22644	-1.36	0.61	-1.01	-0.41	60S ribosomal protein L23A	translation (GO:0006412);
ANE-Chitin	GLYMA19G06460	O80929	-1.24	0.17	-1.23	-0.65	60S ribosomal protein L36-1	translation (GO:0006412);
ANE-Chitin	GLYMA19G32680	Q9STY6	-1.06	0.09	-1.04	-0.90	40S ribosomal protein S20-2	translation (GO:0006412);
ANE-Chitin	GLYMA19G36180	Q9SF40	-2.16	-0.06	-2.07	-0.79	60S ribosomal protein L4-1	translation (GO:0006412);
ANE-Chitin	GLYMA19G36250	Q9M5Z9	-1.38	0.01	-1.35	-0.68	40S ribosomal protein S23	translation (GO:0006412);

ANE-Chitin	GLYMA19G38170	P49636	-1.27	0.08	-1.23	-0.64	Ubiquitin-60S ribosomal protein L40	translation (GO:0006412);
ANE-Chitin	GLYMA19G40080	P49693	-1.36	0.06	-1.30	-0.66	60S ribosomal protein L19-3	translation (GO:0006412);
ANE-Chitin	GLYMA19G42030	Q940B0	-1.25	-0.05	-1.06	-0.48	60S ribosomal protein L18-3	translation (GO:0006412);
ANE-Chitin	GLYMA19G42720	O22584	-1.72	0.19	-1.50	-0.91	40S ribosomal protein S14	translation (GO:0006412);
ANE-Chitin	GLYMA20G29981	Q9LZ57	-1.45	0.33	-1.33	-0.67	60S ribosomal protein L36-3	translation (GO:0006412);
ANE-Chitin	GLYMA20G29990	Q9LZ57	-1.35	0.31	-1.31	-0.87	60S ribosomal protein L36-3	translation (GO:0006412);
ANE-Chitin	GLYMA20G30730	P49202	-1.80	0.04	-1.43	-0.75	40S ribosomal protein S18	translation (GO:0006412);
ANE-Chitin	GLYMA20G35000	P55844	-1.65	-0.18	-1.71	-0.93	Probable 60S ribosomal protein L14	translation (GO:0006412);
ANE-Chitin	GLYMA20G37190	Q8LC83	-1.19	0.41	-1.12	-0.95	40S ribosomal protein S24-2	translation (GO:0006412);
ANE-Chitosan	GLYMA02G11540	P46293	-1.12	0.00	-0.97	-1.05	40S ribosomal protein S16	translation (GO:0006412);
ANE-Chitosan	GLYMA02G38450	Q9M5M7	-1.05	0.24	-0.78	-1.01	60S ribosomal protein L10	translation (GO:0006412);
ANE-Chitosan	GLYMA06G14360	P30707	-1.30	0.07	-1.00	-1.09	60S ribosomal protein L9	translation (GO:0006412);
ANE-Chitosan	GLYMA06G20540	Q9FLF0	-1.16	0.17	-0.90	-1.07	40S ribosomal protein S9-2	translation (GO:0006412);
ANE-Chitosan	GLYMA07G06580	Q6UNT2	-1.36	0.64	-0.99	-1.09	60S ribosomal protein L5	translation (GO:0006412);
ANE-Chitosan	GLYMA08G15000	P34091	-1.04	0.00	-0.83	-1.28	60S ribosomal protein L6	translation (GO:0006412);
ANE-Chitosan	GLYMA11G02190	Q9SCM3	-1.01	-0.08	-0.79	-1.25	40S ribosomal protein S2-4	translation (GO:0006412);
ANE-Chitosan	GLYMA20G21190	Q9SIP7	-1.24	0.37	-0.86	-1.02	40S ribosomal protein S3-1	response to salt stress (GO:0009651); translation (GO:0006412);
ANE	GLYMA01G05740	P41098	-1.19	0.45	-0.85	-0.44	60S ribosomal protein L34	translation (GO:0006412);
ANE	GLYMA02G04400	P49637	-1.16	0.00	-1.00	-0.63	60S ribosomal protein L27a-3	translation (GO:0006412);
ANE	GLYMA02G10550	Q9AT35	-1.07	0.32	-0.70	-0.41	60S ribosomal protein L23a	translation (GO:0006412);
ANE	GLYMA02G11920	Q42351	-1.31	0.60	-0.84	-0.40	60S ribosomal protein L34-1	translation (GO:0006412);
ANE	GLYMA03G32380	Q07760	-1.29	0.25	-0.69	-0.49	60S ribosomal protein L23	translation (GO:0006412);
ANE	GLYMA03G35540	P49636	-1.31	0.00	-1.00	-0.71	Ubiquitin-60S ribosomal protein L40	translation (GO:0006412);
ANE	GLYMA03G40530	Q8LC83	-1.11	0.39	-0.87	-0.33	40S ribosomal protein S24-2	translation (GO:0006412);
ANE	GLYMA04G33900	Q9FLF0	-1.20	0.12	-0.95	-0.90	40S ribosomal protein S9-2	translation (GO:0006412);
ANE	GLYMA04G39940	Q9LYK9	-1.13	0.30	-0.84	-0.70	40S ribosomal protein S26-3	translation (GO:0006412);
ANE	GLYMA04G40430	P30707	-1.06	0.47	-0.60	-0.42	60S ribosomal protein L9	translation (GO:0006412);

ANE	GLYMA04G40980	P51423	-1.14	0.62	-0.73	-0.53	Ubiquitin-60S ribosomal protein L40	translation (GO:0006412);
ANE	GLYMA05G03850	P62302	-1.28	0.51	-0.88	-0.43	40S ribosomal protein S13	translation (GO:0006412);
ANE	GLYMA05G03880	P62302	-1.14	0.55	-0.86	-0.77	40S ribosomal protein S13	translation (GO:0006412);
ANE	GLYMA05G03900	P62302	-1.00	0.33	-0.91	-0.54	40S ribosomal protein S13	translation (GO:0006412);
ANE	GLYMA05G24930	Q96499	-1.00	0.39	-0.97	-0.94	60S ribosomal protein L44	translation (GO:0006412);
ANE	GLYMA05G27570	O22860	-1.14	0.00	-0.96	-0.31	60S ribosomal protein L38	translation (GO:0006412);
ANE	GLYMA05G28880	P49211	-1.05	0.03	-0.97	-0.30	60S ribosomal protein L32-1	translation (GO:0006412);
ANE	GLYMA05G34945	A0A0B2QJM8	-1.10	0.42	-0.92	-0.68	30S ribosomal protein S18	translation (GO:0006412);
ANE	GLYMA05G35030	Q945U1	-1.16	0.52	-0.85	-0.47	40S ribosomal protein S15	translation (GO:0006412);
ANE	GLYMA05G36160	P46290	-1.33	0.50	-0.88	-0.28	60S ribosomal protein L31	translation (GO:0006412);
ANE	GLYMA06G14330	P30707	-1.30	0.18	-0.84	-0.91	60S ribosomal protein L9	translation (GO:0006412);
ANE	GLYMA06G14950	P49206	-1.20	0.37	-0.84	-0.68	40S ribosomal protein S26-1	translation (GO:0006412);
ANE	GLYMA06G18800	Q9LR33	-1.21	0.29	-0.87	-0.50	60S ribosomal protein L27a-2	flower development (GO:0009908); inflorescence development (GO:0010229); post-embryonic development (GO:0009791); translation (GO:0006412);
ANE	GLYMA06G36460	P26861	-1.01	0.00	-0.78	-1.00	60S ribosomal protein L6, mitochondrial	translation (GO:0006412);
ANE	GLYMA06G42680	P46287	-1.01	0.38	-0.96	-0.53	60S ribosomal protein L11	translation (GO:0006412);
ANE	GLYMA07G00700	Q8LEM8	-1.06	0.07	-0.99	-0.62	60S ribosomal protein L37-3	translation (GO:0006412);
ANE	GLYMA07G10330	Q9C514	-1.02	0.22	-0.87	-0.79	40S ribosomal protein S7-1	translation (GO:0006412);
ANE	GLYMA08G09200	Q9C912	-1.30	0.43	-0.98	-0.54	60S ribosomal protein L35a-3	translation (GO:0006412);
ANE	GLYMA08G10540	O22860	-1.23	0.15	-0.97	-0.35	60S ribosomal protein L38	translation (GO:0006412);
ANE	GLYMA08G12030	Q9FHG2	-1.47	0.04	-0.91	-0.71	60S ribosomal protein L32-2	translation (GO:0006412);
ANE	GLYMA08G12041	P49211	-1.12	0.16	-0.63	-0.14	60S ribosomal protein L32-1	translation (GO:0006412);
ANE	GLYMA08G16130	Q9FZ76	-1.31	0.09	-0.87	-0.70	60S ribosomal protein L6-1	response to cytokinin (GO:0009735); translation (GO:0006412);
ANE	GLYMA08G18460	Q5RCE6	-1.18	0.00	-0.64	0.00	Ribosomal L1 domain-containing protein 1	regulation of apoptotic process (GO:0042981); regulation of cellular senescence (GO:2000772);
ANE	GLYMA08G19320	Q96499	-1.06	0.50	-0.92	-0.45	60S ribosomal protein L44	translation (GO:0006412);
ANE	GLYMA08G34520	P82413	-1.09	0.00	-0.59	-0.20	50S ribosomal protein L19, chloroplastic	translation (GO:0006412);

ANE	GLYMA08G45770	Q9M3V8	-1.23	0.35	-0.87	-0.60	40S ribosomal protein S6	translation (GO:0006412);
ANE	GLYMA08G46070	Q08069	-1.39	0.22	-0.90	-0.69	40S ribosomal protein S8	translation (GO:0006412);
ANE	GLYMA08G46850	Q9FKC0	-1.22	0.20	-0.83	-0.73	60S ribosomal protein L13a-4	translation (GO:0006412);
ANE	GLYMA09G12420	P17093	-1.19	0.28	-0.78	-0.46	40S ribosomal protein S11	translation (GO:0006412);
ANE	GLYMA09G37360	P40590	-1.18	0.05	-0.65	0.00	60S ribosomal protein L34	translation (GO:0006412);
ANE	GLYMA10G26790	Q9SIP7	-1.13	0.31	-0.72	-0.73	40S ribosomal protein S3-1	response to salt stress (GO:0009651); translation (GO:0006412);
ANE	GLYMA10G26890	Q9SIP7	-1.07	0.34	-0.69	-0.74	40S ribosomal protein S3-1	response to salt stress (GO:0009651); translation (GO:0006412);
ANE	GLYMA10G32580	P55844	-1.08	0.20	-0.78	-0.39	Probable 60S ribosomal protein L14	translation (GO:0006412);
ANE	GLYMA10G43770	Q9M5L0	-1.18	0.22	-0.91	-0.60	60S ribosomal protein L35	translation (GO:0006412);
ANE	GLYMA12G31040	Q8VZB9	-1.23	0.45	-0.95	-0.60	60S ribosomal protein L10a-1	translation (GO:0006412);
ANE	GLYMA13G20330	O50003	-1.22	0.15	-0.90	-0.45	60S ribosomal protein L12	translation (GO:0006412);
ANE	GLYMA13G24630	Q9M3V8	-1.18	0.23	-0.99	-0.77	40S ribosomal protein S6	translation (GO:0006412);
ANE	GLYMA13G28831	P51418	-1.13	0.37	-0.72	-0.61	60S ribosomal protein L18a-2	translation (GO:0006412);
ANE	GLYMA13G40700	Q9M5L0	-1.00	0.00	-0.74	-0.89	60S ribosomal protein L35	translation (GO:0006412);
ANE	GLYMA13G42090	O22860	-1.22	0.00	-0.83	-0.52	60S ribosomal protein L38	translation (GO:0006412);
ANE	GLYMA14G36620	P93847	-1.21	0.18	-0.99	-0.92	60S ribosomal protein L10	translation (GO:0006412);
ANE	GLYMA14G39435	P07134	-1.08	0.00	0.00	-0.77	30S ribosomal protein S12, chloroplastic	translation (GO:0006412);
ANE	GLYMA15G01860	Q8LEM8	-1.05	0.27	-0.85	-0.46	60S ribosomal protein L37-3	translation (GO:0006412);
ANE	GLYMA15G41870	B8ELE4	-1.00	0.00	-0.83	-0.73	50S ribosomal protein L15	translation (GO:0006412);
ANE	GLYMA16G13580	Q9FKC0	-1.01	0.22	-0.72	-0.76	60S ribosomal protein L13a-4	translation (GO:0006412);
ANE	GLYMA16G33230	Q9SIP7	-1.00	0.30	-0.90	-0.77	40S ribosomal protein S3-1	response to salt stress (GO:0009651); translation (GO:0006412);
ANE	GLYMA17G00820	P31674	-1.11	0.26	-0.77	-0.80	40S ribosomal protein S15	translation (GO:0006412);
ANE	GLYMA17G03550	Q84WM0	-1.07	0.50	-0.89	-0.17	60S ribosomal protein L29-2	translation (GO:0006412);
ANE	GLYMA17G09280	P49637	-1.01	0.36	-0.81	-0.62	60S ribosomal protein L27a-3	translation (GO:0006412);
ANE	GLYMA17G11430	P17093	-1.27	0.32	-1.00	-0.93	40S ribosomal protein S11	translation (GO:0006412);
ANE	GLYMA18G26190	Q9M3V8	-1.27	0.39	-0.96	-0.76	40S ribosomal protein S6	translation (GO:0006412);

ANE	GLYMA19G03520	Q9LF30	-1.21	0.50	-0.73	-0.34	40S ribosomal protein S19-2	translation (GO:0006412);
ANE	GLYMA19G42090	Q9M9W1	-1.08	0.48	-0.81	-0.43	60S ribosomal protein L22-2	translation (GO:0006412);
ANE	GLYMA20G38080	Q9M9W1	-1.14	0.39	-0.94	-0.30	60S ribosomal protein L22-2	translation (GO:0006412);
ANE-Alginate-Chitin	GLYMA11G12200	O04658	-1.80	-1.04	-2.36	-0.41	Probable nucleolar protein 5-1	ribosome biogenesis (GO:0042254);
ANE-Alginate-Chitin	GLYMA12G04400	O04658	-1.74	-1.42	-2.30	-0.39	Probable nucleolar protein 5-1	ribosome biogenesis (GO:0042254);
ANE-Chitin-Chitosan	GLYMA12G03230	P35685	-1.23	0.00	-1.39	-1.05	60S ribosomal protein L7a-1	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA02G39510	Q9U3Z7	-1.29	0.12	-1.11	-0.48	NHP2-like protein 1 homolog	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA10G24060	Q9M8Z5	-1.68	0.00	-1.39	-0.42	Guanine nucleotide-binding protein-like NSN1	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA12G06170	Q9C6I8	-1.79	0.15	-1.24	-0.91	Nucleolar GTP-binding protein 1	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA14G06630	P50346	-1.75	0.19	-1.48	-0.97	60S acidic ribosomal protein P0	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA11G11040	P35685	-1.51	0.27	-1.28	-0.87	60S ribosomal protein L7a-1	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA02G42260	P50346	-1.72	0.23	-1.26	-0.80	60S acidic ribosomal protein P0	ribosome biogenesis (GO:0042254);
ANE-Chitin	GLYMA02G00540	P49692	-1.08	0.18	-1.10	-0.63	60S ribosomal protein L7a-1	ribosome biogenesis (GO:0042254);
ANE	GLYMA05G13330	A9UNU6	-1.38	0.00	-0.94	-0.05	Bystin	ribosome biogenesis (GO:0042254);
ANE	GLYMA08G19660	A4FV84	-1.05	0.11	-0.82	0.00	mRNA turnover protein 4 homolog	ribosome biogenesis (GO:0042254);
ANE	GLYMA10G29560	Q9LUJ5	-1.03	0.00	-0.75	-0.17	Probable rRNA-processing protein EBP2 homolog	ribosome biogenesis (GO:0042254);
ANE	GLYMA11G14190	Q9C6I8	-1.04	0.25	-0.50	-0.72	Nucleolar GTP-binding protein 1	ribosome biogenesis (GO:0042254);
ANE	GLYMA13G26190	F4I366	-1.13	0.39	-0.88	-0.48	DNA-directed RNA polymerase I subunit 2	ribosome biogenesis (GO:0042254);
ANE	GLYMA14G37600	Q6P8E9	-1.37	0.31	-0.99	-0.60	NHP2-like protein 1	ribosome biogenesis (GO:0042254);
ANE	GLYMA15G05360	A4FV84	-1.10	0.20	-0.97	-0.10	mRNA turnover protein 4 homolog	ribosome biogenesis (GO:0042254);
ANE	GLYMA19G28070	Q9M2F1	-1.28	0.41	-0.95	-0.32	40S ribosomal protein S27-2	ribosomal small subunit assembly (GO:0000028);
ANE-Chitin-Chitosan	GLYMA07G04890	O22518	-1.48	0.04	-1.36	-1.17	40S ribosomal protein SA	ribosomal small subunit assembly (GO:0000028);
ANE-Chitin-Chitosan	GLYMA10G36780	P35684	-2.03	0.41	-1.80	-1.46	60S ribosomal protein L3	ribosomal large subunit assembly (GO:0000027);
ANE	GLYMA16G05090	Q9M2F1	-1.08	0.52	-0.71	-0.23	40S ribosomal protein S27-2	ribosomal small subunit assembly (GO:0000028);
ANE	GLYMA02G26840	Q9ZUT9	-1.16	0.24	-0.99	-0.52	40S ribosomal protein S5-1	ribosomal small subunit assembly (GO:0000028);

ANE	GLYMA02G07430	Q9M2F1	-1.03	0.16	-0.88	-0.64	40S ribosomal protein S27-2	ribosomal small subunit assembly (GO:0000028);
ANE-Chitin-Chitosan	GLYMA17G10340	A4QND5	-1.99	0.00	-1.77	-1.07	60S ribosome subunit biogenesis protein NIP7 homolog	ribosome assembly (GO:0042255);
ANE-Chitin-Chitosan	GLYMA20G30810	P35684	-1.41	0.04	-1.35	-1.03	60S ribosomal protein L3	ribosomal large subunit assembly (GO:0000027);
ANE-Chitin-Chitosan	GLYMA07G03980	Q869L3	-1.22	0.09	-1.01	-1.17	Midasin	ribosomal large subunit assembly (GO:0000027);
ANE-Chitin-Chitosan	GLYMA16G01460	O22518	-1.39	0.04	-1.14	-1.10	40S ribosomal protein SA	ribosomal small subunit assembly (GO:0000028); rRNA export from nucleus (GO:0006407);
ANE-Chitin-Chitosan	GLYMA15G42150	O22518	-1.45	0.00	-1.47	-1.13	40S ribosomal protein SA	ribosomal small subunit assembly (GO:0000028); rRNA export from nucleus (GO:0006407);
ANE-Chitin	GLYMA05G01560	A4QND5	-1.46	-0.14	-1.45	-0.68	60S ribosome subunit biogenesis protein NIP7 homolog	ribosome assembly (GO:0042255);
ANE-Chitin	GLYMA10G08910	P49215	-1.40	0.02	-1.46	-0.89	40S ribosomal protein S17	ribosomal small subunit assembly (GO:0000028); translation (GO:0006412);
ANE-Chitin	GLYMA09G15680	Q9ZUT9	-1.17	0.07	-1.16	-0.62	40S ribosomal protein S5-1	response to cytokinin (GO:0009735); ribosomal small subunit assembly (GO:0000028);
ANE-Chitin	GLYMA20G30970	P34788	-1.39	0.15	-1.24	-0.95	40S ribosomal protein S18	translational initiation (GO:0006413);
ANE-Chitin-Chitosan	GLYMA06G14080	P52855	-1.29	0.00	-1.30	-1.02	60S acidic ribosomal protein P1	translational elongation (GO:0006414);
ANE	GLYMA13G39490	O24415	-1.02	0.36	-0.74	-0.61	60S acidic ribosomal protein P2B	translational elongation (GO:0006414);
ANE	GLYMA05G30780	Q9LVC9	-1.02	0.28	-0.96	-0.44	60S acidic ribosomal protein P3-2	translational elongation (GO:0006414);
ANE-Chitin	GLYMA19G44916	Q8LCW9	-1.35	0.23	-1.43	-0.96	60S acidic ribosomal protein P1-1	translational elongation (GO:0006414);
ANE-Chitin	GLYMA19G37780	P41099	-1.60	0.04	-1.61	-0.82	60S acidic ribosomal protein P2	translational elongation (GO:0006414);
ANE-Chitin	GLYMA12G30800	O24415	-1.20	0.30	-1.11	-0.63	60S acidic ribosomal protein P2B	translational elongation (GO:0006414);
ANE-Chitin	GLYMA08G13970	Q9LVC9	-1.38	0.19	-1.14	-0.71	60S acidic ribosomal protein P3-2	translational elongation (GO:0006414);
ANE-Chitin	GLYMA04G40720	P52855	-1.34	-0.01	-1.39	-0.75	60S acidic ribosomal protein P1	translational elongation (GO:0006414);
ANE-Chitin	GLYMA03G42180	Q8LCW9	-1.36	0.33	-1.13	-0.54	60S acidic ribosomal protein P1-1	translational elongation (GO:0006414);
ANE-Chitin	GLYMA03G35080	P41099	-1.25	0.16	-1.25	-0.91	60S acidic ribosomal protein P2	translational elongation (GO:0006414);
ANE-Chitin	GLYMA0169S00200	P41099	-1.36	0.12	-1.58	-0.93	60S acidic ribosomal protein P2	translational elongation (GO:0006414);
ANE-Chitin	GLYMA13G26300	B5BAM9	-1.61	0.00	-1.50	-0.81	Ribosome-recycling factor	translational termination (GO:0006415);

ANE-Chitin-Chitosan	GLYMA07G02270	P60039	-1.38	0.15	-1.23	-1.05	60S ribosomal protein L7-3	No-GOBP
ANE	GLYMA10G07630	Q9LUG5	-1.09	0.00	-0.77	-0.34	Ribosome production factor 2 homolog	No-GOBP
ANE-Chitin	GLYMA13G00430	A0A0B2QS30	-1.07	-0.64	-1.54	-0.31	Nucleolar and coiled-body phosphoprotein 1	No-GOBP
ANE-Chitin	GLYMA17G23890	Q9ZPV5	-1.75	0.00	-1.53	-0.58	Nucleolar complex protein 2 homolog	No-GOBP
ANE-Chitin	GLYMA15G04450	Q9ZPV5	-1.11	-0.64	-1.53	-0.06	Nucleolar complex protein 2 homolog	No-GOBP
ANE-Chitin	GLYMA09G30130	Q6NVM6	-1.29	-0.27	-1.59	-0.64	Nucleolar protein 10	No-GOBP
ANE-Chitin	GLYMA01G38280	Q6NRY2	-1.51	0.00	-1.23	0.00	Nucleolar protein 6	No-GOBP
ANE	GLYMA11G12300	Q9SIK2	-1.03	0.16	-0.80	-0.51	40S ribosomal protein S25-2	No-GOBP
ANE	GLYMA12G04510	Q9SIK2	-1.00	0.79	-0.51	-0.37	40S ribosomal protein S25-2	No-GOBP
ANE	GLYMA15G18170	P60039	-1.02	0.21	-0.94	-0.77	60S ribosomal protein L7-3	No-GOBP
ANE-Chitin	GLYMA13G21520	O65743	-1.26	0.00	-1.09	-0.59	60S ribosomal protein L24	No-GOBP
ANE-Chitin	GLYMA11G02410	Q9LTF2	-1.34	0.00	-1.11	-0.79	40S ribosomal protein S10-3	No-GOBP
ANE-Chitin-Chitosan	GLYMA08G23750	P60040	-1.33	0.09	-1.12	-1.06	60S ribosomal protein L7-2	No-GOBP
ANE-Chitin	GLYMA01G43110	Q9LTF2	-1.15	-0.05	-1.11	-0.85	40S ribosomal protein S10-3	No-GOBP
ANE-Chitin	GLYMA10G07680	O65743	-1.09	0.07	-1.14	-0.73	60S ribosomal protein L24	No-GOBP
ANE-Chitin	GLYMA03G34700	Q9FUL4	-1.15	0.00	-1.14	-0.92	60S ribosomal protein L24	No-GOBP

Table 17: ANE down-regulated genes involved in RNA processing

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA05G03150	Q54MT2	-1.90	-1.19	-2.36	-0.44	Nucleolar protein 56	rRNA processing (GO:0006364);
ANE-Alginate-Chitin	GLYMA17G13770	O00567	-2.34	-1.08	-2.69	-0.54	Nucleolar protein 56	rRNA modification (GO:0000154); rRNA processing (GO:0006364);
ANE-Chitin-Chitosan	GLYMA19G28740	Q6FSB0	-2.11	-0.20	-2.08	-1.08	H/ACA ribonucleoprotein complex subunit 1	pseudouridine synthesis (GO:0001522); rRNA processing (GO:0006364);

ANE-Chitin-Chitosan	GLYMA19G37051	Q8BHB4	-2.51	0.00	-1.74	-1.52	WD repeat-containing protein 3	No-GOBP
ANE-Chitin	GLYMA01G01390	Q6YS30	-1.18	-0.58	-1.60	0.51	DEAD-box ATP-dependent RNA helicase 5	RNA methylation (GO:0001510); RNA secondary structure unwinding (GO:0010501); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA02G45960	Q9C8Z4	-1.50	0.00	-1.08	-0.53	Uncharacterized protein	rRNA processing (GO:0006364);
ANE-Chitin	GLYMA03G32150	Q93XX8	-1.20	0.27	-1.09	-0.08	H/ACA ribonucleoprotein complex subunit 3-like protein	polar nucleus fusion (GO:0010197); pseudouridine synthesis (GO:0001522); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA09G02690	Q3MKM6	-1.42	0.00	-2.07	0.00	U3 snoRNP-associated protein-like EMB2271	rRNA processing (GO:0006364);
ANE-Chitin	GLYMA08G47440	Q8NI36	-1.60	0.00	-1.30	-0.86	WD repeat-containing protein 36	response to stimulus (GO:0050896); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA09G15786	F4J8K6	-1.38	0.22	-2.04	-0.73	rRNA biogenesis protein RRP5	embryo sac development (GO:0009553); mRNA processing (GO:0006397); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA09G34390	Q9C551	-1.65	-0.35	-1.81	0.00	DEAD-box ATP-dependent RNA helicase 5	RNA secondary structure unwinding (GO:0010501); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA11G07340	Q9H6R4	-2.03	0.00	-1.38	-0.58	Nucleolar protein 6	rRNA processing (GO:0006364); tRNA export from nucleus (GO:0006409);
ANE-Chitin	GLYMA11G10790	Q7XTT4	-1.32	0.23	-1.27	-0.81	Nucleolin 2	rRNA processing (GO:0006364);
ANE-Chitin	GLYMA16G04570	Q8VZT0	-1.80	0.00	-1.58	-0.57	Putative H/ACA ribonucleoprotein complex subunit 1-like protein 1	pseudouridine synthesis (GO:0001522); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA17G09590	Q810V0	-1.84	0.00	-1.58	-0.11	U3 small nucleolar ribonucleoprotein protein MPP10	negative regulation of phosphatase activity (GO:0010923); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA12G03070	Q7XTT4	-1.45	-0.13	-1.52	-0.76	Nucleolin 2	rRNA processing (GO:0006364);
ANE	GLYMA02G26710	F4J8K6	-1.11	0.05	-0.74	-0.75	rRNA biogenesis protein RRP5	embryo sac development (GO:0009553); mRNA processing (GO:0006397); rRNA processing (GO:0006364);
ANE	GLYMA02G41900	Q803X4	-1.06	0.04	-0.75	-0.60	DDB1- and CUL4-associated factor 13	protein ubiquitination (GO:0016567); rRNA processing (GO:0006364);
ANE	GLYMA04G27730	O13823	-1.55	0.00	-0.70	-0.42	U3 small nucleolar ribonucleoprotein protein imp4	rRNA processing (GO:0006364);
ANE	GLYMA08G44490	O94268	-1.19	0.00	-0.94	-0.36	25S rRNA (cytosine-C(5))-methyltransferase nop2	rRNA methylation (GO:0031167); rRNA processing (GO:0006364);
ANE	GLYMA11G21290	Q5R947	-1.16	0.08	-0.72	-0.13	U3 small nucleolar ribonucleoprotein protein IMP4	rRNA processing (GO:0006364);
ANE	GLYMA15G13570	Q9M0V4	-1.20	0.00	-0.72	-0.19	U3 snoRNP-associated protein-like YAO	acceptance of pollen (GO:0060321); embryo development ending in seed dormancy (GO:0009793); embryo sac development (GO:0009553); rRNA processing (GO:0006364);

ANE	GLYMA19G34910	Q93XX8	-1.18	0.29	-0.97	-0.18	H/ACA ribonucleoprotein complex subunit 3-like protein	polar nucleus fusion (GO:0010197); pseudouridine synthesis (GO:0001522); rRNA processing (GO:0006364);
ANE	GLYMA03G34360	Q8BHB4	-1.42	0.28	-0.99	-0.63	WD repeat-containing protein 3	No-GOBP
ANE-Chitin-Chitosan	GLYMA18G02340	Q94AH9	-1.75	-0.22	-1.77	-1.01	Mediator of RNA polymerase II transcription subunit 36a	rRNA methylation (GO:0031167); transcription, DNA-templated (GO:0006351); tRNA processing (GO:0008033);
ANE-Chitin-Chitosan	GLYMA02G43400	Q8L633	-1.41	0.04	-1.57	-1.77	Ribonuclease Z, chloroplastic	tRNA 3'-end processing (GO:0042780); tRNA 3'-trailer cleavage, endonucleolytic (GO:0034414); tRNA processing (GO:0008033);
ANE-Chitin-Chitosan	GLYMA05G35530	Q66GI4	-1.18	0.00	-2.22	-1.46	Proteinaceous RNase P 1, chloroplastic/mitochondrial	tRNA 5'-leader removal (GO:0001682); tRNA processing (GO:0008033);
ANE-Chitin-Chitosan	GLYMA14G05580	Q8L633	-1.18	0.07	-1.21	-1.38	Ribonuclease Z, chloroplastic	tRNA 3'-trailer cleavage, endonucleolytic (GO:0034414); tRNA processing (GO:0008033);
ANE-Chitin	GLYMA06G17310	Q94AH9	-1.98	0.17	-1.20	-0.73	Mediator of RNA polymerase II transcription subunit 36a	rRNA methylation (GO:0031167); transcription, DNA-templated (GO:0006351); tRNA processing (GO:0008033);
ANE-Chitin	GLYMA04G37770	Q94AH9	-2.20	0.24	-2.00	-0.87	Mediator of RNA polymerase II transcription subunit 36a	rRNA methylation (GO:0031167); transcription, DNA-templated (GO:0006351); tRNA processing (GO:0008033);
ANE-Chitin	GLYMA11G36090	P22509	-2.28	0.00	-1.96	-1.00	rRNA 2'-O-methyltransferase fibrillarin	RNA processing (GO:0006396); rRNA processing (GO:0006364); snoRNA localization (GO:0048254); snoRNA metabolic process (GO:0016074); tRNA processing (GO:0008033);
ANE-Chitin	GLYMA19G00280	Q9S7G6	-1.67	0.00	-1.18	-0.39	Polyribonucleotide nucleotidyltransferase 2, mitochondrial	RNA phosphodiester bond hydrolysis, exonucleolytic (GO:0090503); rRNA processing (GO:0006364); tRNA processing (GO:0008033);
ANE-Chitin-Chitosan	GLYMA04G32180	Q8L4J2	-1.08	-0.58	-1.17	-2.20	Cleavage stimulation factor subunit 50	mRNA processing (GO:0006397);
ANE-Chitin-Chitosan	GLYMA01G34980	Q9SL79	-1.72	0.00	-1.45	-1.02	CRS2-associated factor 1, chloroplastic	Group II intron splicing (GO:0000373); mRNA processing (GO:0006397);
ANE-Chitin	GLYMA15G15056	F4KDH9	-1.74	0.00	-2.09	-0.94	FIP1[V]-like protein	mRNA processing (GO:0006397);
ANE	GLYMA15G13600	P38456	-1.52	0.00	-0.56	-0.33	Uncharacterized mitochondrial protein ymf11	mRNA processing (GO:0006397);
ANE	GLYMA17G07060	Q9SUM2	-1.60	0.66	-0.81	-0.69	Probable small nuclear ribonucleoprotein F	mRNA splicing, via spliceosome (GO:0000398);
ANE	GLYMA17G11410	Q5F3X4	-1.18	0.00	-0.95	-0.68	116 kDa U5 small nuclear ribonucleoprotein component	mRNA processing (GO:0006397); RNA splicing (GO:0008380);
ANE	GLYMA09G04031	F4KDH9	-1.25	-0.36	-0.75	-0.79	FIP1[V]-like protein	mRNA processing (GO:0006397);
ANE-Chitin-Chitosan	GLYMA09G30770	Q8S9K3	-1.35	0.21	-2.20	-1.54	Zinc finger protein VAR3, chloroplastic	chloroplast mRNA modification (GO:1900871); chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA17G08630	Q9FNR1	-1.29	0.13	-1.34	-1.36	Glycine-rich RNA-binding protein 3, mitochondrial	mitochondrial mRNA modification (GO:0080156); response to cold (GO:0009409);

ANE-Chitin	GLYMA13G34670	Q9LKA5	-1.02	-0.32	-1.06	-0.69	Multiple organellar RNA editing factor 8, chloroplastic/mitochondrial	mitochondrial mRNA modification (GO:0080156); RNA modification (GO:0009451);
ANE-Chitin	GLYMA07G11495	Q8S9K3	-1.30	0.00	-1.36	-0.57	Zinc finger protein VAR3, chloroplastic	chloroplast mRNA modification (GO:1900871); chloroplast organization (GO:0009658);
ANE	GLYMA02G09300	Q8S9K3	-1.02	0.00	-0.96	-0.76	Zinc finger protein VAR3, chloroplastic	chloroplast mRNA modification (GO:1900871); chloroplast organization (GO:0009658);
ANE	GLYMA15G02070	Q8CGS5	-1.31	0.28	-0.68	-0.48	Zinc phosphodiesterase ELAC protein 2	mitochondrial tRNA 3'-trailer cleavage, endonucleolytic (GO:0072684);
ANE	GLYMA17G23520	I1MW40	-1.01	0.25	-0.61	-0.75	Uncharacterized protein	RNA methylation (GO:0001510);
ANE	GLYMA18G50250	O94260	-1.03	-0.21	-0.86	-0.60	Putative G3BP-like protein	mRNA transport (GO:0051028); protein deubiquitination (GO:0016579);
ANE-Chitin-Chitosan	GLYMA08G01200	A0A0R0IMG1	-1.78	0.00	-2.48	-1.19	Uncharacterized protein	RNA processing (GO:0006396);
ANE-Chitosan	GLYMA20G04820	P42813	-1.11	0.00	0.73	-1.33	Ribonuclease 1	anthocyanin-containing compound biosynthetic process (GO:0009718); response to wounding (GO:0009611); RNA phosphodiester bond hydrolysis (GO:0090501);
ANE-Chitin	GLYMA03G30620	Q9ZVT7	-1.14	-0.21	-1.28	0.00	Exosome complex component RRP4 homolog	exonucleolytic trimming to generate mature 3'-end of 5.8S rRNA from tricistronic rRNA transcript (SSU-rRNA, 5.8S rRNA, LSU-rRNA) (GO:0000467);
ANE-Chitin	GLYMA05G02240	Q12788	-1.63	0.00	-1.36	-0.50	Transducin beta-like protein 3	maturation of SSU-rRNA from tricistronic rRNA transcript (SSU-rRNA, 5.8S rRNA, LSU-rRNA) (GO:0000462);
ANE-Chitin	GLYMA09G10290	P25635	-1.51	-0.13	-1.67	0.00	Periodic tryptophan protein 2	endonucleolytic cleavage to generate mature 5'-end of SSU-rRNA from (SSU-rRNA, 5.8S rRNA, LSU-rRNA) (GO:0000472);
ANE-Chitin	GLYMA10G34390	Q9P4X3	-2.02	0.03	-1.47	-0.81	Probable U3 small nucleolar RNA-associated protein 7	maturation of SSU-rRNA (GO:0030490);
ANE-Chitin	GLYMA20G33160	Q9P4X3	-1.18	0.00	-1.20	-0.89	Probable U3 small nucleolar RNA-associated protein 7	maturation of SSU-rRNA (GO:0030490);
ANE	GLYMA17G01570	Q9NYH9	-1.34	0.00	-0.93	-0.45	U3 small nucleolar RNA-associated protein 6 homolog	maturation of SSU-rRNA from tricistronic rRNA transcript (SSU-rRNA, 5.8S rRNA, LSU-rRNA) (GO:0000462);
ANE	GLYMA17G35220	Q8R2N2	-1.01	0.00	-0.66	-0.21	U3 small nucleolar RNA-associated protein 4 homolog	maturation of SSU-rRNA (GO:0030490); regulation of transcription, DNA-templated (GO:0006355);
ANE-Alginate-Chitin	GLYMA08G18860	Q9LD90	-1.80	-1.41	-2.35	-0.52	H/ACA ribonucleoprotein complex subunit 4	box H/ACA snoRNA 3'-end processing (GO:0000495); mRNA pseudouridine synthesis (GO:1990481);
ANE-Alginate-Chitin	GLYMA15G06160	Q9LD90	-2.35	-1.23	-2.70	-0.62	H/ACA ribonucleoprotein complex subunit 4	box H/ACA snoRNA 3'-end processing (GO:0000495); mRNA pseudouridine synthesis (GO:1990481

ANE	GLYMA17G09070	Q9LEY9	-1.09	-0.12	-1.00	-0.98	H/ACA ribonucleoprotein complex subunit 2-like protein	rRNA pseudouridine synthesis (GO:0031118);
ANE	GLYMA17G09100	Q9LEY9	-1.00	-0.17	-0.85	-0.81	H/ACA ribonucleoprotein complex subunit 2-like protein	rRNA pseudouridine synthesis (GO:0031118);
ANE-Chitin-Chitosan	GLYMA07G06240	Q5ZBH5	-1.40	-0.38	-1.71	-1.94	DEAD-box ATP-dependent RNA helicase 25	RNA secondary structure unwinding (GO:0010501);
ANE-Chitin-Chitosan	GLYMA10G28100	Q8L7S8	-1.80	0.02	-2.29	-2.26	DEAD-box ATP-dependent RNA helicase 3, chloroplastic	RNA secondary structure unwinding (GO:0010501);
ANE-Chitin-Chitosan	GLYMA16G02880	Q0DBU5	-1.49	-0.28	-1.65	-1.47	DEAD-box ATP-dependent RNA helicase 31	RNA secondary structure unwinding (GO:0010501);
ANE-Chitin-Chitosan	GLYMA20G22120	Q0DM51	-2.24	-0.17	-2.47	-2.88	DEAD-box ATP-dependent RNA helicase 3, chloroplastic	RNA secondary structure unwinding (GO:0010501);
ANE-Chitin	GLYMA17G13230	Q84T03	-1.05	-0.57	-1.44	0.00	DEAD-box ATP-dependent RNA helicase 27	RNA secondary structure unwinding (GO:0010501);
ANE-Chitosan	GLYMA08G17220	Q6H874	-1.13	0.02	-0.92	-1.57	DEAD-box ATP-dependent RNA helicase 47A	RNA secondary structure unwinding (GO:0010501);
ANE-Chitosan	GLYMA15G41980	Q8W4E1	-1.11	0.02	-0.89	-1.53	DEAD-box ATP-dependent RNA helicase 47, mitochondrial	plasmodesma organization (GO:0009663); plasmodesmata-mediated intercellular transport (GO:0010497); RNA secondary structure unwinding (GO:0010501);
ANE	GLYMA14G02940	Q9PV94	-1.28	-0.12	-0.96	-0.67	Small nuclear ribonucleoprotein-associated protein B'	mRNA splicing, via spliceosome (GO:0000398); spliceosomal snRNP assembly (GO:0000387);

Table 18: ANE down-regulated genes involved in N-assimilation.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin	GLYMA14G33480	P39866	-1.10	0.36	-1.00	0.00	Nitrate reductase [NADH] 2	nitrate assimilation (GO:0042128);

Table 19: ANE down-regulated genes involved in amino acid biosynthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes

ANE-Chitin-Chitosan	GLYMA06G13280	Q03460	-1.40	0.30	-1.77	-1.70	Glutamate synthase [NADH], amyloplastic	glutamine metabolic process (GO:0006541); L-glutamate biosynthetic process (GO:0097054);
ANE-Chitin-Chitosan	GLYMA04G41540	Q03460	-1.38	0.00	-1.12	-1.59	Glutamate synthase [NADH], amyloplastic	glutamine metabolic process (GO:0006541); L-glutamate biosynthetic process (GO:0097054);
ANE-Chitin-Chitosan	GLYMA08G05250	Q04797	-1.35	-0.40	-1.62	-1.44	Aspartate-semialdehyde dehydrogenase	'de novo' L-methionine biosynthetic process (GO:0071266); threonine biosynthetic process (GO:0009088);
ANE-Chitin-Chitosan	GLYMA16G03180	Q8LB01	-1.42	-0.08	-1.77	-1.99	4-hydroxy-tetrahydrodipicolinate reductase 2, chloroplastic	diaminopimelate biosynthetic process (GO:0019877); lysine biosynthetic process via diaminopimelate (GO:0009089);
ANE-Chitin-Chitosan	GLYMA15G03120	Q9MT28	-1.55	0.12	-1.27	-1.20	Threonine synthase, chloroplastic	Threonine biosynthetic process (GO:0009088);
ANE-Chitin-Chitosan	GLYMA13G36730	O82043	-1.21	0.08	-1.09	-1.28	Ketol-acid reductoisomerase, chloroplastic	isoleucine biosynthetic process (GO:0009097); valine biosynthetic process (GO:0009099);
ANE-Chitin-Chitosan	GLYMA10G40750	O49485	-2.06	-0.64	-2.28	-1.62	D-3-phosphoglycerate dehydrogenase 1, chloroplastic	serine family amino acid biosynthetic process (GO:0009070);
ANE-Chitin-Chitosan	GLYMA20G26530	O49485	-2.16	-0.09	-2.25	-1.57	D-3-phosphoglycerate dehydrogenase 1, chloroplastic	serine family amino acid biosynthetic process (GO:0009070);
ANE-Chitin	GLYMA08G23860	O04130	-1.61	0.10	-1.40	-0.31	D-3-phosphoglycerate dehydrogenase 2, chloroplastic	L-serine biosynthetic process (GO:0006564);
ANE-Chitin	GLYMA13G34290	P93164	-1.14	0.47	-1.57	-0.86	Gamma-glutamyl hydrolase	glutamine metabolic process (GO:0006541); proteolysis (GO:0006508);
ANE-Chitosan	GLYMA07G29630	Q93Z70	-1.27	0.14	-0.82	-1.56	Probable N-acetyl-gamma-glutamyl-phosphate reductase, chloroplastic	arginine biosynthetic process (GO:0006526); response to cadmium ion (GO:0046686);
ANE-Chitin-Chitosan	GLYMA13G27810	Q9LIR4	-1.73	-0.11	-2.23	-1.91	Dihydroxy-acid dehydratase, chloroplastic	branched-chain amino acid biosynthetic process (GO:0009082); response to salt stress (GO:0009651); valine biosynthetic process (GO:0009099);
ANE-Chitin-Chitosan	GLYMA02G42800	Q28HE5	-1.14	-0.56	-2.62	-1.36	Probable sodium-coupled neutral amino acid transporter 6	amino acid transmembrane transport (GO:0003333); sodium ion transport (GO:0006814);
ANE-Chitin-Chitosan	GLYMA14G10260	Q9H2H9	-1.01	-0.65	-1.48	-1.53	Sodium-coupled neutral amino acid transporter 1	amino acid transmembrane transport (GO:0003333); glutamine transport (GO:0006868);
ANE-Chitin-Chitosan	GLYMA13G02960	Q9LPF1	-1.02	-0.86	-2.94	-1.61	WAT1-related protein	L-alpha-amino acid transmembrane transport (GO:1902475);
ANE	GLYMA05G03190	Q9SZX3	-1.12	0.00	-0.71	-0.48	Argininosuccinate synthase, chloroplastic	arginine biosynthetic process (GO:0006526);

Table 20: ANE down-regulated genes involved in translation-related processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA19G07240	O64937	-1.59	0.48	-1.19	-1.18	Elongation factor 1-alpha	No-GOBP
ANE-Chitin-Chitosan	GLYMA09G01590	Q8GWE0	-1.68	0.00	-1.60	-2.44	Pentatricopeptide repeat-containing protein, chloroplastic	chloroplast organization (GO:0009658); chloroplast RNA processing (GO:0031425); positive regulation of translation (GO:0045727);
ANE-Chitin	GLYMA02G25240	Q0INC5	-1.08	0.00	-1.01	-0.19	DEAD-box ATP-dependent RNA helicase 28	translational initiation (GO:0006413);
ANE-Chitin	GLYMA03G42410	Q9C552	-1.05	-0.14	-1.51	-0.19	Pumilio homolog 23	auxin homeostasis (GO:0010252); regulation of translation (GO:0006417); response to glucose (GO:0009749); response to sucrose (GO:0009744);
ANE-Chitin	GLYMA17G06830	Q9C5E7	-1.00	0.00	-1.12	0.00	Pumilio homolog 6, chloroplastic	regulation of translation (GO:0006417);
ANE-Chitin	GLYMA03G30720	B2UQY9	-2.05	0.00	-1.48	-0.44	Elongation factor Tu	No-GOBP
ANE-Chitin	GLYMA16G00360	Q9FUM1	-1.44	0.03	-1.28	-0.92	Elongation factor 1-gamma	No-GOBP
ANE-Chitin	GLYMA19G33570	Q3SLQ1	-1.54	0.00	-1.36	-0.53	Elongation factor Tu	No-GOBP
ANE-Chitin	GLYMA18G08360	O15160	-1.44	0.00	-1.08	-0.77	DNA-directed RNA polymerases I and III subunit RPAC1	transcription from RNA polymerase III promoter (GO:0006383);transcription initiation from RNA polymerase I promoter (GO:0006361);
ANE-Chitosan	GLYMA06G17930	P29545	-1.15	0.41	-0.75	-1.17	Elongation factor 1-beta	No-GOBP
ANE-Chitosan	GLYMA12G28670	Q6YW46	-1.11	0.24	-0.96	-1.05	Elongation factor 1-gamma 2	glutathione metabolic process (GO:0006749);
ANE-Chitosan	GLYMA17G13371	P46280	-1.41	0.68	-0.33	-1.34	Elongation factor Tu, chloroplastic	mitochondrial translational elongation (GO:0070125);
ANE-Chitosan	GLYMA09G01570	Q8GWE0	-1.05	0.02	-0.50	-1.70	Pentatricopeptide repeat-containing protein, chloroplastic	chloroplast organization (GO:0009658); chloroplast RNA processing (GO:0031425); positive regulation of translation (GO:0045727);
ANE-Chitosan	GLYMA1180S00210	Q8GWE0	-1.10	0.00	-0.91	-1.62	Pentatricopeptide repeat-containing protein, chloroplastic	chloroplast organization (GO:0009658); chloroplast RNA processing (GO:0031425); positive regulation of translation (GO:0045727);
ANE-Chitosan	GLYMA17G12450	Q9SUV1	-2.78	-0.62	-0.94	-1.62	Adenine nucleotide transporter BT1, chloroplastic/mitochondrial	translation (GO:0006412); transmembrane transport (GO:0055085);
ANE	GLYMA05G24110	O49169	-1.22	0.48	-0.95	-0.93	Elongation factor 1-alpha	No-GOBP
ANE	GLYMA11G16460	Q9C641	-1.03	0.98	-0.33	-0.91	Elongation factor G-1, mitochondrial	No-GOBP
ANE	GLYMA13G04050	P29545	-1.00	0.52	-0.54	-0.78	Elongation factor 1-beta	No-GOBP

ANE	GLYMA09G36870	Q38884	-1.01	0.06	-0.74	-0.99	Eukaryotic translation initiation factor 3 subunit I	formation of translation preinitiation complex (GO:0001731); response to salt stress (GO:0009651);
ANE	GLYMA11G00790	A0A0B2 RSF1	-1.42	0.00	0.00	-0.92	Uncharacterized protein	translation (GO:0006412);
ANE	GLYMA16G04890	P35614	-1.68	-0.09	0.00	-0.28	Eukaryotic peptide chain release factor subunit 1-3	regulation of growth (GO:0040008); translational termination (GO:0006415);
ANE	GLYMA17G35890	Q9FXA2	-1.01	0.37	-0.87	-0.87	Polyadenylate-binding protein 8	regulation of translation (GO:0006417); response to cadmium ion (GO:0046686); viral process (GO:0016032);
ANE	GLYMA20G31120	Q05196	-1.29	0.09	-0.39	-0.58	Polyadenylate-binding protein 5	regulation of translation (GO:0006417);

Table 21: ANE down-regulated genes encoding Pentatricopeptide repeat-containing proteins (likely involved in translation regulation).

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA03G38690	Q9SHZ8	-1.48	0.00	-1.43	-1.12	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA04G00910	Q9LTV8	-1.10	0.00	-1.37	-1.13	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA04G06020	Q9SMZ2	-1.97	0.00	-1.09	-1.43	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA06G00940	Q9CAA8	-1.62	0.00	-1.20	-1.28	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA06G12586	O49287	-1.22	0.00	-1.28	-1.50	Putative pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitin-Chitosan	GLYMA06G22850	Q0WN60	-1.86	0.00	-1.37	-1.94	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA07G30720	Q9LG23	-1.62	0.00	-1.17	-1.30	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitin-Chitosan	GLYMA07G38200	Q9SJK9	-1.31	0.00	-1.01	-1.78	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP

ANE-Chitin-Chitosan	GLYMA08G09830	Q9SMZ2	-1.20	0.00	-1.22	-1.22	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA09G02010	Q56XI1	-1.75	0.00	-1.87	-2.06	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA13G38960	Q9MA50	-1.02	0.00	-1.11	-1.63	Pentatricopeptide repeat-containing protein, chloroplastic	RNA modification (GO:0009451);
ANE-Chitin-Chitosan	GLYMA18G04921	Q9FFN1	-2.39	0.00	-1.56	-1.22	Pentatricopeptide repeat-containing protein	embryo development ending in seed dormancy (GO:0009793);
ANE-Chitin-Chitosan	GLYMA19G39670	Q9FMA1	-1.12	0.00	-1.11	-1.15	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA20G36285	Q9FHR3	-1.32	0.00	-1.11	-1.27	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin	GLYMA02G01460	Q9FZ24	-1.85	0.00	-1.43	-0.94	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitin	GLYMA02G07861	Q9SVP7	-1.26	0.00	-1.07	-0.41	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin	GLYMA08G03870	Q3ECB8	-1.20	0.00	-1.16	-0.68	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin	GLYMA08G25345	Q9LUC2	-1.25	0.00	-1.29	-0.46	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin	GLYMA10G01500	Q9FZ24	-1.89	0.00	-1.69	-0.77	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitin	GLYMA12G07220	Q9LQQ1	-1.31	0.24	-1.06	-0.91	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitin	GLYMA16G31949	Q9LQ16	-1.02	-0.25	-1.03	-0.76	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitin	GLYMA17G36970	Q940Z1	-1.39	0.00	-1.17	-0.70	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA03G15860	Q9SHZ8	-1.00	0.00	-0.32	-1.17	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA03G34150	Q9FHR3	-1.11	0.00	-0.55	-1.37	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA04G41420	Q9M3A8	-1.08	-0.13	-0.92	-1.10	Pentatricopeptide repeat-containing protein	embryo development ending in seed dormancy (GO:0009793);
ANE-Chitosan	GLYMA05G27390	Q9ZUU3	-1.06	0.00	-0.54	-1.08	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA05G34000	Q9SY02	-1.13	0.00	-0.77	-1.09	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA06G23620	Q9FM64	-1.03	0.15	-0.19	-1.20	Pentatricopeptide repeat-containing protein, chloroplastic	mRNA modification (GO:0016556);
ANE-Chitosan	GLYMA06G44400	Q9SY75	-1.01	0.00	-0.80	-1.12	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA08G12390	Q9SN39	-1.84	0.00	-0.64	-1.98	Pentatricopeptide repeat-containing protein DOT4, chloroplastic	chloroplast RNA modification (GO:1900865);
ANE-Chitosan	GLYMA08G17040	Q9FK33	-1.05	0.00	-0.51	-1.27	Pentatricopeptide repeat-containing protein, chloroplastic	No-GOBP

ANE-Chitosan	GLYMA08G41430	Q9M2Y7	-1.05	0.00	-0.90	-1.30	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA09G37140	Q9FK93	-1.28	0.00	-0.85	-1.01	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA09G40850	Q9FXB9	-1.07	0.00	-0.59	-1.04	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitosan	GLYMA10G28930	O80488	-1.16	0.13	-0.84	-1.26	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA10G40610	Q9LTV8	-1.42	0.00	-0.45	-1.04	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA11G00960	Q9LUJ4	-1.32	0.16	-0.98	-1.16	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE-Chitosan	GLYMA11G06990	Q9FLZ9	-1.33	0.00	0.00	-1.92	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA15G11730	Q9XE98	-1.33	0.00	-0.83	-1.08	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA15G22730	Q9STE1	-1.23	0.00	-0.57	-1.73	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA16G33110	Q9C501	-1.20	0.00	-0.81	-1.53	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA17G11010	Q9FJY7	-1.21	0.00	-0.19	-1.63	Pentatricopeptide repeat-containing protein	chloroplast RNA modification (GO:1900865);
ANE-Chitosan	GLYMA18G48780	Q1PEU4	-1.63	0.10	-0.50	-1.04	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA18G49710	Q9CA54	-1.07	0.00	-0.98	-1.31	Pentatricopeptide repeat-containing protein	No-GOBP
ANE-Chitosan	GLYMA19G40875	Q9SIT7	-1.09	0.00	-0.50	-1.23	Pentatricopeptide repeat-containing protein	mitochondrial mRNA modification (GO:0080156); sugar mediated signaling pathway (GO:0010182);
ANE-Chitosan	GLYMA20G29500	Q9M1V3	-1.34	0.00	-0.67	-1.41	Pentatricopeptide repeat-containing protein, chloroplastic	mRNA processing (GO:0006397);
ANE	GLYMA02G00270	Q9C977	-1.39	0.43	-0.62	-0.62	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE	GLYMA02G10461	Q9LTV8	-1.26	0.00	-0.49	-0.91	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA03G37041	Q9LSB8	-1.73	0.00	-0.69	-0.70	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA05G05870	Q9LS72	-1.27	0.06	0.00	-0.61	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA07G21700	Q1PFH7	-1.40	0.00	0.00	-0.03	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA07G31620	P93011	-1.20	0.00	0.00	-0.67	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA08G06580	Q9LG23	-1.45	0.14	-0.92	-0.89	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE	GLYMA08G10260	A8MQA3	-1.52	0.00	-0.67	-0.80	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA08G28400	Q9M891	-1.31	0.00	-0.43	-0.44	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE	GLYMA10G00280	Q9C977	-1.15	0.00	-0.55	-0.43	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE	GLYMA10G41080	Q9C9A2	-1.06	0.16	0.00	-0.71	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP

ANE	GLYMA11G11110	Q9SX45	-1.10	0.00	-0.52	-0.94	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA11G12940	Q9LHN5	-1.01	0.00	-0.49	-0.23	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA12G00690	Q9SB36	-1.13	0.00	-0.94	-0.93	Pentatricopeptide repeat-containing protein, chloroplastic	RNA splicing (GO:0008380);
ANE	GLYMA12G22290	Q9SVP7	-1.16	0.00	-0.51	-0.34	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA12G31350	Q9MA50	-1.18	0.03	-0.68	-0.39	Pentatricopeptide repeat-containing protein, chloroplastic	RNA modification (GO:0009451);
ANE	GLYMA13G40750	O23169	-1.53	0.02	-0.86	-0.92	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA14G04900	Q0WP85	-1.05	0.00	0.00	-0.48	Pentatricopeptide repeat-containing protein, mitochondrial	No-GOBP
ANE	GLYMA14G07170	O64705	-1.03	0.00	-0.32	-0.59	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA15G04311	Q940Q2	-1.14	0.00	-0.33	-0.36	Pentatricopeptide repeat-containing protein, mitochondrial	response to cadmium ion (GO:0046686);
ANE	GLYMA16G00280	Q9SY69	-1.33	0.22	-0.68	-0.55	Pentatricopeptide repeat-containing protein	cell division (GO:0051301); transcription, DNA-templated (GO:0006351);
ANE	GLYMA18G46431	Q9LSB8	-1.76	0.00	0.00	-0.02	Putative pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA19G25830	A8MQA3	-2.01	0.00	0.00	-0.89	Pentatricopeptide repeat-containing protein	No-GOBP
ANE	GLYMA19G32350	Q9LTF4	-1.15	0.00	-0.45	-0.90	Putative pentatricopeptide repeat-containing protein	cytidine to uridine editing (GO:0016554);

Table 22: ANE down-regulated genes involved in protein folding and transport.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA14G11420	P05477	-2.61	-1.13	-3.31	-1.21	17.9 kDa class II heat shock protein	No-GOBP
ANE-Alginate-Chitin	GLYMA17G34220	P05477	-1.75	-1.06	-1.28	-0.94	17.9 kDa class II heat shock protein	No-GOBP
ANE-Chitin-Chitosan	GLYMA01G09310	Q9SIF2	-1.14	0.07	-1.45	-1.34	Heat shock protein 90-5, chloroplastic	protein folding (GO:0006457); response to salt stress (GO:0009651); response to water deprivation (GO:0009414);
ANE-Chitin-Chitosan	GLYMA08G43120	P31170	-1.57	-0.65	-1.39	-3.50	25.3 kDa heat shock protein, chloroplastic	response to heat (GO:0009408); response to hydrogen peroxide (GO:0042542);
ANE-Chitin-Chitosan	GLYMA18G10760	P31170	-1.88	0.00	-1.87	-1.65	25.3 kDa heat shock protein, chloroplastic	response to heat (GO:0009408response to hydrogen peroxide (GO:0042542);
ANE-Chitin-Chitosan	GLYMA20G01930	P19242	-1.11	-0.73	-1.34	-1.33	17.1 kDa class II heat shock protein	No-GOBP

ANE-Chitin-Chitosan	GLYMA02G13790	Q9SIF2	-1.28	0.00	-1.62	-1.27	Heat shock protein 90-5, chloroplastic	protein folding (GO:0006457); response to chlorate (GO:0010157); response to heat (GO:0009408); response to salt stress;
ANE-Chitin-Chitosan	GLYMA03G41210	Q38867	-2.53	-0.38	-2.61	-1.50	Peptidyl-prolyl cis-trans isomerase CYP19-3	protein folding (GO:0006457); response to cadmium ion (GO:0046686); signal transduction (GO:0007165);
ANE-Chitin-Chitosan	GLYMA07G30290	Q01899	-1.64	-0.27	-1.50	-1.41	Heat shock 70 kDa protein, mitochondrial	protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA08G06950	Q01899	-1.43	-0.02	-1.19	-1.30	Heat shock 70 kDa protein, mitochondrial	protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA08G47920	Q9M888	-1.40	-0.18	-1.07	-1.08	T-complex protein 1 subunit zeta 1	protein folding (GO:0006457); response to cadmium ion (GO:0046686); response to zinc ion (GO:0010043);
ANE-Chitin-Chitosan	GLYMA13G17260	Q02073	-1.55	-0.29	-2.06	-2.69	20 kDa chaperonin, chloroplastic	positive regulation of superoxide dismutase activity (GO:1901671); protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA15G08420	P59910	-1.41	-0.02	-1.38	-1.93	DnaJ homolog subfamily B member 13	protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA16G00410	Q02028	-1.15	-0.25	-1.48	-1.87	Stromal 70 kDa heat shock-related protein, chloroplastic	protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA17G05230	O65282	-1.56	-0.35	-2.07	-2.52	20 kDa chaperonin, chloroplastic	positive regulation of superoxide dismutase activity (GO:1901671); protein folding (GO:0006457); response to cadmium ion (GO:0046686); response to cold (GO:0009409);
ANE-Chitin-Chitosan	GLYMA19G29841	Q59978	-1.04	0.32	-1.26	-1.61	Protein GrpE	protein folding (GO:0006457);
ANE-Chitin-Chitosan	GLYMA20G02110	P34893	-1.59	-0.16	-2.08	-1.22	10 kDa chaperonin	protein folding (GO:0006457); response to heat (GO:0009408);
ANE-Chitin-Chitosan	GLYMA08G18760	P08927	-1.01	-0.25	-1.26	-1.89	RuBisCO large subunit-binding protein subunit beta, chloroplastic	protein refolding (GO:0042026);
ANE-Chitin-Chitosan	GLYMA08G20560	Q93ZM7	-1.59	0.00	-2.36	-1.29	Chaperonin CPN60-like 2, mitochondrial	protein refolding (GO:0042026); response to cadmium ion (GO:0046686);
ANE-Chitin-Chitosan	GLYMA10G25630	Q05045	-1.58	-0.02	-1.65	-1.20	Chaperonin CPN60-1, mitochondrial	protein refolding (GO:0042026);
ANE-Chitin-Chitosan	GLYMA11G20180	P08926	-1.27	0.00	-1.57	-1.88	RuBisCO large subunit-binding protein subunit alpha, chloroplastic	protein refolding (GO:0042026);
ANE-Chitin-Chitosan	GLYMA15G40110	P08927	-1.55	0.00	-1.87	-1.99	RuBisCO large subunit-binding protein subunit beta, chloroplastic	protein refolding (GO:0042026);
ANE-Chitin-Chitosan	GLYMA20G19980	P29185	-1.88	-0.05	-2.14	-1.78	Chaperonin CPN60-1, mitochondrial	protein refolding (GO:0042026);

ANE-Chitin-Chitosan	GLYMA20G33910	Q05046	-2.16	0.06	-1.75	-1.14	Chaperonin CPN60-2, mitochondrial	protein refolding (GO:0042026);
ANE-Chitin	GLYMA02G47580	F4JFN3	-1.30	0.04	-1.26	-0.61	Heat shock protein 90-6, mitochondrial	protein folding (GO:0006457); response to stress (GO:0006950);
ANE-Chitin	GLYMA03G28120	Q96539	-1.28	0.34	-1.40	-0.49	10 kDa chaperonin	protein folding (GO:0006457);
ANE-Chitin	GLYMA04G30630	F4J9Q6	-1.30	-0.94	-1.55	-0.05	Peptidyl-prolyl cis-trans isomerase FKBP43	protein folding (GO:0006457);
ANE-Chitin	GLYMA07G34380	P34893	-1.39	0.00	-1.27	-0.26	10 kDa chaperonin	protein folding (GO:0006457); response to heat (GO:0009408);
ANE-Chitin	GLYMA10G28890	Q40401	-1.21	-0.43	-1.03	-0.80	Calreticulin	protein folding (GO:0006457);
ANE-Chitin	GLYMA07G01190	Q93ZM7	-1.46	0.00	-1.40	-0.70	Chaperonin CPN60-like 2, mitochondrial	protein refolding (GO:0042026); response to cadmium ion (GO:0046686);
ANE-Chitin	GLYMA10G33680	Q05046	-1.78	0.20	-1.51	-0.87	Chaperonin CPN60-2, mitochondrial	protein refolding (GO:0042026);
ANE-Chitin	GLYMA11G01660	Q93ZG9	-1.44	-0.94	-1.99	-0.40	Peptidyl-prolyl cis-trans isomerase FKBP53	chaperone-mediated protein folding (GO:0061077); nucleosome assembly (GO:0006334);
ANE-Chitin	GLYMA14G01100	F4JFN3	-1.31	0.00	-1.32	-0.82	Heat shock protein 90-6, mitochondrial	protein folding (GO:0006457); response to stress (GO:0006950);
ANE-Chitin	GLYMA19G30860	Q96539	-1.08	0.28	-1.09	-0.80	10 kDa chaperonin	protein folding (GO:0006457);
ANE-Chitin	GLYMA19G43830	Q38867	-1.39	0.00	-1.20	-0.54	Peptidyl-prolyl cis-trans isomerase CYP19-3	protein folding (GO:0006457); response to cadmium ion (GO:0046686);
ANE-Chitin	GLYMA04G05720	P19242	-1.55	-0.10	-1.06	-0.10	17.1 kDa class II heat shock protein	No-GOBP
ANE-Chitosan	GLYMA10G44560	Q9ASS6	-1.02	0.00	-0.99	-1.39	Photosynthetic NDH subunit of lumenal location 5, chloroplastic	NAD(P)H dehydrogenase complex assembly (GO:0010275); protein folding (GO:0006457); transport (GO:0006810);
ANE-Chitosan	GLYMA12G09250	Q940P8	-1.09	0.04	-0.96	-1.05	T-complex protein 1 subunit beta	protein folding (GO:0006457);
ANE	GLYMA08G05470	Q9LV21	-1.07	0.13	-0.38	-0.40	T-complex protein 1 subunit delta	protein folding (GO:0006457); response to cadmium ion (GO:0046686);
ANE	GLYMA08G12970	P28769	-1.02	0.32	-0.48	-0.74	T-complex protein 1 subunit alpha	protein folding (GO:0006457);
ANE	GLYMA11G19220	Q940P8	-1.36	0.18	-0.86	-0.99	T-complex protein 1 subunit beta	protein folding (GO:0006457);
ANE	GLYMA11G37630	O04450	-1.08	0.42	-0.43	-0.27	T-complex protein 1 subunit epsilon	protein folding (GO:0006457);
ANE	GLYMA14G05520	Q9MAU6	-1.05	0.00	-0.76	-0.72	Protein disulfide-isomerase like 2-2	cell redox homeostasis (GO:0045454); protein folding (GO:0006457); response to endoplasmic reticulum stress (GO:0034976);
ANE	GLYMA18G53590	Q8L7N0	-1.04	-0.10	-0.91	-0.91	T-complex protein 1 subunit zeta 2	protein folding (GO:0006457);
ANE	GLYMA07G32050	P05478	-1.10	-0.08	-0.96	-0.40	18.5 kDa class I heat shock protein	No-GOBP
ANE-Alginate-Chitin-Chitosan	GLYMA16G18075	Q8L748	-2.75	-2.70	-2.87	-3.34	Nuclear pore complex protein NUP107	mRNA transport (GO:0051028); protein transport (GO:0015031);

ANE-Chitin-Chitosan	GLYMA11G07661	Q9SI13	-1.15	0.00	-1.85	-1.64	Phosphatidylinositol/phosphatidylcholine transfer protein SFH10	protein transport (GO:0015031);
ANE-Chitin	GLYMA03G19460	Q9XGX8	-1.81	0.00	-1.30	-0.08	Mitochondrial import inner membrane translocase subunit Tim9	protein transport (GO:0015031);
ANE-Chitosan	GLYMA18G36340	F4J7S8	-1.67	0.00	0.00	-1.61	Phosphatidylinositol/phosphatidylcholine transfer protein SFH9	protein transport (GO:0015031);
ANE	GLYMA02G04900	A0A0B2NYX1	-1.40	0.00	0.00	0.00	IST1-like protein	protein transport (GO:0015031);
ANE	GLYMA03G27570	Q9M612	-1.27	0.00	-0.79	-0.71	Nascent polypeptide-associated complex subunit alpha-like protein	protein transport (GO:0015031);
ANE	GLYMA04G37570	Q9XGY4	-1.17	0.18	-0.68	-0.12	Mitochondrial import inner membrane translocase subunit TIM8	protein transport (GO:0015031);
ANE	GLYMA10G36500	Q9LMK7	-1.00	0.08	-0.74	-0.29	Ran-binding protein 1 homolog a	intracellular transport (GO:0046907); mRNA transport (GO:0051028); protein transport (GO:0015031);
ANE	GLYMA12G06770	Q9XGX7	-1.08	0.05	-0.90	-0.06	Mitochondrial import inner membrane translocase subunit Tim9	chaperone-mediated protein transport (GO:0072321); protein import into mitochondrial inner membrane (GO:0045039);
ANE	GLYMA16G12090	Q2PMP0	-1.02	-0.45	-0.43	1.86	Protein TIC 214	protein transport (GO:0015031);
ANE	GLYMA20G21860	Q94CG2	-1.04	-0.08	-0.12	0.00	Bet1-like SNARE 1-2	ER to Golgi vesicle-mediated transport (GO:0006888); protein transport (GO:0015031);

Table 23: ANE down-regulated genes involved in nucleotide and DNA synthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA08G15610	Q38W66	-1.38	0.10	-1.34	-1.68	Uridylate kinase	'de novo' CTP biosynthetic process (GO:0044210);
ANE-Chitin-Chitosan	GLYMA05G36900	O49203	-1.05	0.09	-1.19	-1.33	Nucleoside diphosphate kinase III	CTP biosynthetic process (GO:0006241); UTP biosynthetic process (GO:0006228);
ANE-Chitin-Chitosan	GLYMA10G39305	Q9I0K9	-1.00	-0.11	-1.01	-1.72	Adenylosuccinate lyase	'de novo' AMP biosynthetic process (GO:0044208);'de novo' IMP biosynthetic process (GO:0006189);
ANE-Chitosan	GLYMA04G16670	B7GFU2	-1.03	0.14	-0.93	-1.24	Bifunctional purine biosynthesis protein PurH	'de novo' IMP biosynthetic process (GO:0006189);
ANE-Chitin	GLYMA08G25030	Q8R720	-1.99	-0.21	-1.04	-0.39	CTP synthase	'de novo' CTP biosynthetic process (GO:0044210);
ANE-Chitin	GLYMA18G05501	Q10FF9	-2.90	0.00	-2.46	-0.62	Deoxyuridine 5'-triphosphate nucleotidohydrolase	dUMP biosynthetic process (GO:0006226); dUTP catabolic process (GO:0046081);

ANE	GLYMA05G37760	O66601	-1.20	0.00	-0.66	-0.69	GMP synthase [glutamine-hydrolyzing]	GMP biosynthetic process (GO:0006177);
ANE	GLYMA08G01820	O66601	-1.04	0.28	-0.59	-0.41	GMP synthase [glutamine-hydrolyzing]	GMP biosynthetic process (GO:0006177);
ANE	GLYMA02G46290	Q43086	-1.30	0.00	-0.77	-0.78	Aspartate carbamoyltransferase 1, chloroplastic	'de novo' UMP biosynthetic process (GO:0044205);
ANE	GLYMA03G25820	Q39839	-1.00	0.62	-0.66	-0.59	Nucleoside diphosphate kinase 1	CTP biosynthetic process (GO:0006241); GTP biosynthetic process (GO:0006183); UTP biosynthetic process (GO:0006228);
ANE-Chitin-Chitosan	GLYMA01G39430	Q9FHA3	-1.32	0.00	-1.10	-1.15	DNA polymerase alpha catalytic subunit	DNA replication initiation (GO:0006270);
ANE-Chitin-Chitosan	GLYMA10G11610	P30182	-1.08	-0.54	-1.40	-1.42	DNA topoisomerase 2	DNA topological change (GO:0006265); mitotic DNA integrity checkpoint (GO:0044774);
ANE-Chitin-Chitosan	GLYMA14G18300	Q1RIM1	-1.80	0.00	-2.13	-2.16	DNA topoisomerase 1	DNA topological change (GO:0006265);
ANE-Chitin-Chitosan	GLYMA02G45610	P20664	-2.92	0.00	-1.67	-1.04	DNA primase small subunit	No-GOBP
ANE-Chitin	GLYMA03G37770	P43299	-2.87	0.00	-1.97	-0.27	DNA replication licensing factor MCM7	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268); sugar mediated signaling pathway (GO:0010182);
ANE-Chitin	GLYMA04G39990	Q501D5	-1.52	0.00	-1.30	0.00	Mini-chromosome maintenance complex-binding protein	cell division (GO:0051301); DNA replication (GO:0006260); mitotic nuclear division (GO:0007067);
ANE-Chitin	GLYMA06G14860	Q501D5	-2.01	0.00	-1.79	-0.18	Mini-chromosome maintenance complex-binding protein	cell division (GO:0051301); DNA replication (GO:0006260); mitotic nuclear division (GO:0007067);
ANE-Chitin	GLYMA08G11130	Q84WJ2	-1.01	0.00	-1.13	-0.53	Probable DNA primase large subunit	No-GOBP
ANE-Chitin	GLYMA02G10605	Q9FNE9	-1.47	0.00	-2.09	-0.35	Histone-lysine N-methyltransferase ATXR6	histone H3-K27 methylation (GO:0070734); regulation of cell cycle (GO:0051726); regulation of DNA replication (GO:0006275);
ANE-Chitin	GLYMA05G25980	Q43704	-1.92	0.00	-1.91	-0.66	DNA replication licensing factor MCM3 homolog 1	cell cycle (GO:0007049); DNA replication initiation (GO:0006270);
ANE-Chitin	GLYMA07G36680	Q2R482	-2.73	0.00	-2.50	-0.67	DNA replication licensing factor MCM2	cell proliferation (GO:0008283);cytokinesis by cell plate formation (GO:0000911); (GO:0042127);regulation of DNA replication (GO:0006275);regulation of flower development (GO:0009909);
ANE-Chitin	GLYMA09G05240	Q6F353	-2.32	-0.20	-2.56	-0.86	DNA replication licensing factor MCM6	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268);
ANE-Chitin	GLYMA11G12110	Q0WVF5	-2.29	0.00	-2.15	-0.98	DNA replication licensing factor MCM4	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268);

ANE-Chitin	GLYMA12G04320	Q5JKB0	-1.28	0.00	-2.03	-0.53	DNA replication licensing factor MCM4	cytokinesis by cell plate formation (GO:0000911);DNA methylation (GO:0006306);DNA replication initiation (GO:0006270);DNA unwinding involved in DNA replication (GO:0006268); regulation of flower development (GO:0009909);
ANE-Chitin	GLYMA13G22420	O80786	-2.13	0.00	-2.19	-0.78	DNA replication licensing factor MCM5	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268);
ANE-Chitin	GLYMA17G11220	O80786	-1.61	0.00	-2.15	-0.44	DNA replication licensing factor MCM5	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268);
ANE-Chitin	GLYMA04G05870	Q8I7P9	-1.14	-0.78	-2.01	-0.12	Retrovirus-related Pol polyprotein from transposon opus	DNA integration (GO:0015074); RNA-dependent DNA replication (GO:0006278); transposition, DNA-mediated (GO:0006313);
ANE-Chitin	GLYMA05G00370	Q9FME0	-1.95	0.46	-2.02	-0.69	Replication protein A 70 kDa DNA-binding subunit D	DNA recombination (GO:0006310); DNA repair (GO:0006281); DNA replication (GO:0006260);
ANE-Chitin	GLYMA19G28126	Q7LHG5	-1.52	0.57	-2.38	-0.27	Transposon Ty3-I Gag-Pol polyprotein	DNA biosynthetic process (GO:0071897); RNA-dependent DNA replication (GO:0006278); viral release from host cell (GO:0019076);
ANE-Chitin	GLYMA06G15350	Q9SJ20	-2.13	0.07	-1.65	-0.57	Ribonucleoside-diphosphate reductase large subunit	deoxyribonucleoside triphosphate biosynthetic process (GO:0009202); DNA replication (GO:0006260);
ANE-Chitin	GLYMA16G06070	P49730	-2.76	0.25	-1.61	-0.89	Ribonucleoside-diphosphate reductase small chain	deoxyribonucleoside diphosphate metabolic process (GO:0009186); DNA replication (GO:0006260);
ANE-Chitosan	GLYMA08G46385	Q55GA4	-1.20	0.00	-0.95	-1.94	Cell division control protein 45 homolog	cell cycle (GO:0007049); DNA replication initiation (GO:0006270);
ANE-Chitosan	GLYMA20G14210	O81263	-1.63	0.00	-0.64	-1.17	Thymidine kinase	DNA biosynthetic process (GO:0071897);
ANE	GLYMA08G08920	Q9FL33	-1.17	0.00	-0.30	-0.40	DNA replication licensing factor MCM3	cell cycle (GO:0007049); DNA replication initiation (GO:0006270); DNA unwinding involved in DNA replication (GO:0006268);
ANE	GLYMA18G07444	O82387	-1.76	0.00	0.00	-0.02	Cell division control protein 6 homolog	cell division (GO:0051301); DNA replication initiation (GO:0006270);
ANE	GLYMA01G05540	Q9ZVH3	-1.05	0.00	-0.41	-0.39	Origin of replication complex subunit 6	DNA replication (GO:0006260); pollen development (GO:0009555);
ANE	GLYMA15G43170	O24308	-1.24	-0.36	-0.94	-0.68	DNA topoisomerase 2	DNA topological change (GO:0006265);
ANE	GLYMA18G01290	A0A0B2QRT3	-1.24	0.00	-0.75	0.00	DNA polymerase alpha subunit B	DNA replication (GO:0006260);

ANE-Alginate-Chitin-Chitosan	GLYMA07G18400	P10978	-2.47	-1.79	-2.48	-3.26	Retrovirus-related Pol polyprotein from transposon TNT 1-94	DNA integration (GO:0015074);
ANE-Alginate-Chitin	GLYMA08G16220	Q00874	-1.33	-1.17	-1.89	3.38	DNA-damage-repair/toleration protein DRT100	DNA repair (GO:0006281);
ANE-Alginate-Chitin	GLYMA1029S00200	P04323	-1.09	-1.23	-1.81	-0.06	Retrovirus-related Pol polyprotein from transposon 17.6	DNA integration (GO:0015074);
ANE-Alginate-Chitin	GLYMA13G41650	Q00874	-1.59	-1.69	-3.47	1.43	DNA-damage-repair/toleration protein DRT100	DNA repair (GO:0006281);
ANE-Chitin-Chitosan	GLYMA02G47240	I1JJL9	-1.55	-0.37	-1.77	-1.41	Uncharacterized protein	regulation of DNA repair (GO:0006282);
ANE-Chitin-Chitosan	GLYMA07G10400	Q96PM5	-1.37	0.90	-1.06	-1.30	RING finger and CHY zinc finger domain-containing protein 1	DNA repair (GO:0006281);
ANE-Chitin-Chitosan	GLYMA08G25841	P10978	-1.19	0.00	-1.76	-2.19	Retrovirus-related Pol polyprotein from transposon TNT 1-94	DNA integration (GO:0015074);
ANE-Chitin-Chitosan	GLYMA10G31180	P10978	-1.13	-0.07	-1.75	-1.91	Retrovirus-related Pol polyprotein from transposon TNT 1-94	DNA integration (GO:0015074);
ANE-Chitin-Chitosan	GLYMA18G17515	P10978	-1.09	0.00	-1.32	-1.53	Retrovirus-related Pol polyprotein from transposon TNT 1-94	DNA integration (GO:0015074);
ANE-Chitin-Chitosan	GLYMA11G03750	F4JP48	-1.42	-0.26	-1.28	-1.02	DNA mismatch repair protein MSH4	meiotic mismatch repair involved in reciprocal meiotic recombination (GO:0010777);
ANE-Chitin	GLYMA06G42935	P04146	-1.71	0.00	-1.33	0.77	Copia protein	DNA integration (GO:0015074);
ANE-Chitin	GLYMA14G38871	Q9STG6	-1.82	0.00	-1.62	-0.96	Deoxyuridine 5'-triphosphate nucleotidohydrolase	DNA repair (GO:0006281); dUMP biosynthetic process (GO:0006226); dUTP catabolic process (GO:0046081);
ANE-Chitin	GLYMA15G22450	P10978	-1.25	0.00	-1.41	0.00	Retrovirus-related Pol polyprotein from transposon TNT 1-94	DNA integration (GO:0015074);
ANE	GLYMA04G33750	Q8LC68	-1.17	0.00	-1.00	-0.14	NAP1-related protein 2	nucleosome assembly (GO:0006334); somatic cell DNA recombination (GO:0016444);
ANE	GLYMA06G20700	Q8LC68	-1.10	-0.08	-0.99	-0.27	NAP1-related protein 2	nucleosome assembly (GO:0006334); somatic cell DNA recombination (GO:0016444);
ANE	GLYMA17G10770	Q8LC68	-1.42	0.22	-0.79	-0.06	NAP1-related protein 2	nucleosome assembly (GO:0006334); somatic cell DNA recombination (GO:0016444);

Table 24: ANE down-regulated genes involved in fatty acids metabolism.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA17G14130	Q9S746	-1.49	-2.64	-2.57	1.10	Protein HOTHEAD	fatty acid omega-oxidation (GO:0010430);
ANE-Chitin-Chitosan	GLYMA05G25970	P52410	-1.35	0.00	-1.24	-1.17	3-oxoacyl-[acyl-carrier-protein] synthase I, chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE-Chitin-Chitosan	GLYMA08G01180	P52414	-1.07	-0.31	-1.65	-2.23	Acyl carrier protein 4, chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE-Chitin-Chitosan	GLYMA09G38440	Q42783	-1.28	0.08	-1.06	-1.49	Biotin carboxyl carrier protein of acetyl-CoA carboxylase, chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE-Chitin-Chitosan	GLYMA12G03060	Q9SLA8	-1.45	0.00	-1.84	-1.54	Enoyl-[acyl-carrier-protein] reductase [NADH], chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE-Chitin	GLYMA08G45990	P80030	-2.18	-0.37	-1.75	-0.42	Enoyl-[acyl-carrier-protein] reductase [NADH], chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE-Chitin	GLYMA18G31780	P80030	-1.13	0.00	-1.15	-0.15	Enoyl-[acyl-carrier-protein] reductase [NADH], chloroplastic	fatty acid biosynthetic process (GO:0006633);
ANE	GLYMA04G20620	O48780	-2.18	-0.72	-0.82	0.00	3-ketoacyl-CoA synthase 11	fatty acid biosynthetic process (GO:0006633);
ANE	GLYMA09G01630	Q9FGJ4	-1.52	0.00	-0.91	-0.78	Acyl carrier protein 3, mitochondrial	fatty acid biosynthetic process (GO:0006633);
ANE	GLYMA10G38670	Q9XF43	-1.24	-0.12	-0.82	-0.76	3-ketoacyl-CoA synthase 6	fatty acid biosynthetic process (GO:0006633); wax biosynthetic process (GO:0010025);

Table 25: ANE down-regulated genes involved in lipid catabolic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA07G01680	Q9LU14	-1.11	-1.86	-4.55	1.22	GDSL esterase/lipase APG	lipid catabolic process (GO:0016042);

ANE-Alginate-Chitin	GLYMA18G13540	Q67ZI9	-1.55	-1.09	-2.13	1.40	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE-Alginate-Chitin	GLYMA09G41460	Q9M5X8	-2.30	-2.79	-2.37	0.90	Non-specific lipid-transfer protein	lipid transport (GO:0006869);
ANE-Alginate-Chitin	GLYMA13G22940	Q9SU35	-4.37	-2.85	-5.15	1.21	pEARLI1-like lipid transfer protein 1	cellular response to cold (GO:0070417); cold acclimation (GO:0009631); defense response to fungus (GO:0050832); induced systemic resistance (GO:0009682); systemic acquired resistance (GO:0009627);
ANE-Alginate-Chitin	GLYMA17G32100	Q9SU35	-1.86	-2.68	-4.23	0.27	pEARLI1-like lipid transfer protein 1	cellular response to cold (GO:0070417); cold acclimation (GO:0009631); defense response to fungus (GO:0050832);
ANE-Alginate	GLYMA13G30680	Q9FJ41	-1.15	-2.10	0.00	0.00	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE-Alginate	GLYMA17G37900	Q94CH6	-1.47	-1.28	0.06	3.66	GDSL esterase/lipase EXL3	lipid catabolic process (GO:0016042);
ANE-Chitin-Chitosan	GLYMA16G21970	Q9ASU1	-1.20	0.00	-1.12	-1.67	Diacylglycerol O-acyltransferase 2	neutral lipid biosynthetic process (GO:0046460); triglyceride biosynthetic process (GO:0019432);
ANE-Chitin-Chitosan	GLYMA16G07450	Q9LY84	-1.40	0.00	-1.93	-1.61	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE-Chitin-Chitosan	GLYMA01G00541	Q8S8N6	-1.21	-0.65	-1.47	-1.17	Phospholipase A2-alpha	lipid catabolic process (GO:0016042);
ANE-Chitin-Chitosan	GLYMA03G24890	Q9M903	-1.31	0.00	-1.42	-1.09	Protein TRIGALACTOSYLDIACYLGLYCEROL 4, chloroplastic	acylglycerol transport (GO:0034196); ER to chloroplast lipid transport (GO:1990052);
ANE-Chitin-Chitosan	GLYMA07G12970	Q9M903	-2.08	0.00	-2.58	-1.99	Protein TRIGALACTOSYLDIACYLGLYCEROL 4, chloroplastic	ER to chloroplast lipid transport (GO:1990052);
ANE-Chitin-Chitosan	GLYMA04G41510	Q9LV04	-1.28	0.91	-1.50	-1.10	Probable plastid-lipid-associated protein 14, chloroplastic	No-GOBP
ANE-Chitin	GLYMA02G11820	Q3EBF7	-1.82	-0.05	-1.94	0.00	Delta (8)-fatty-acid desaturases 2	cellular response to cold (GO:0070417); sphingolipid biosynthetic process (GO:0030148);
ANE-Chitin	GLYMA19G29810	Q9FXB6	-1.69	-0.02	-1.23	0.00	GDSL esterase/lipase LIP-4	lipid catabolic process (GO:0016042);
ANE-Chitin	GLYMA19G41470	Q9FXB6	-1.27	0.00	-1.46	-0.80	GDSL esterase/lipase LIP-4	lipid catabolic process (GO:0016042);
ANE-Chitin	GLYMA09G02550	O80959	-1.58	0.00	-1.12	-0.58	Patatin-like protein 6	defense response (GO:0006952); lipid catabolic process (GO:0016042);
ANE-Chitin	GLYMA07G15580	Q8S8N6	-1.07	-0.55	-2.24	-0.47	Phospholipase A2-alpha	lipid catabolic process (GO:0016042);
ANE-Chitosan	GLYMA03G28690	O48917	-1.01	0.55	-0.35	-1.52	UDP-sulfoquinovose synthase, chloroplastic	cellular response to phosphate starvation (GO:0016036); glycolipid biosynthetic process (GO:0009247);
ANE-Chitosan	GLYMA19G31430	O48917	-1.07	0.30	-0.79	-1.45	UDP-sulfoquinovose synthase, chloroplastic	cellular response to phosphate starvation (GO:0016036); glycolipid biosynthetic process (GO:0009247);

ANE	GLYMA02G43440	Q9SJB4	-1.68	-0.10	0.21	1.03	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE	GLYMA14G05550	Q9SJB4	-1.05	-0.40	-0.28	1.20	GDSL esterase/lipase	lipid catabolic process (GO:0016042);
ANE	GLYMA15G13460	Q9SV43	-1.12	0.00	-0.56	0.68	Patatin-like protein 7	lipid catabolic process (GO:0016042); response to other organism (GO:0051707);
ANE	GLYMA18G41590	Q8W453	-1.22	1.26	0.00	0.68	Putative lipid-transfer protein DIR1	lipid transport (GO:0006869); systemic acquired resistance, salicylic acid mediated signaling pathway (GO:0009862);

Table 26: ANE down-regulated genes involved in protein degradation.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA01G11870	P01070	-1.26	-0.85	-2.16	-1.85	Trypsin inhibitor A	No-GOBP
ANE-Chitin-Chitosan	GLYMA08G45540	P01070	-1.53	-0.70	-1.73	-1.31	Trypsin inhibitor A	No-GOBP
ANE-Chitin	GLYMA08G45550	P01070	-2.05	-0.57	-2.60	-0.22	Trypsin inhibitor A	No-GOBP
ANE-Chitin	GLYMA08G45560	P01070	-1.99	-0.66	-1.00	-0.72	Trypsin inhibitor A	No-GOBP
ANE	GLYMA08G45590	P01070	-3.67	-0.48	-0.57	-0.79	Trypsin inhibitor A	No-GOBP

Table 27: ANE down-regulated genes associated with somatic storage proteins.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA07G01740	P15490	-1.71	1.93	-1.83	-2.30	Stem 28 kDa glycoprotein	No-GOBP
ANE-Chitin	GLYMA07G01730	P10742	-2.12	2.69	-2.60	-0.61	Stem 31 kDa glycoprotein	No-GOBP
ANE-Chitin	GLYMA08G21410	P10743	-3.26	3.02	-2.50	-0.89	Stem 31 kDa glycoprotein	No-GOBP
ANE-Chitin	GLYMA15G07850	P24805	-2.17	-0.76	-2.10	0.00	Stem-specific protein TSJT1	No-GOBP

Table 28: ANE down-regulated genes involved in cell division.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA06G04575	P42753	-1.52	-1.28	-1.67	0.37	Cyclin-D3-1	cell division (GO:0051301); G1/S transition of mitotic cell cycle (GO:0000082); response to sucrose (GO:0009744); seed development (GO:0048316);
ANE-Alginate-Chitin	GLYMA09G03300	Q94C59	-1.16	-1.00	-1.20	-0.67	Patellin-4	cell cycle (GO:0007049); cell division (GO:0051301);
ANE-Alginate	GLYMA06G44460	F4I2H7	-1.08	-1.10	-0.91	-0.48	Protein TPX2	cell division (GO:0051301); regulation of mitotic spindle organization (GO:0060236);
ANE-Chitin-Chitosan	GLYMA02G05170	Q9SD20	-1.16	-0.47	-1.76	-1.03	Protein POLLENLESS 3-LIKE 2	cell cycle (GO:0007049); cell division (GO:0051301);
ANE-Chitin-Chitosan	GLYMA03G29760	O48533	-1.37	-0.57	-2.21	-1.13	Protein POLYCHOME	cell division (GO:0051301); defense response (GO:0006952); positive regulation of defense response to bacterium (GO:1900426);
ANE-Chitin-Chitosan	GLYMA03G36021	A7UL74	-1.18	-0.47	-1.30	-2.12	Protein CHROMOSOME TRANSMISSION FIDELITY 7	anther development (GO:0048653);cell division (GO:0051301);
ANE-Chitin-Chitosan	GLYMA18G43920	Q9SCU1	-1.49	-0.79	-1.85	-1.42	Patellin-6	cell cycle (GO:0007049); cell division (GO:0051301); transport (GO:0006810);
ANE-Chitin-Chitosan	GLYMA18G10030	Q42545	-1.24	-0.38	-1.81	-1.18	Cell division protein FtsZ homolog 1, chloroplastic	cell cycle (GO:0007049); chloroplast fission (GO:0010020); chloroplast organization (GO:0009658); chloroplast relocation (GO:0009902);
ANE-Chitin-Chitosan	GLYMA17G05042	Q8LNZ2	-1.84	-0.37	-1.02	-1.97	Kinesin-like protein NACK2	cytokinesis by cell plate formation (GO:0000911); pollen development (GO:0009555); radial microtubular system formation (GO:0010245);
ANE-Chitin	GLYMA01G36430	P30278	-1.01	-0.21	-1.53	-0.40	G2/mitotic-specific cyclin-2	cell division (GO:0051301); mitotic nuclear division (GO:0007067);
ANE-Chitin	GLYMA02G37560	Q6YXH8	-1.13	-0.68	-1.36	-0.88	Cyclin-D4-1	cell cycle (GO:0007049); cell division (GO:0051301);
ANE-Chitin	GLYMA07G18500	M1CZC0	-1.39	0.03	-1.05	-0.67	ERBB-3 BINDING PROTEIN 1	auxin-activated signaling pathway (GO:0009734); cell cycle G1/S phase transition (GO:0044843); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA10G02745	O82266	-1.53	-0.05	-1.28	-0.72	Protein SLOW WALKER 1	cell cycle (GO:0007049); embryo sac development (GO:0009553); megagametogenesis (GO:0009561); rRNA processing (GO:0006364);
ANE-Chitin	GLYMA11G05560	Q9FGQ6	-1.93	-0.25	-1.46	-0.34	Microtubule-associated protein RP/EB family member 1C	cell division (GO:0051301); cortical cytoskeleton organization (GO:0030865); mitotic nuclear division (GO:0007067);
ANE-Chitin	GLYMA14G16130	Q147G5	-1.58	0.00	-1.02	0.00	Cyclin-A2-2	cell cycle (GO:0007049); cell division (GO:0051301); positive regulation of stomatal complex development (GO:2000123);
ANE-Chitin	GLYMA15G14220	Q94C59	-1.05	-0.80	-1.06	0.00	Patellin-4	cell cycle (GO:0007049); cell division (GO:0051301);

ANE-Chitin	GLYMA16G27361	Q689D6	-1.02	0.00	-1.33	-0.03	Protein ROOT PRIMORDIUM DEFECTIVE 1	lateral root morphogenesis (GO:0010102); negative regulation of cell proliferation (GO:0008285);
ANE-Chitin	GLYMA12G31730	Q27IK7	-1.24	-0.81	-1.53	-0.73	Phragmoplast orienting kinesin-1	cytokinesis by cell plate formation (GO:0000911); microtubule-based movement (GO:0007018);
ANE-Chitin	GLYMA17G10720	Q9FGQ6	-1.31	0.00	-1.31	0.49	Microtubule-associated protein RP/EB family member 1C	cell division (GO:0051301); cortical cytoskeleton organization (GO:0030865); mitotic nuclear division (GO:0007067);
ANE-Chitosan	GLYMA11G31850	Q689D6	-1.33	0.00	-0.75	-1.30	Protein ROOT PRIMORDIUM DEFECTIVE 1	lateral root morphogenesis (GO:0010102); negative regulation of cell proliferation (GO:0008285);
ANE	GLYMA01G39720	Q9FGQ6	-1.27	0.00	-0.17	-0.51	Microtubule-associated protein RP/EB family member 1C	cell division (GO:0051301); cortical cytoskeleton organization (GO:0030865); mitotic nuclear division (GO:0007067);
ANE	GLYMA02G17050	O82266	-1.84	0.02	-0.60	-0.77	Protein SLOW WALKER 1	cell cycle (GO:0007049); cell division (GO:0051301); megagametogenesis (GO:0009561); rRNA processing (GO:0006364);
ANE	GLYMA04G04620	Q9FMH5	-2.31	0.00	-0.47	-0.96	Putative cyclin-A3-1	cell cycle (GO:0007049); cell division (GO:0051301);
ANE	GLYMA11G34060	Q8LPL5	-1.02	-0.14	-0.70	-0.84	Protein FIZZY-RELATED 3	cell division (GO:0051301); mitotic nuclear division (GO:0007067); protein ubiquitination (GO:0016567);
ANE	GLYMA13G05430	Q9FJL0	-1.08	0.00	-0.49	-0.62	Structural maintenance of chromosomes protein 4	cell division (GO:0051301); mitotic chromosome condensation (GO:0007076);
ANE	GLYMA17G18360	Q9FGQ7	-1.17	0.00	-0.98	-0.67	Cyclin-D3-2	cell cycle (GO:0007049); cell division (GO:0051301); seed development (GO:0048316);
ANE	GLYMA19G06110	A0A0B2S 5H5	-1.21	0.00	-0.96	-0.69	Cell division cycle-associated 7-like protein	cell division (GO:0051301);
ANE	GLYMA14G39760	Q8LG64	-1.04	0.00	-0.83	-0.12	Cyclin-dependent kinase B2-2	hormone-mediated signaling pathway (GO:0009755); regulation of G2/M transition of mitotic cell cycle (GO:0010389); regulation of meristem structural organization (GO:0009934);
ANE	GLYMA07G14830	Q689D6	-2.15	0.00	0.00	-0.62	Protein ROOT PRIMORDIUM DEFECTIVE 1	lateral root morphogenesis (GO:0010102); negative regulation of cell proliferation (GO:0008285);
ANE	GLYMA18G10090	Q9FLP0	-1.30	0.00	0.00	0.00	65-kDa microtubule-associated protein 1	cortical microtubule organization (GO:0043622); cytokinesis (GO:0000910); mitotic nuclear division (GO:0007067);
ANE	GLYMA13G38700	Q27IK7	-1.15	0.00	-0.49	-0.42	Phragmoplast orienting kinesin-1	cytokinesis by cell plate formation (GO:0000911); microtubule-based movement (GO:0007018);
ANE	GLYMA07G02400	Q2V419	-1.08	0.00	-0.90	0.00	Cyclin-dependent kinase B1-2	regulation of cell cycle (GO:0051726);

Table 29: ANE down-regulated genes involved in pollen tube growth.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA06G46350	Q00624	-2.07	-2.26	-2.68	1.25	L-ascorbate oxidase homolog	Probably involved in pollen tube growth;
ANE-Alginate-Chitin	GLYMA12G10420	P29162	-2.19	-1.21	-2.42	1.05	L-ascorbate oxidase homolog	Probably involved in pollen tube growth
ANE-Alginate-Chitin	GLYMA18G47700	Q9FFD5	-1.40	-1.36	-1.17	0.00	CRIB domain-containing protein RIC4	pollen tube growth (GO:0009860); regulation of actin filament polymerization (GO:0030833);
ANE-Alginate	GLYMA14G04530	Q40588	-1.23	-1.19	-0.83	-0.65	L-ascorbate oxidase	Probably involved in pollen tube growth
ANE-Chitin-Chitosan	GLYMA11G06290	Q00624	-1.92	-0.98	-1.69	-1.05	L-ascorbate oxidase homolog	Probably involved in pollen tube growth
ANE-Chitin-Chitosan	GLYMA16G04770	Q9SQG2	-1.42	0.00	-2.83	-1.09	Apyrase 1	pollen germination (GO:0009846);
ANE-Chitin-Chitosan	GLYMA14G37680	Q500U8	-1.77	-1.00	-2.81	-2.40	Tetraketide alpha-pyrone reductase 1	pollen development (GO:0009555); pollen exine formation (GO:0010584);
ANE-Chitin-Chitosan	GLYMA07G02250	Q9CA89	-1.44	-0.16	-1.99	-1.39	Rop guanine nucleotide exchange factor 12	pollen tube growth (GO:0009860); positive regulation of GTPase activity (GO:0043547); regulation of pollen tube growth (GO:0080092);
ANE-Chitin	GLYMA04G02140	Q00624	-1.37	-0.22	-1.52	0.12	L-ascorbate oxidase homolog	Probably involved in pollen tube growth
ANE-Chitin	GLYMA14G39880	Q00624	-1.05	0.00	-1.02	0.00	L-ascorbate oxidase homolog	Probably involved in pollen tube growth
ANE-Chitosan	GLYMA15G16940	Q9FLC6	-1.33	0.00	-0.57	-1.92	RING-H2 finger protein ATL73	pollen development (GO:0009555); regulation of jasmonic acid biosynthetic process (GO:0080141); stamen development (GO:0048443);

Table 30: ANE down-regulated genes involved in cell wall-related processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA05G15700	Q9FH92	-1.23	-1.68	-2.33	-1.29	Protein IRX15-LIKE	plant-type secondary cell wall biogenesis (GO:0009834);

ANE-Alginate-Chitin-Chitosan	GLYMA12G06730	Q38865	-2.14	-1.38	-3.05	-1.92	Expansin-A6	plant-type cell wall organization (GO:0009664);
ANE-Alginate-Chitin	GLYMA01G40460	Q8LF99	-2.10	-2.06	-3.29	0.26	Probable xyloglucan endotransglucosylase/hydrolase protein 6	cell wall biogenesis (GO:0042546); ce response to water deprivation (GO:0009414); xyloglucan metabolic process (GO:0010411);
ANE-Alginate-Chitin	GLYMA04G41460	Q9FK05	-1.24	-3.14	-1.77	1.11	Probable pectinesterase/pectinesterase inhibitor 61	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);
ANE-Alginate-Chitin	GLYMA06G03770	Q9S7G2	-1.25	-1.11	-1.10	-0.57	Probable galacturonosyltransferase-like 2	cell wall organization (GO:0071555); pectin biosynthetic process (GO:0045489);
ANE-Alginate-Chitin	GLYMA07G05660	Q84WP6	-1.03	-1.10	-1.77	0.00	NAC domain-containing protein 43	fruit dehiscence (GO:0010047); plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Alginate-Chitin	GLYMA07G15120	Q9LHF1	-1.76	-1.46	-1.41	0.11	Leucine-rich repeat extensin-like protein 4	cell wall organization (GO:0071555);
ANE-Alginate-Chitin	GLYMA08G03610	Q9LUI1	-1.52	-1.29	-1.74	0.34	Leucine-rich repeat extensin-like protein 6	cell wall organization (GO:0071555);
ANE-Alginate-Chitin	GLYMA12G32390	Q8LDW9	-1.40	-1.11	-2.13	-0.26	Xyloglucan endotransglucosylase/hydrolase protein 9	cell wall biogenesis (GO:0042546); cell wall organization (GO:0071555); xyloglucan metabolic process (GO:0010411);
ANE-Alginate-Chitin	GLYMA16G22680	Q09023	-1.26	-1.02	-3.69	-0.72	Endochitinase CH25	cell wall macromolecule catabolic process (GO:0016998); chitin catabolic process (GO:0006032);
ANE-Alginate-Chitin	GLYMA19G02810	Q38864	-1.50	-2.85	-4.19	2.43	Expansin-A5	plant-type cell wall organization (GO:0009664); primary root development (GO:0080022);
ANE-Alginate-Chitin	GLYMA19G36280	Q9ZQC6	-1.03	-1.42	-1.72	-0.85	Probable beta-1,4-xylosyltransferase IRX9	cell wall organization (GO:0071555); glucuronoxylan biosynthetic process (GO:0010417);
ANE-Alginate-Chitin	GLYMA19G42360	O81635	-1.46	-1.41	-2.66	-0.58	Kinesin-4	microtubule-based movement (GO:0007018);
ANE-Alginate-Chitin	GLYMA13G40960	Q9SN95	-1.66	-2.52	-2.67	0.52	UDP-glucuronic acid decarboxylase 5	D-xylose metabolic process (GO:0042732); UDP-D-xylose biosynthetic process (GO:0033320);
ANE-Alginate-Chitin	GLYMA16G00330	Q6NMK1	-1.22	-1.75	-2.61	-0.83	Glucuronoxylan 4-O-methyltransferase 1	xylan biosynthetic process (GO:0045492); xylan metabolic process (GO:0045491);
ANE-Alginate-Chitin	GLYMA04G43510	Q9T0F7	-1.27	-1.01	-1.62	-0.25	Glucuronoxylan 4-O-methyltransferase 2	xylan biosynthetic process (GO:0045492); xylan metabolic process (GO:0045491);
ANE-Alginate-Chitosan	GLYMA07G23250	Q7XZU3	-1.01	-1.48	0.00	-1.24	Phosphoinositide phosphatase SAC1	cytoskeleton organization (GO:0007010); phosphatidylinositol-3-phosphate biosynthetic process (GO:0036092);
ANE-Alginate	GLYMA06G47200	Q9FF77	-2.01	-1.37	-0.35	0.32	Probable pectinesterase/pectinesterase inhibitor 47	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);

ANE-Alginate	GLYMA15G15370	Q9FGY1	-1.57	-1.53	-0.90	0.01	Beta-D-xylosidase 1	arabinan catabolic process (GO:0031222); seed coat development (GO:0010214); xylan catabolic process (GO:0045493);
ANE-Alginate	GLYMA13G19580	P82266	-1.54	-1.11	-0.78	0.00	Kinesin-like protein KIN-5C	microtubule-based movement (GO:0007018);
ANE-Chitin-Chitosan	GLYMA01G20460	Q9S7Y7	-1.79	0.00	-1.37	-1.54	Alpha-xylosidase 1	cell wall organization (GO:0071555); response to cadmium ion (GO:0046686); xyloglucan metabolic process (GO:0010411);
ANE-Chitin-Chitosan	GLYMA02G03420	O48676	-1.55	0.00	-2.17	-2.32	UDP-glycosyltransferase 74B1	defense response by callose deposition in cell wall (GO:0052544);
ANE-Chitin-Chitosan	GLYMA05G32370	Q9LSB1	-1.21	-0.84	-1.63	-2.24	UDP-glucuronate: xylan alpha-glucuronosyltransferase 1	plant-type secondary cell wall biogenesis (GO:0009834); xylan biosynthetic process (GO:0045492);
ANE-Chitin-Chitosan	GLYMA08G39340	Q949Z1	-2.36	-0.09	-2.61	-1.98	Polygalacturonase	carbohydrate metabolic process (GO:0005975); cell wall organization (GO:0071555);
ANE-Chitin-Chitosan	GLYMA09G03620	Q949Z1	-1.61	-0.63	-1.98	-1.52	Polygalacturonase	carbohydrate metabolic process (GO:0005975); cell wall organization (GO:0071555);
ANE-Chitin-Chitosan	GLYMA15G14540	Q949Z1	-1.06	-0.06	-1.31	-2.03	Polygalacturonase	carbohydrate metabolic process (GO:0005975); cell wall organization (GO:0071555);
ANE-Chitin-Chitosan	GLYMA01G03760	P92990	-1.80	-0.35	-2.03	-2.00	Polygalacturonase 1 beta-like protein 3	No-GOBP
ANE-Chitin-Chitosan	GLYMA02G03960	P92990	-2.24	-0.16	-2.61	-1.92	Polygalacturonase 1 beta-like protein 3	No-GOBP
ANE-Chitin-Chitosan	GLYMA15G26790	Q9M5J9	-2.26	0.00	-1.44	-2.65	Polygalacturonase inhibitor 1	negative regulation of catalytic activity (GO:0043086);
ANE-Chitin-Chitosan	GLYMA08G02050	A7PZL3	-1.20	-0.84	-1.80	-2.13	Probable polygalacturonase	carbohydrate metabolic process (GO:0005975);
ANE-Chitin-Chitosan	GLYMA18G47130	A7PZL3	-2.21	0.00	-2.92	-2.80	Probable polygalacturonase	carbohydrate metabolic process (GO:0005975);
ANE-Chitin-Chitosan	GLYMA18G44820	Q8RX86	-1.24	-0.01	-1.48	-1.92	Alpha-galactosidase 2	cell wall organization (GO:0071555); leaf morphogenesis (GO:0009965); positive regulation of flower development (GO:0009911); response to fungus (GO:0009620);
ANE-Chitin-Chitosan	GLYMA12G36870	Q7XKV2	-1.57	0.00	-1.08	-1.69	Beta-glucosidase 13	carbohydrate metabolic process (GO:0005975);
ANE-Chitin-Chitosan	GLYMA02G33550	A7LXU3	-1.66	-0.05	-1.61	-1.83	Beta-glucosidase BoGH3B	mutualism (GO:0085030); xyloglucan catabolic process (GO:2000899);
ANE-Chitin-Chitosan	GLYMA03G41670	Q41808	-1.03	-0.27	-1.23	-1.04	Tubulin gamma-2 chain	cytoplasmic microtubule organization (GO:0031122); microtubule nucleation (GO:0007020);

ANE-Chitin-Chitosan	GLYMA18G03470	P05522	-3.17	-0.20	-2.02	-2.03	Endoglucanase 1	cellulose catabolic process (GO:0030245); fruit ripening (GO:0009835);
ANE-Chitin-Chitosan	GLYMA10G40690	Q8L7W8	-1.22	0.00	-1.10	-1.75	Alpha-L-fucosidase 2	No-GOBP
ANE-Chitin-Chitosan	GLYMA07G30395	O81884	-1.09	0.00	-1.02	-2.01	L-galactose dehydrogenase	galactose metabolic process (GO:0006012); L-ascorbic acid biosynthetic process (GO:0019853);
ANE-Chitin	GLYMA01G36780	Q43872	-1.06	-0.43	-1.53	0.86	Peroxidase 64	cellular oxidant detoxification (GO:0098869); hydrogen peroxide catabolic process (GO:0042744); plant-type cell wall organization (GO:0009664);
ANE-Chitin	GLYMA02G11720	Q9FH92	-1.67	-0.71	-1.01	0.00	Protein IRX15-LIKE	plant-type secondary cell wall biogenesis (GO:0009834); xylan biosynthetic process (GO:0045492);
ANE-Chitin	GLYMA02G38990	O80434	-1.14	-0.58	-1.70	-0.26	Laccase-4	lignin catabolic process (GO:0046274); plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Chitin	GLYMA01G06030	Q9C554	-2.48	-0.46	-1.71	0.46	Expansin-A1	plant-type cell wall organization (GO:0009664);
ANE-Chitin	GLYMA02G12140	Q9C554	-1.95	-0.26	-1.38	0.46	Expansin-A1	plant-type cell wall organization (GO:0009664);
ANE-Chitin	GLYMA04G40000	O22874	-3.29	-0.98	-2.60	0.17	Expansin-A8	plant-type cell wall organization (GO:0009664);
ANE-Chitin	GLYMA06G14850	O22874	-3.10	-0.99	-2.79	0.72	Expansin-A8	plant-type cell wall organization (GO:0009664);
ANE-Chitin	GLYMA18G06063	O22874	-2.20	-0.62	-2.56	2.45	Expansin-A8	plant-type cell wall organization (GO:0009664); syncytium formation (GO:0006949);
ANE-Chitin	GLYMA11G26240	O22874	-1.27	-0.90	-1.13	3.23	Expansin-A8	plant-type cell wall organization (GO:0009664); syncytium formation (GO:0006949);
ANE-Chitin	GLYMA05G36930	O48766	-1.34	-0.50	-1.08	0.69	Endoglucanase 11	cell wall organization (GO:0071555); cellulose catabolic process (GO:0030245);
ANE-Chitin	GLYMA06G02760	Q9FXI9	-1.17	0.00	-1.65	-0.45	Endoglucanase 2	cell wall organization (GO:0071555); cellulose catabolic process (GO:0030245);
ANE-Chitin	GLYMA08G02610	Q42059	-1.53	-0.53	-1.15	0.69	Endoglucanase 6	cell wall organization (GO:0071555); cellulose catabolic process (GO:0030245);
ANE-Chitin	GLYMA08G03670	Q9FJU9	-1.09	-0.10	-1.39	-0.97	Glucan endo-1,3-beta-glucosidase 13	carbohydrate metabolic process (GO:0005975); cell wall organization (GO:0071555); defense response (GO:0006952);
ANE-Chitin	GLYMA06G13400	Q9FK05	-1.71	-0.98	-1.51	0.38	Probable pectinesterase/pectinesterase inhibitor 61	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);
ANE-Chitin	GLYMA02G45620	P83948	-1.52	0.00	-1.13	-0.05	Pectinesterase 3	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);
ANE-Chitin	GLYMA07G05145	Q43111	-1.40	0.00	-1.01	0.00	Pectinesterase 3	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);

ANE-Chitin	GLYMA02G46880	Q9FM79	-1.18	0.00	-1.30	-0.56	Pectinesterase QRT1	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);
ANE-Chitin	GLYMA07G00590	Q69L19	-1.12	-0.56	-1.71	0.58	Probable xyloglucan glycosyltransferase 2	cell wall organization (GO:0071555);
ANE-Chitin	GLYMA08G23820	Q9SJA2	-1.00	-0.44	-1.75	0.69	Probable xyloglucan glycosyltransferase 8	cell wall organization (GO:0071555);
ANE-Chitin	GLYMA14G03710	A7PZL3	-1.10	-0.19	-1.31	-0.54	Probable polygalacturonase	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA09G32080	Q9LHF1	-1.58	-0.06	-1.40	-0.55	Leucine-rich repeat extensin-like protein 4	cell wall organization (GO:0071555);
ANE-Chitin	GLYMA10G39760	Q8L9A9	-2.03	-0.66	-1.04	0.38	Probable xyloglucan endotransglucosylase/hydrolase protein 8	cell wall biogenesis (GO:0042546); cell wall organization (GO:0071555); xyloglucan metabolic process (GO:0010411);
ANE-Chitin	GLYMA15G02750	Q9SCW1	-1.46	-0.70	-2.04	-0.74	Beta-galactosidase 1	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA06G16420	Q9FN08	-1.75	0.00	-1.28	0.56	Beta-galactosidase 10	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA01G37541	Q9SCV9	-1.59	-0.44	-1.91	-0.82	Beta-galactosidase 3	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA02G05790	Q9SCV9	-1.01	0.82	-1.30	-0.28	Beta-galactosidase 3	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA11G07760	Q9SCV9	-1.16	-0.34	-1.41	-0.78	Beta-galactosidase 3	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA09G30910	Q9LV34	-1.57	-0.40	-1.72	-0.83	Beta-glucosidase 43	carbohydrate metabolic process (GO:0005975);
ANE-Chitin	GLYMA08G31890	Q9S7Y7	-2.26	0.00	-1.36	0.00	Alpha-xylosidase 1	cell wall organization (GO:0071555); response to cadmium ion (GO:0046686); xylan catabolic process (GO:0045493);
ANE-Chitin	GLYMA11G19490	Q9T0L2	-1.00	-0.54	-1.90	0.31	Probable mannan synthase 15	cell wall organization (GO:0071555);
ANE-Chitin	GLYMA12G36200	F4IWA8	-1.81	-0.83	-2.78	-0.17	Protein trichome birefringence-like 41	cell wall organization or biogenesis (GO:0071554);
ANE-Chitin	GLYMA13G21440	Q9LZR3	-1.80	-0.45	-2.98	-0.36	Glucomannan 4-beta-mannosyltransferase 9	cell wall organization (GO:0071555); mannosylation (GO:0097502); response to bacterium (GO:0009617);
ANE-Chitin	GLYMA07G10790	Q8S950	-1.14	0.00	-1.01	-0.25	Kinesin-like protein NACK1	cell plate assembly (GO:0000919); microtubule-based movement (GO:0007018);
ANE-Chitin	GLYMA04G09350	P33629	-1.75	-0.64	-1.26	0.51	Tubulin alpha chain	microtubule-based process (GO:0007017);
ANE-Chitin	GLYMA20G27280	P33623	-1.32	-0.78	-1.60	0.08	Tubulin alpha-1 chain	microtubule-based process (GO:0007017);
ANE-Chitin	GLYMA16G02980	Q84JH9	-1.24	-0.57	-1.27	0.00	Protein trichome birefringence-like 25	cell wall organization or biogenesis (GO:0071554);
ANE-Chitin	GLYMA16G26650	Q94CD1	-1.62	-0.92	-1.62	1.20	Omega-hydroxypalmitate O-feruloyl transferase	cell wall pectin biosynthetic process (GO:0052325); suberin biosynthetic process (GO:0010345);
ANE-Chitin	GLYMA06G17590	Q94CD1	-1.00	0.00	-1.12	0.00	Omega-hydroxypalmitate O-feruloyl transferase	cell wall pectin biosynthetic process (GO:0052325); suberin biosynthetic process (GO:0010345);
ANE-Chitin	GLYMA04G32130	Q94KT8	-1.80	-0.73	-3.30	0.94	Protein COBRA	cellulose microfibril organization (GO:0010215); multidimensional cell growth (GO:0009825); response to salt stress (GO:0009651);

ANE-Chitin	GLYMA06G22410	Q94KT8	-1.06	-0.95	-2.67	0.44	Protein COBRA	cellulose microfibril organization (GO:0010215); multidimensional cell growth (GO:0009825); response to salt stress (GO:0009651);
ANE-Chitosan	GLYMA16G08710	Q6ZLK0	-1.00	0.00	-0.59	-1.59	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase 48 kDa subunit	cell wall modification (GO:0042545); cellulose biosynthetic process (GO:0030244); unidimensional cell growth (GO:0009826);
ANE-Chitosan	GLYMA18G39850	Q38865	-1.27	0.00	-0.91	-1.08	Expansin-A6	plant-type cell wall organization (GO:0009664);
ANE	GLYMA02G41590	Q38865	-1.72	-0.48	-0.22	0.00	Expansin-A6	plant-type cell wall organization (GO:0009664);
ANE	GLYMA12G33070	Q9M0I2	-1.28	-0.36	-0.26	2.58	Expansin-B3	cell wall organization (GO:0071555); sexual reproduction (GO:0019953);
ANE	GLYMA06G44930	Q9M0I2	-1.04	-0.50	-0.33	0.00	Expansin-B3	cell wall organization (GO:0071555); sexual reproduction (GO:0019953);
ANE	GLYMA07G35620	Q9C554	-1.12	0.00	0.30	-0.26	Expansin-A1	plant-type cell wall organization (GO:0009664);
ANE	GLYMA04G33350	O80622	-1.14	-0.35	-0.78	1.59	Expansin-A15	plant-type cell wall organization (GO:0009664); syncytium formation (GO:0006949);
ANE	GLYMA06G05190	Q8LIY0	-1.05	-0.39	-0.80	-0.45	Probable xyloglucan glycosyltransferase 1	cell wall organization (GO:0071555);
ANE	GLYMA18G18931	Q8LC45	-1.46	0.00	-0.40	0.00	Probable xyloglucan endotransglucosylase/hydrolase protein 33	plant-type cell wall modification involved in multidimensional cell growth (GO:0009831);
ANE	GLYMA10G34550	Q9LQC9	-1.12	0.00	-0.15	0.13	Probable mannan synthase 3	cell wall organization (GO:0071555);
ANE	GLYMA12G36630	Q9LXV2	-1.52	-0.96	-0.49	0.00	Transcription factor MYB46	defense response to fungus (GO:0050832); regulation of secondary cell wall biogenesis (GO:2000652); response to salicylic acid (GO:0009751);
ANE	GLYMA13G17740	Q66GM8	-1.27	-0.17	-0.66	0.04	Pectin acetylesterase 10	cell wall organization (GO:0071555);
ANE	GLYMA16G01650	O49006	-1.44	0.00	-0.62	0.61	Pectinesterase/pectinesterase inhibitor 3	cell wall modification (GO:0042545); defense response to Gram-negative bacterium (GO:0050829); response to fungus (GO:0009620); response to nematode (GO:0009624);
ANE	GLYMA19G40020	P85076	-1.37	-0.64	0.00	0.00	Pectinesterase	cell wall modification (GO:0042545); pectin catabolic process (GO:0045490);
ANE	GLYMA18G06570	Q9FGH4	-2.36	0.00	0.00	-0.22	Glucan endo-1,3-beta-glucosidase 9	carbohydrate metabolic process (GO:0005975); cell wall organization (GO:0071555); defense response (GO:0006952);
ANE	GLYMA19G40170	Q9SRW9	-1.09	0.00	0.00	-0.15	Cellulose synthase-like protein D5	cell wall biogenesis (GO:0042546); response to osmotic stress (GO:0006970); response to salt stress (GO:0009651);
ANE	GLYMA03G37550	Q9SRW9	-1.07	0.00	-0.42	-0.42	Cellulose synthase-like protein D5	cellulose biosynthetic process (GO:0030244); mannosylation (GO:0097502); response to osmotic stress (GO:0006970); response to salt stress;
ANE	GLYMA09G15620	Q84ZN6	-1.67	-0.42	0.00	0.44	Probable cellulose synthase A catalytic subunit 8 [UDP-forming]	cell wall organization (GO:0071555); cellulose biosynthetic process (GO:0030244);

ANE	GLYMA19G40180	Q8VYF4	-1.08	-0.80	-0.76	-0.56	Probable galacturonosyltransferase-like 7	cell wall organization (GO:0071555); pectin biosynthetic process (GO:0045489);
ANE	GLYMA20G04490	O80622	-2.02	0.00	-0.95	-0.36	Expansin-A15	plant-type cell wall organization (GO:0009664);
ANE	GLYMA16G24440	Q10RB4	-1.12	0.00	0.00	0.00	Beta-galactosidase 5	carbohydrate metabolic process (GO:0005975);
ANE	GLYMA10G15980	A7LXU3	-1.10	-0.05	-0.96	-0.88	Beta-glucosidase BoGH3B	mutualism (GO:0085030); xyloglucan catabolic process (GO:2000899);
ANE	GLYMA15G01840	Q940B8	-1.01	0.00	-0.85	0.00	Kinesin-13A	microtubule-based movement (GO:0007018);
ANE	GLYMA10G37960	Q40106	-1.15	0.08	-0.70	-0.30	Tubulin beta-2 chain	microtubule-based process (GO:0007017);
ANE	GLYMA20G29840	Q40106	-1.04	0.00	-0.16	0.00	Tubulin beta-2 chain	microtubule-based process (GO:0007017);

Table 31: ANE down-regulated genes involved in cell expansion.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA03G33720	Q8LEJ6	-1.04	-1.53	-2.26	0.00	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Alginate-Chitin	GLYMA03G33730	Q8LEJ6	-1.32	-1.41	-2.87	-0.69	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Alginate-Chitin	GLYMA19G36470	Q8LEJ6	-2.25	-2.11	-2.98	0.71	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE-Chitin	GLYMA02G47790	Q9SJ81	-1.06	-0.06	-1.01	-0.56	Fasciclin-like arabinogalactan protein 7	No-GOBP
ANE-Chitin	GLYMA08G44210	Q9FM65	-1.63	-0.62	-1.46	0.00	Fasciclin-like arabinogalactan protein 1	root development (GO:0048364);
ANE-Chitin	GLYMA11G02130	Q9FK16	-3.68	-0.50	-2.89	-0.60	Arabinogalactan peptide 22	No-GOBP
ANE-Chitin	GLYMA14G00830	Q9SJ81	-1.01	-0.04	-1.10	-0.29	Fasciclin-like arabinogalactan protein 7	No-GOBP
ANE-Chitin	GLYMA18G45420	O22126	-1.08	-0.76	-1.87	-0.36	Fasciclin-like arabinogalactan protein 8	No-GOBP
ANE-Chitin	GLYMA12G29670	Q8LEJ6	-1.47	-0.96	-2.38	-0.41	Fasciclin-like arabinogalactan protein 11	plant-type secondary cell wall biogenesis (GO:0009834);
ANE	GLYMA18G08530	Q9FM65	-1.49	0.00	-0.93	-0.73	Fasciclin-like arabinogalactan protein 1	root development (GO:0048364);

Table 32: ANE down-regulated genes involved in lignin catabolic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA04G40580	P28002	-1.12	-1.42	-1.93	-1.29	Caffeic acid 3-O-methyltransferase	lignin biosynthetic process (GO:0009809);
ANE-Alginate-Chitin	GLYMA07G16080	Q9FJD5	-3.10	-3.86	-2.97	0.00	Laccase-17	lignin catabolic process (GO:0046274);
ANE-Alginate-Chitin	GLYMA11G35700	Q9SIY8	-1.16	-2.98	-1.24	1.03	Laccase-5	lignin catabolic process (GO:0046274);
ANE-Alginate-Chitin	GLYMA18G02690	Q9SIY8	-1.85	-2.40	-1.17	0.66	Laccase-5	lignin catabolic process (GO:0046274);
ANE-Alginate-Chitin	GLYMA18G40070	Q9FJD5	-2.35	-2.65	-2.81	0.00	Laccase-17	lignin catabolic process (GO:0046274);
ANE-Chitin-Chitosan	GLYMA16G24341	Q42600	-1.07	-0.50	-1.11	-2.61	Cytochrome P450 84A1	lignin biosynthetic process (GO:0009809); phenylpropanoid biosynthetic process (GO:0009699);
ANE-Chitosan	GLYMA06G26610	Q07488	-1.76	-0.77	0.00	-1.24	Blue copper protein	defense response to fungus (GO:0050832); regulation of lignin biosynthetic process (GO:1901141);
ANE	GLYMA06G14210	P28002	-1.52	0.00	0.00	-0.64	Caffeic acid 3-O-methyltransferase	lignin biosynthetic process (GO:0009809);

Table 33: ANE down-regulated genes involved in pectin catabolic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA05G02890	Q93Z25	-2.08	-1.35	-1.98	1.94	Probable pectate lyase 22	pectin catabolic process (GO:0045490);
ANE-Alginate-Chitin	GLYMA17G13550	Q93Z25	-2.25	-1.80	-2.29	2.45	Probable pectate lyase 22	pectin catabolic process (GO:0045490);
ANE	GLYMA10G24630	Q944R1	-1.65	-0.11	-0.66	0.31	Probable pectate lyase 15	pectin catabolic process (GO:0045490);
ANE	GLYMA20G19200	Q940Q1	-1.98	-0.81	-0.87	1.78	Probable pectate lyase 1	pectin catabolic process (GO:0045490);

Table 34: ANE down-regulated genes involved in wax biosynthesis.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin	GLYMA10G36690	Q96520	-1.91	-0.02	-1.78	-0.63	Peroxidase 12	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE-Chitosan	GLYMA12G15460	P16147	-1.86	-0.98	0.35	-1.11	Peroxidase	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE	GLYMA09G41440	P22195	-2.03	0.00	3.16	-0.08	Cationic peroxidase 1	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE	GLYMA19G09290	Q9FVS9	-1.12	-0.05	-0.81	2.13	Alkane hydroxylase MAH1	wax biosynthetic process (GO:0010025);
ANE	GLYMA13G07550	Q93ZR6	-1.00	0.00	0.00	0.03	O-acyltransferase WSD1	triglyceride biosynthetic process (GO:0019432); wax biosynthetic process (GO:0010025);
ANE	GLYMA18G44310	P22195	-1.73	-0.41	0.75	-0.26	Cationic peroxidase 1	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE	GLYMA08G19180	P22196	-1.01	0.00	0.46	-0.54	Cationic peroxidase 2	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE	GLYMA16G27880	Q96520	-1.74	-0.72	-0.45	0.00	Peroxidase 12	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979); biosynthesis and degradation of lignin, suberization, auxin catabolism

Table 35: ANE down-regulated genes involved in other synthetic processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA20G33450	Q8W3Z2	-1.67	-2.08	-2.39	2.09	Lupeol synthase	pentacyclic triterpenoid biosynthetic process (GO:0019745);
ANE-Alginate-Chitin	GLYMA08G21300	Q8H1S0	-1.76	-1.72	-1.02	-0.02	Inositol oxygenase 4	inositol catabolic process (GO:0019310); L-ascorbic acid biosynthetic process (GO:0019853);
ANE-Chitin-Chitosan	GLYMA06G47430	Q8GW43	-1.06	-0.04	-1.45	-1.46	8-amino-7-oxononanoate synthase	biotin biosynthetic process (GO:0009102);
ANE-Chitin	GLYMA06G09400	O80594	-2.06	-0.54	-3.45	-0.89	Ergosterol biosynthetic protein 28	sterol biosynthetic process (GO:0016126);
ANE-Chitin	GLYMA03G42160	O81032	-2.30	0.22	-1.82	0.00	Probable L-gulonolactone oxidase 6	L-ascorbic acid biosynthetic process (GO:0019853);

ANE-Chitin	GLYMA20G05530	P29057	-1.54	-0.65	-1.82	1.23	3-hydroxy-3-methylglutaryl-coenzyme A reductase 1	coenzyme A metabolic process (GO:0015936); isoprenoid biosynthetic process (GO:0008299);
ANE-Chitin	GLYMA13G40771	Q6L506	-1.12	0.34	-1.16	-0.57	Probable monofunctional riboflavin biosynthesis protein RIBA 3, chloroplastic	riboflavin biosynthetic process (GO:0009231);
ANE-Chitin	GLYMA05G38260	Q9LUF2	-1.24	-0.88	-1.08	2.37	11-beta-hydroxysteroid dehydrogenase-like 4B	steroid biosynthetic process (GO:0006694);
ANE-Chitosan	GLYMA18G18880	O82147	-1.35	0.06	-0.98	-1.48	Spermidine synthase	spermidine biosynthetic process (GO:0008295);
ANE	GLYMA04G09260	O80594	-1.01	0.00	-0.84	-0.68	Ergosterol biosynthetic protein 28	sterol biosynthetic process (GO:0016126);
ANE	GLYMA12G36270	Q6EQG2	-1.08	0.00	-0.60	-0.52	Probable NADH kinase	NAD metabolic process (GO:0019674); NADP biosynthetic process (GO:0006741);
ANE	GLYMA14G05360	Q08506	-1.46	-0.73	-0.38	0.00	1-aminocyclopropane-1-carboxylate oxidase 1	ethylene biosynthetic process (GO:0009693);
ANE	GLYMA06G16990	Q9LFU9	-1.08	0.00	-0.19	0.00	Glucosamine 6-phosphate N-acetyltransferase	N-acetylglucosamine metabolic process (GO:0006044); UDP-N-acetylglucosamine biosynthetic process (GO:0006048);
ANE	GLYMA04G14361	P43309	-2.03	0.00	-0.18	-0.16	Polyphenol oxidase, chloroplastic	pigment biosynthetic process (GO:0046148);
ANE	GLYMA18G50290	Q84KK5	-1.89	0.00	-0.11	-0.12	Isoflavone 7-O-methyltransferase	isoflavonoid biosynthetic process (GO:0009717);
ANE	GLYMA06G42170	P43309	-1.33	-0.49	0.00	0.00	Polyphenol oxidase, chloroplastic	pigment biosynthetic process (GO:0046148);

Table 36: ANE down-regulated genes involved in plant defense and other responses.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA12G31320	Q9SJ52	-3.08	-1.81	-2.25	0.61	Protein YLS9	defense response to virus (GO:0051607); leaf senescence (GO:0010150);
ANE-Alginate	GLYMA19G38600	Q9SJ52	-1.08	-1.51	0.00	1.20	Protein YLS9	defense response to virus (GO:0051607); leaf senescence (GO:0010150);
ANE-Chitin-Chitosan	GLYMA06G11820	Q93ZH0	-1.20	-0.69	-1.44	-1.28	LysM domain-containing GPI-anchored protein 1	defense response (GO:0006952); immune response (GO:0006955);
ANE-Chitin-Chitosan	GLYMA05G21780	Q9SNV9	-1.99	1.22	-1.71	-1.63	Metal transporter Nramp3	defense response to bacterium (GO:0042742); response to bacterium (GO:0009617);
ANE-Chitin-Chitosan	GLYMA17G18010	Q9SNV9	-2.81	1.27	-2.08	-2.22	Metal transporter Nramp3	defense response to bacterium (GO:0042742); response to bacterium (GO:0009617);

ANE-Chitin-Chitosan	GLYMA19G43460	P02877	-2.49	-0.08	-2.26	-1.31	Pro-hevein	defense response to bacterium (GO:0042742); defense response to fungus (GO:0050832);
ANE-Chitin-Chitosan	GLYMA14G09620	Q948Z4	-1.65	-0.88	-2.78	-1.10	Snakin-1	defense response (GO:0006952);
ANE-Chitin-Chitosan-	GLYMA04G09740	O04496	-2.62	-0.52	-2.62	-2.41	Aspartyl protease AED3	systemic acquired resistance (GO:0009627);
ANE-Chitin-Chitosan-	GLYMA04G17600	O04496	-2.81	0.05	-3.00	-3.00	Aspartyl protease AED3	systemic acquired resistance (GO:0009627);
ANE-Chitin-Chitosan-	GLYMA06G09830	O04496	-2.63	-0.65	-3.96	-2.48	Aspartyl protease AED3	systemic acquired resistance (GO:0009627);
ANE-Chitin	GLYMA01G04070	Q93VR4	-1.66	-0.55	-3.23	2.25	MLP-like protein 423	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE-Chitin	GLYMA02G03680	Q93VR4	-3.04	-0.21	-2.18	1.89	MLP-like protein 423	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE-Chitin	GLYMA13G01250	Q40374	-2.55	0.00	-1.77	-0.63	Pathogenesis-related protein PR-1	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE-Chitin	GLYMA12G35420	Q9LUH3	-1.12	0.00	-1.24	-0.45	Protein DETOXIFICATION 18	defense response to oomycetes (GO:0002229); defense response to other organism (GO:0098542);
ANE-Chitin	GLYMA17G35530	Q948Z4	-3.00	-0.62	-3.29	-0.69	Snakin-1	defense response (GO:0006952);
ANE-Chitin	GLYMA05G01845	Q93X17	-1.46	-0.37	-2.23	-0.74	Snakin-2	defense response (GO:0006952);
ANE-Chitin	GLYMA17G07740	Q9M0B6	-1.11	-0.95	-1.30	0.10	UDP-glucuronate 4-epimerase 1	defense response to fungus (GO:0050832); defense response to Gram-negative bacterium (GO:0050829); galacturonate biosynthetic process (GO:0033481);
ANE-Chitin	GLYMA11G36600	Q9LIS3	-1.05	-0.94	-1.30	0.40	UDP-glucuronate 4-epimerase 6	defense response to fungus (GO:0050832); defense response to Gram-negative bacterium (GO:0050829); galacturonate biosynthetic process (GO:0033481);
ANE-Chitosan	GLYMA14G05300	Q9ZU91	-1.11	-0.23	-0.94	-1.09	Glucan endo-1,3-beta-glucosidase 3	carbohydrate metabolic process (GO:0005975); defense response (GO:0006952);
ANE	GLYMA14G05355	Q0WPW4	-1.31	-0.30	-0.16	-0.26	1-aminocyclopropane-1-carboxylate oxidase 5	defense response (GO:0006952); ethylene biosynthetic process (GO:0009693);
ANE	GLYMA17G03360	Q43560	-1.35	-0.28	0.87	0.00	Class-10 pathogenesis-related protein 1	defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE	GLYMA16G18480	A3FPF2	-1.17	0.59	-0.34	0.45	Defensin-like protein	defense response to fungus (GO:0050832); killing of cells of other organism (GO:0031640);
ANE	GLYMA03G28850	Q03773	-3.83	0.00	-0.30	0.68	Glucan endo-1,3-beta-glucosidase	carbohydrate metabolic process (GO:0005975); defense response (GO:0006952);

ANE	GLYMA08G46110	Q06915	-1.12	-0.49	-0.73	-0.48	Probable glucan endo-1,3-beta-glucosidase A6	carbohydrate metabolic process (GO:0005975); defense response (GO:0006952);
ANE	GLYMA17G16600	Q93ZB1	-1.13	0.00	-0.89	0.09	Protein LOL1	apoptotic process (GO:0006915); defense response, incompatible interaction (GO:0009814);
ANE	GLYMA11G08240	P83332	-1.38	-0.18	-0.67	-0.97	Thaumatin-like protein 1	defense response (GO:0006952);
ANE	GLYMA18G00505	Q9LIS3	-1.78	-0.75	-0.47	0.36	UDP-glucuronate 4-epimerase 6	defense response to fungus (GO:0050832); defense response to Gram-negative bacterium (GO:0050829); galacturonate biosynthetic process (GO:0033481);
ANE-Alginate-Chitin-Chitosan	GLYMA08G47070	Q8GXU5	-2.12	-2.40	-2.08	-1.70	Protein SULFUR DEFICIENCY-INDUCED 1	cellular response to sulfur starvation (GO:0010438); regulation of sulfur utilization (GO:0006792);
ANE-Alginate-Chitin-Chitosan	GLYMA18G38060	Q8GXU5	-2.39	-2.83	-2.93	-2.00	Protein SULFUR DEFICIENCY-INDUCED 1	cellular response to sulfur starvation (GO:0010438); regulation of sulfur utilization (GO:0006792);
ANE-Alginate	GLYMA18G44320	P22195	-1.71	-1.06	1.15	-0.69	Cationic peroxidase 1	hydrogen peroxide catabolic process (GO:0042744); response to oxidative stress (GO:0006979);
ANE-Chitin-Chitosan	GLYMA14G09650	Q9STX2	-1.22	-0.53	-2.08	-1.49	3-oxo-Delta (4,5)-steroid 5-beta-reductase	response to wounding (GO:0009611); steroid metabolic process (GO:0008202);
ANE-Chitin-Chitosan	GLYMA19G05120	Q9FWA3	-1.38	-0.79	-1.42	-1.33	6-phosphogluconate dehydrogenase, decarboxylating 3	D-gluconate metabolic process (GO:0019521); response to salt stress (GO:0009651);
ANE-Chitin-Chitosan	GLYMA17G11170	Q91XA9	-1.16	0.00	-1.24	-1.44	Acidic mammalian chitinase	apoptotic process (GO:0006915); chitin catabolic process (GO:0006032); immune system process (GO:0002376)
ANE-Chitin-Chitosan	GLYMA09G03600	Q8VYR7	-1.16	0.00	-1.76	-1.64	Boron transporter 1	response to boron-containing substance (GO:0010036);
ANE-Chitin-Chitosan	GLYMA17G14280	Q93YR3	-1.46	0.00	-1.20	-1.34	FAM10 family protein	response to cadmium ion (GO:0046686);
ANE-Chitin-Chitosan	GLYMA09G08210	A6X935	-1.10	-0.47	-1.09	-1.35	Inter alpha-trypsin inhibitor, heavy chain 4	acute-phase response (GO:0006953); hyaluronan metabolic process (GO:0030212); response to cytokine (GO:0034097);
ANE-Chitin-Chitosan	GLYMA15G15990	Q39644	-1.63	-0.59	-1.80	-2.26	Late embryogenesis abundant protein Lea5	response to stress (GO:0006950);
ANE-Chitin-Chitosan	GLYMA09G01160	Q94CF0	-2.22	0.00	-2.34	-1.04	EH domain-containing protein 1	endocytic recycling (GO:0032456); endocytosis (GO:0006897); hyperosmotic salinity response;
ANE-Chitin-Chitosan	GLYMA15G09970	Q5PP26	-1.12	0.00	-1.24	-1.18	Piriformospora indica-insensitive protein 2	response to symbiotic fungus (GO:0009610); symbiosis, encompassing mutualism through parasitism (GO:0044403);
ANE-Chitin	GLYMA09G23640	Q9FK81	-1.06	-0.42	-1.01	-0.27	Stress-response A/B barrel domain-containing protein	No-GOBP

ANE	GLYMA03G41880	Q9LNJ3	-1.42	-0.07	0.00	0.15	Aspartyl protease family protein 2	response to karrikin (GO:0080167);
ANE	GLYMA07G06100	Q9LNJ3	-1.56	-0.26	-0.38	-0.33	Aspartyl protease family protein 2	response to karrikin (GO:0080167);
ANE	GLYMA19G44540	Q9LNJ3	-1.29	-0.13	-0.87	0.00	Aspartyl protease family protein 2	response to karrikin (GO:0080167);
ANE	GLYMA05G03770	Q93YR3	-1.01	0.00	-0.69	-0.97	FAM10 family protein	response to cadmium ion (GO:0046686);
ANE	GLYMA20G38690	Q94JV5	-1.02	-0.34	-0.25	-0.69	Nitrilase-like protein 2	nitrogen compound metabolic process (GO:0006807); response to cadmium ion (GO:0046686);
ANE	GLYMA05G22751	Q058P4	-1.21	0.00	-0.33	-0.12	U-box domain-containing protein 30	cellular response to salt stress (GO:0071472); negative regulation of response to salt stress (GO:1901001);

Table 37: ANE down-regulated genes involved in oxidation-reduction processes

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA05G30380	P00303	-2.56	-3.25	-1.58	1.72	Basic blue protein	oxidation-reduction process (GO:0055114);
ANE-Alginate-Chitin	GLYMA06G10500	Q41001	-1.64	-2.15	-1.84	0.25	Blue copper protein	oxidation-reduction process (GO:0055114);
ANE-Alginate	GLYMA06G12680	P00303	-1.46	-1.41	-0.75	1.30	Basic blue protein	oxidation-reduction process (GO:0055114);
ANE-Alginate	GLYMA14G35530	Q41001	-1.05	-2.51	-0.63	0.00	Blue copper protein	oxidation-reduction process (GO:0055114);
ANE-Chitin-Chitosan	GLYMA17G02670	Q8L8Z8	-2.92	1.52	-2.14	-3.40	Monothiol glutaredoxin-S2	cell redox homeostasis (GO:0045454);
ANE-Chitin-Chitosan	GLYMA07G38030	Q8L8Z8	-2.95	0.00	-1.69	-4.01	Monothiol glutaredoxin-S2	cell redox homeostasis (GO:0045454);
ANE-Chitin-Chitosan	GLYMA10G02590	Q9LYC8	-1.25	0.00	-1.37	-2.35	Monothiol glutaredoxin-S6	cell redox homeostasis (GO:0045454);
ANE-Chitosan	GLYMA13G28750	O23419	-1.13	0.87	0.00	-2.27	Monothiol glutaredoxin-S4	cell redox homeostasis (GO:0045454);
ANE	GLYMA13G22650	Q41001	-1.28	-0.51	0.21	0.40	Blue copper protein	oxidation-reduction process (GO:0055114);
ANE	GLYMA02G47700	Q9SJ74	-1.18	-0.78	0.00	0.12	Cytochrome b561 and DOMON domain-containing protein	oxidation-reduction process (GO:0055114);

ANE	GLYMA04G42690	P29828	-1.04	0.18	-0.57	-0.69	Protein disulfide-isomerase	cell redox homeostasis (GO:0045454);
ANE	GLYMA16G27900	Q96520	-1.09	0.00	0.00	-0.52	Peroxidase 12	biosynthesis and degradation of lignin, suberization, auxin catabolism
ANE	GLYMA09G16810	Q9LEH3	-1.33	-0.41	-0.57	0.00	Peroxidase 15	biosynthesis and degradation of lignin, suberization, auxin catabolism

Table 38: ANE down-regulated genes involved in photosynthesis

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA11G19840	P11964	-1.23	-1.15	-1.27	-1.76	Superoxide dismutase [Cu-Zn], chloroplastic	cellular response to high light intensity (GO:0071486);
ANE-Alginate-Chitin-Chitosan	GLYMA12G08650	P11964	-1.58	-1.46	-1.63	-1.81	Superoxide dismutase [Cu-Zn], chloroplastic	cellular response to high light intensity (GO:0071486);
ANE-Alginate	GLYMA11G18541	Q9S9P8	-2.36	-1.22	-0.69	-0.33	Ferredoxin--NADP reductase, root isozyme 2, chloroplastic	photosynthesis (GO:0015979);
ANE-Chitin-Chitosan	GLYMA03G00900	O22682	-1.41	0.00	-1.43	-1.88	Probable S-sulfocysteine synthase, chloroplastic	cysteine biosynthetic process from serine (GO:0006535); photosynthesis (GO:0015979); photosynthetic acclimation (GO:0009643);
ANE-Chitin-Chitosan	GLYMA02G14870	Q8VY09	-1.03	-0.62	-1.78	-1.38	Protein DJ-1 homolog C	cellular response to hydrogen peroxide (GO:0070301); chloroplast organization (GO:0009658); regulation of transcription from RNA polymerase II promoter (GO:0006357);
ANE-Chitin-Chitosan	GLYMA19G29741	O22682	-1.18	0.00	-1.48	-2.61	Probable S-sulfocysteine synthase, chloroplastic	cysteine biosynthetic process from serine (GO:0006535); photosynthesis (GO:0015979); photosynthetic acclimation (GO:0009643);
ANE-Chitin-Chitosan	GLYMA01G34300	P39657	-2.10	1.82	-4.16	-2.18	RuBisCO-associated protein	carbohydrate metabolic process (GO:0005975);
ANE-Chitin-Chitosan	GLYMA08G01050	Q9SW18	-1.61	-0.15	-2.56	-1.89	Magnesium protoporphyrin IX methyltransferase, chloroplastic	chlorophyll biosynthetic process (GO:0015995);
ANE-Chitin-Chitosan	GLYMA08G01060	Q9SW18	-1.67	-0.02	-2.44	-2.07	Magnesium protoporphyrin IX methyltransferase, chloroplastic	chlorophyll biosynthetic process (GO:0015995);
ANE-Chitin-Chitosan	GLYMA11G10040	Q43082	-1.27	0.00	-1.25	-1.59	Porphobilinogen deaminase, chloroplastic	chlorophyll biosynthetic process (GO:0015995);
ANE-Chitin-Chitosan	GLYMA03G33120	Q9LS48	-1.04	0.08	-1.16	-1.57	protein SLOW GREEN 1, chloroplastic	chloroplast organization (GO:0009658);

ANE-Chitin-Chitosan	GLYMA13G19610	Q9LS48	-2.14	0.00	-2.17	-2.18	protein SLOW GREEN 1, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA19G35820	Q9LS48	-1.45	0.28	-1.10	-1.63	protein SLOW GREEN 1, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA13G34500	Q9AXB0	-1.08	0.18	-1.29	-1.53	Uroporphyrinogen decarboxylase 1, chloroplastic	chlorophyll biosynthetic process (GO:0015995); heme biosynthetic process (GO:0006783);
ANE-Chitin-Chitosan	GLYMA01G07780	F4I0K2	-2.05	0.00	-2.67	-1.76	Fructokinase-like 2, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA01G26990	Q9M394	-1.90	0.00	-2.08	-1.77	Fructokinase-like 1, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA02G13260	F4I0K2	-1.27	0.00	-1.57	-2.07	Fructokinase-like 2, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA03G14960	Q9M394	-2.02	0.40	-1.99	-2.17	Fructokinase-like 1, chloroplastic	chloroplast organization (GO:0009658);
ANE-Chitin-Chitosan	GLYMA13G41370	Q9STE8	-1.69	0.00	-1.57	-1.31	Protein TOC75-3, chloroplastic	chloroplast organization (GO:0009658); protein targeting to chloroplast (GO:0045036); response to cytokinin (GO:0009735);
ANE-Chitin-Chitosan	GLYMA20G12510	Q9FMX0	-1.56	-0.15	-1.91	-1.94	Superoxide dismutase [Fe] 3, chloroplastic	Plays important role in chloroplast development
ANE-Chitin-Chitosan	GLYMA15G04030	Q9STE8	-1.42	0.00	-1.51	-1.24	Protein TOC75-3, chloroplastic	chloroplast organization (GO:0009658); protein targeting to chloroplast (GO:0045036); response to cytokinin (GO:0009735);
ANE-Chitin-Chitosan	GLYMA13G43920	Q39002	-1.40	0.03	-1.34	-2.84	ADP, ATP carrier protein 1, chloroplastic	ADP transport (GO:0015866); ATP transport (GO:0015867);
ANE-Chitin-Chitosan	GLYMA12G24991	P14853	-1.51	0.00	-2.15	-2.41	Cytochrome c oxidase subunit 3	aerobic electron transport chain (GO:0019646);
ANE-Chitin	GLYMA04G03920	O22227	-1.27	-1.00	-1.95	0.52	Protein MIZU-KUSSEI 1	chloroplast organization (GO:0009658); hydrotropism (GO:0010274);
ANE-Chitosan	GLYMA15G01420	Q39002	-1.07	0.08	-0.96	-2.19	ADP, ATP carrier protein 1, chloroplastic	ADP transport (GO:0015866); ATP transport (GO:0015867);
ANE	GLYMA03G02271	Q9LE33	-1.40	0.00	-0.04	-0.35	Glyoxylate/hydroxypyruvate reductase HPR3	oxidative photosynthetic carbon pathway (GO:0009854); photorespiration (GO:0009853);
ANE	GLYMA09G26731	Q09G46	-1.00	0.00	0.00	0.00	Photosystem I P700 chlorophyll a apoprotein A1	photosynthesis (GO:0015979); protein-chromophore linkage (GO:0018298);
ANE	GLYMA11G16391	Q2PMU3	-1.01	0.00	0.83	-0.40	Photosystem I P700 chlorophyll a apoprotein A1	photosynthesis (GO:0015979); protein-chromophore linkage (GO:0018298);
ANE	GLYMA19G41510	O23403	-1.23	0.00	0.00	0.00	PsbP domain-containing protein 1, chloroplastic	photosystem I assembly (GO:0048564);

ANE	GLYMA17G37450	Q9FIG9	-1.35	0.00	-0.90	-0.54	Protein ACCUMULATION AND REPLICATION OF CHLOROPLASTS 6	chloroplast fission (GO:0010020); chloroplast organization (GO:0009658);

Table 39: ANE down-regulated genes involved in the stabilization of mitochondrial proteins.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Chitin-Chitosan	GLYMA09G30410	O49460	-1.19	0.00	-1.07	-1.50	Prohibitin-1, mitochondrial	No-GOBP
ANE-Chitin-Chitosan	GLYMA04G16350	Q9LK25	-1.40	0.00	-1.32	-1.01	Prohibitin-4, mitochondrial	No-GOBP
ANE-Chitin	GLYMA11G11670	O49460	-1.16	0.00	-1.15	-0.68	Prohibitin-1, mitochondrial	No-GOBP
ANE-Chitin	GLYMA12G33520	O49460	-1.12	0.02	-1.12	-0.97	Prohibitin-1, mitochondrial	No-GOBP
ANE-Chitin	GLYMA19G35670	Q9FZ84	-1.22	0.00	-1.16	-0.54	Glycine-rich RNA-binding protein 6, mitochondrial	RNA transcription or processing during stress
ANE-Chitin-Chitosan	GLYMA03G32960	Q9FZ84	-1.00	0.00	-1.10	-1.38	Glycine-rich RNA-binding protein 6, mitochondrial	response to salt stress (GO:0009651);
ANE-Chitin-Chitosan	GLYMA06G47520	O04331	-1.43	0.00	-1.28	-1.14	Prohibitin-3, mitochondrial	cell division (GO:0051301); cell growth (GO:0016049); lateral root development (GO:0048527); mitochondrion organization;

Table 40: ANE down-regulated genes involved in transport.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA19G04450	Q41951	-3.03	-1.12	-4.56	-4.16	Aquaporin TIP2-1	glycerol transport (GO:0015793); methylammonium transmembrane transport (GO:0072489); water transport (GO:0006833);
ANE-Alginate	GLYMA06G13110	Q6NQN5	-1.20	-1.32	0.00	0.00	Bidirectional sugar transporter SWEET3	carbohydrate transmembrane transport (GO:0034219);
ANE-Chitin-Chitosan	GLYMA13G01570	Q9M0B8	-3.98	-0.73	-3.74	-3.94	WAT1-related protein	transmembrane transport (GO:0055085);

ANE-Chitin-Chitosan	GLYMA04G37700	Q0WPN8	-1.59	-0.27	-1.98	-1.44	Cysteine-rich repeat secretory protein 60	transport (GO:0006810); viral process (GO:0016032);
ANE-Chitin-Chitosan	GLYMA07G03220	P21616	-2.47	0.00	-1.48	-1.47	Pyrophosphate-energized vacuolar membrane proton pump	proton transport (GO:0015992);
ANE-Chitin-Chitosan	GLYMA08G10710	Q9LK64	-1.64	0.31	-2.66	-1.96	ABC transporter C family member 3	No-GOBP
ANE-Chitin	GLYMA05G37730	Q7XSQ9	-1.41	-0.26	-1.62	0.00	Probable aquaporin PIP1-2	glycerol transport (GO:0015793); ion transmembrane transport (GO:0034220); water transport (GO:0006833);
ANE-Chitin	GLYMA08G01860	Q7XSQ9	-1.11	-0.09	-1.61	-0.96	Probable aquaporin PIP1-2	glycerol transport (GO:0015793); ion transmembrane transport (GO:0034220); water transport (GO:0006833);
ANE-Chitosan	GLYMA08G22880	P21616	-1.14	-0.05	-0.91	-1.63	Pyrophosphate-energized vacuolar membrane proton pump	proton transport (GO:0015992);
ANE-Chitosan	GLYMA05G31520	Q9C9A3	-1.10	0.00	-0.66	-1.29	Heavy metal-associated isoprenylated plant protein 20	cellular transition metal ion homeostasis (GO:0046916); metal ion transport (GO:0030001);
ANE	GLYMA13G34520	Q39688	-1.58	-0.30	-0.17	0.00	Epidermis-specific secreted glycoprotein EP1	water transport (GO:0006833);
ANE	GLYMA16G27370	Q9LE20	-1.47	0.00	0.00	-0.06	Protein DETOXIFICATION 54	drug transmembrane transport (GO:0006855);
ANE	GLYMA14G05220	Q9SHZ3	-1.43	-0.06	0.00	0.00	Nucleobase-ascorbate transporter 1	transmembrane transport (GO:0055085);
ANE	GLYMA13G39965	Q9SL29	-1.33	-0.44	0.00	0.00	Putative cyclic nucleotide-gated ion channel 15	potassium ion transmembrane transport (GO:0071805); regulation of membrane potential (GO:0042391);
ANE	GLYMA06G05460	O04089	-1.21	0.72	-0.91	-0.48	Zinc transporter 4, chloroplastic	No-GOBP

Table 41: ANE down-regulated genes encoding various receptors.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA15G13840	C0LGQ9	-2.26	-1.11	-2.38	-1.05	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE-Chitin-Chitosan	GLYMA09G02881	C0LGQ9	-1.65	-0.52	-1.32	-1.26	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE-Chitin-Chitosan	GLYMA02G10770	Q9LRT1	-1.25	-0.55	-1.56	-1.49	Probably inactive leucine-rich repeat receptor-like protein kinase	No-GOBP

ANE-Chitin-Chitosan	GLYMA15G29880	Q8LFN2	-1.79	-0.08	-2.15	-2.28	Probable inactive leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE-Chitin-Chitosan	GLYMA12G32880	Q6R2K3	-1.08	-0.76	-1.29	-1.13	Protein STRUBBELIG-RECEPTOR FAMILY 3	circadian rhythm (GO:0007623);
ANE-Chitin	GLYMA02G40340	Q9C9Y8	-1.45	-0.30	-1.53	-0.79	Probable inactive receptor kinase	No-GOBP
ANE-Chitin	GLYMA02G45545	Q9SJG2	-1.13	0.00	-1.39	-0.65	Probable receptor-like protein kinase	No-GOBP
ANE-Chitin	GLYMA03G29670	Q9FL51	-1.43	-0.23	-1.55	-0.12	Probably inactive leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE-Chitin	GLYMA07G11680	Q9LP77	-1.15	-0.77	-1.09	0.33	Probable inactive receptor kinase	No-GOBP
ANE-Chitin	GLYMA09G18550	Q9M9C5	-1.52	-0.33	-1.55	0.00	Probable leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE-Chitin	GLYMA13G42601	Q8RWW0	-1.42	-0.92	-1.54	0.69	Receptor-like serine/threonine-protein kinase ALE2	cuticle development (GO:0042335); organ formation (GO:0048645);
ANE-Chitin	GLYMA14G06051	Q9SCT4	-1.33	-0.13	-1.45	-0.01	Probably inactive leucine-rich repeat receptor-like protein kinase IMK2	No-GOBP
ANE-Chitin	GLYMA15G11820	Q6R2J8	-1.10	-0.53	-1.02	0.00	Protein STRUBBELIG-RECEPTOR FAMILY 8	No-GOBP
ANE-Chitosan	GLYMA17G00680	Q9LT96	-1.07	0.00	-0.30	-1.23	Probable leucine-rich repeat receptor-like protein kinase	No-GOBP
ANE	GLYMA03G29490	P93756	-1.90	0.00	-0.19	-0.11	G-type lectin S-receptor-like serine/threonine-protein kinase SD3-1	recognition of pollen (GO:0048544);
ANE	GLYMA08G06020	Q9LP77	-1.04	-0.15	-0.53	-0.41	Probable inactive receptor kinase	No-GOBP
ANE	GLYMA08G21151	Q9FZB1	-1.46	0.15	0.00	0.00	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE	GLYMA08G21190	Q9FZB1	-1.26	0.00	0.16	0.00	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE	GLYMA09G40870	C0LGS2	-1.34	0.70	0.00	0.00	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE	GLYMA10G37250	C0LGQ5	-1.16	0.40	0.30	0.24	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE	GLYMA10G37260	C0LGQ5	-1.18	0.00	-0.99	-0.07	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE	GLYMA10G37290	C0LGQ5	-1.01	0.33	-0.47	0.00	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE	GLYMA10G37300	C0LGQ5	-1.49	0.14	-0.54	-0.11	LRR receptor-like serine/threonine-protein kinase GSO1	plant epidermis development (GO:0090558);
ANE	GLYMA13G35690	Q9LK35	-1.03	0.00	-0.49	-0.7	Receptor-like protein kinase THESEUS 1	protein autophosphorylation (GO:0046777);
ANE	GLYMA16G33991	Q9SSN3	-1.43	0.00	0.00	0.00	Toll/interleukin-1 receptor-like protein	defense response (GO:0006952);
ANE	GLYMA18G42700	Q8VZG8	-1.33	0.00	-0.06	0.00	Probable LRR receptor-like serine/threonine-protein kinase	No-GOBP
ANE	GLYMA18G43730	Q9C9N5	-1.02	-0.27	-0.97	-0.60	Probable inactive leucine-rich repeat receptor-like protein kinase	No-GOBP

Table 42: ANE down-regulated genes associated with transcription factors.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA07G37410	P42736	-2.11	-2.29	-3.12	-2.17	Ethylene-responsive transcription factor RAP2-3	cell death (GO:0008219); transcription, DNA-templated (GO:0006351);
ANE-Alginate-Chitin-Chitosan	GLYMA10G03530	Q9LUH8	-2.21	-1.48	-2.90	-3.30	Heat stress transcription factor A-6b	transcription, DNA-templated (GO:0006351);
ANE-Alginate-Chitin	GLYMA04G07140	Q9LFN7	-1.17	-1.05	-1.85	0.52	Ethylene-responsive transcription factor SHINE 2	ethylene-activated signaling pathway (GO:0009873); transcription, DNA-templated (GO:0006351);
ANE-Alginate-Chitin	GLYMA09G34461	Q9SAK5	-1.12	-1.98	-1.63	-0.66	Myb family transcription factor APL	phloem development (GO:0010088); positive regulation of transcription, DNA-templated (GO:0045893); xylem development (GO:0010089);
ANE-Alginate-Chitin	GLYMA17G15310	Q8LBQ7	-1.43	-1.03	-1.44	0.94	Ethylene-responsive transcription factor ERF034	ethylene-activated signaling pathway (GO:0009873); transcription, DNA-templated (GO:0006351);
ANE-Alginate-Chitin	GLYMA06G43760	Q9M2K4	-2.28	-1.70	-3.48	1.53	Basic leucine zipper 61	positive regulation of transcription, DNA-templated (GO:0045893); transcription, DNA-templated (GO:0006351);
ANE-Alginate	GLYMA07G05740	O23090	-1.21	-1.77	-0.49	-0.17	Transcription factor bHLH14	regulation of transcription, DNA-templated (GO:0006355); transcription, DNA-templated (GO:0006351);
ANE-Alginate	GLYMA11G30940	Q6S4P4	-1.09	-1.05	0.00	0.00	Transcription factor RF2b	transcription, DNA-templated (GO:0006351);
ANE-Alginate	GLYMA15G19460	Q6AWY8	-1.76	-1.72	-0.75	0.00	Growth-regulating factor 1	developmental process (GO:0032502); regulation of transcription, DNA-templated (GO:0006355); transcription, DNA-templated (GO:0006351);
ANE-Chitin-Chitosan	GLYMA07G05500	Q700E4	-3.08	-0.91	-2.50	-1.91	Transcription factor bHLH67	regulation of transcription, DNA-templated (GO:0006355); transcription, DNA-templated (GO:0006351);
ANE-Chitin-Chitosan	GLYMA11G02140	Q9SUQ2	-1.94	0.00	-1.30	-1.48	Ethylene-responsive transcription factor CRF2	cotyledon development (GO:0048825); root development (GO:0048364); transcription factor imports into nucleus (GO:0042991);
ANE-Chitin-Chitosan	GLYMA04G40661	A0A0B2 QIG5	-1.17	0.00	-1.12	-1.46	Uncharacterized protein	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin-Chitosan	GLYMA19G43580	Q8L8A5	-1.10	-0.39	-1.44	-1.72	GRF1-interacting factor 1	cell proliferation (GO:0008283); leaf development (GO:0048366); transcription, DNA-templated (GO:0006351);
ANE-Chitin-Chitosan	GLYMA12G08131	Q9SV30	-1.23	-0.31	-1.18	-1.57	GATA transcription factor 8	cell differentiation (GO:0030154); circadian rhythm (GO:0007623) ; transcription from RNA polymerase II promoter (GO:0006366);
ANE-Chitin-Chitosan	GLYMA13G29310	F4IHY7	-1.07	0.00	-1.29	-1.71	Protein PLASTID TRANSCRIPTIONALLY ACTIVE 12	response to light stimulus (GO:0009416);
ANE-Chitin	GLYMA06G35330	Q9C7T4	-1.33	-0.77	-1.45	0.00	Transcription factor bHLH96	transcription, DNA-templated (GO:0006351);

ANE-Chitin	GLYMA08G39951	Q7XR02	-1.26	0.85	-1.22	0.00	Transcription factor IBH1	brassinosteroid mediated signaling pathway (GO:0009742); unidimensional cell growth (GO:0009826);
ANE-Chitin	GLYMA08G03910	Q9LYD3	-1.19	-0.57	-1.18	0.07	Dehydration-responsive element-binding protein 3	ethylene-activated signaling pathway (GO:0009873); transcription, DNA-templated (GO:0006351);
ANE-Chitin	GLYMA15G00290	I1MC78	-1.53	0.00	-1.56	-0.73	Uncharacterized protein	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin	GLYMA15G04200	Q6R0E3	-1.44	-0.05	-1.52	-0.81	Telomere repeat-binding protein 5	regulation of transcription, DNA-templated (GO:0006355); transcription, DNA-templated (GO:0006351);
ANE-Chitin	GLYMA15G17550	Q9SUM5	-2.12	-0.95	-2.26	0.96	Uncharacterized protein	regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin	GLYMA08G41000	Q8GW32	-1.79	0.00	-1.08	1.91	Transcription factor PRE6	regulation of growth (GO:0040008); transcription, DNA-templated (GO:0006351);
ANE-Chitin	GLYMA17G19570	Q9FUA4	-1.08	0.00	-1.41	-0.49	Transcription factor SPATULA	fruit development (GO:0010154); transcription, DNA-templated (GO:0006351);
ANE-Chitin	GLYMA06G07911	O00370	-2.16	0.00	-1.53	-0.49	LINE-1 retrotransposable element ORF2 protein	DNA recombination (GO:0006310);
ANE-Chitin	GLYMA08G32190	Q93Y94	-1.01	0.00	-1.20	-0.93	DNA-directed RNA polymerase 1, mitochondrial	transcription, DNA-templated (GO:0006351);
ANE-Chitosan	GLYMA18G03430	Q8S9H7	-1.13	0.00	-0.63	-1.44	Transcription factor DIVARICATA	flower development (GO:0009908); regulation of transcription (GO:0006355);
ANE-Chitosan	GLYMA07G40190	O64722	-1.37	0.00	-0.56	-1.26	Zinc-finger homeodomain protein 3	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA08G46040	Q9SRT2	-1.13	-0.50	-0.33	0.05	Transcription factor bHLH62	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA09G41050	Q9LY00	-1.19	1.18	0.16	0.93	Probable WRKY transcription factor 70	defense response to bacterium (GO:0042742);
ANE	GLYMA17G33891	Q9C5T4	-1.00	-0.43	-0.59	0.00	WRKY transcription factor 18	defense response to bacterium (GO:0042742); defense response to fungus (GO:0050832); response to chitin (GO:0010200);
ANE	GLYMA18G05880	Q6S4P4	-1.18	0.00	-0.08	0.00	Transcription factor RF2b	transcription, DNA-templated (GO:0006351);
ANE	GLYMA05G32430	Q9SXY1	-1.25	0.00	-0.92	-0.99	Chromatin assembly factor 1 subunit FAS2	cell proliferation (GO:0008283); transcription, DNA-templated (GO:0006351);
ANE	GLYMA06G19870	Q9ZWS6	-1.14	-0.76	-0.74	0.00	Two-component response regulator ARR6	response to cytokinin (GO:0009735); transcription, (GO:0006351);
ANE	GLYMA08G03160	F4JRF5	-1.21	0.00	-0.97	-0.80	Homeobox-DDT domain protein RLT3	flower development (GO:0009908); regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA08G04920	Q9C9T2	-1.01	0.00	0.00	0.00	Protein SAR DEFICIENT 1	cellular response to molecule of bacterial origin (GO:0071219); transcription, (GO:0006351);

ANE	GLYMA11G06210	Q9FKZ3	-1.33	-0.77	0.00	0.00	LOB domain-containing protein 36	leaf morphogenesis (GO:0009965); regulation of transcription, (GO:0006355);
ANE	GLYMA12G36970	Q9FLT2	-1.60	0.00	-0.21	-0.33	Inositol polyphosphate multikinase beta	pollen development (GO:0009555); regulation of transcription, (GO:0006355);
ANE	GLYMA15G29870	Q93ZL5	-1.92	-0.72	-0.60	-0.85	Cyclic dof factor 2	flower development (GO:0009908); regulation of transcription (GO:0006355);
ANE	GLYMA17G14030	Q9CAN3	-1.55	-0.60	-0.47	-0.26	Scarecrow-like protein 28	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA18G01040	Q54DA1	-1.41	-0.24	-0.33	0.00	Dr1-associated corepressor homolog	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA18G45391	Q9LZX5	-1.84	0.00	0.00	0.00	Protein LITTLE ZIPPER 2	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA20G35180	Q7XBH4	-1.05	0.00	0.00	0.00	Myb-related protein Myb4	regulation of transcription, DNA-templated (GO:0006355);
ANE	GLYMA08G08220	P42777	-1.13	0.00	-0.78	0.00	G-box-binding factor 4	transcription, DNA-templated (GO:0006351);
ANE	GLYMA11G36580	Q9SVM8	-1.23	0.19	-0.77	-0.17	Glycine-rich RNA-binding protein 2, mitochondrial	cold acclimation (GO:0009631); response to cold (GO:0009409);
ANE	GLYMA15G23160	O00370	-1.30	0.00	-0.53	-0.39	LINE-1 retrotransposable element ORF2 protein	DNA recombination (GO:0006310);
ANE	GLYMA16G01911	B5X570	-1.00	0.00	0.00	-0.16	NAC domain-containing protein 14	transcription, DNA-templated (GO:0006351);
ANE	GLYMA16G13200	Q8L6J4	-1.02	0.27	-0.81	-0.80	DNA-directed RNA polymerase 1A	transcription, DNA-templated (GO:0006351);
ANE	GLYMA18G00480	Q9SVM8	-1.65	0.14	-0.97	-0.42	Glycine-rich RNA-binding protein 2, mitochondrial	response to cold (GO:0009409);
ANE	GLYMA18G51160	Q8L6J3	-1.12	0.00	-0.74	-0.29	DNA-directed RNA polymerase 2B, chloroplastic/mitochondrial	transcription, DNA-templated (GO:0006351);
ANE	GLYMA06G22840	O13046	-1.40	0.00	-0.68	0.00	WD repeat and HMG-box DNA-binding protein 1	No-GOBP

Table 43: ANE down-regulated genes associated with chromatin modulators.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin	GLYMA11G36620	O23255	-1.51	-1.23	-1.70	0.02	Adenosylhomocysteinase 1	methylation-dependent chromatin silencing (GO:0006346);
ANE-Chitin-Chitosan	GLYMA05G32210	P68428	-2.01	0.28	-1.42	-1.05	Histone H3.2	No-GOBP
ANE-Chitin-Chitosan	GLYMA11G04301	Q8VZ17	-1.16	-0.11	-1.03	-1.48	Histone-lysine N-methyltransferase, H3 lysine-9 specific SUVH6	histone methylation (GO:0016571);
ANE-Chitin-Chitosan	GLYMA12G05910	P68428	-1.48	0.00	-1.40	-1.17	Histone H3.2	No-GOBP
ANE-Chitin-Chitosan	GLYMA12G30600	Q8LJS2	-1.97	0.00	-1.82	-1.01	Histone deacetylase HDT1	chromatin modification (GO:0016568);
ANE-Chitin-Chitosan	GLYMA13G24180	P68428	-1.09	0.06	-1.40	-1.03	Histone H3.2	No-GOBP
ANE-Chitin-Chitosan	GLYMA17G05340	Q8LJS2	-2.10	0.82	-1.24	-1.44	Histone deacetylase HDT1	chromatin modification (GO:0016568);
ANE-Chitin-Chitosan	GLYMA19G33360	O65759	-1.30	0.00	-1.05	-1.02	Histone H2AX	No-GOBP
ANE-Chitin-Chitosan	GLYMA02G04060	Q9LXE5	-1.56	-0.79	-1.75	- 1.98	DNA (cytosine-5)-methyltransferase DRM1	defense response to fungus (GO:0050832);
ANE-Chitin-Chitosan	GLYMA11G27622	F4IXE7	-1.12	0.00	-1.24	-1.18	Increased DNA methylation 1	RNA-directed DNA methylation (GO:0080188);
ANE-Chitin	GLYMA07G32360	P68428	-1.38	0.00	-1.40	-0.83	Histone H3.2	No-GOBP
ANE-Chitin	GLYMA04G07841	Q6YXZ7	-1.26	0.17	-1.16	-0.03	Protein arginine N-methyltransferase 5	(regulation of transcription, DNA-templated (GO:0006355);
ANE-Chitin	GLYMA04G16850	Q0WVD6	-1.10	-0.05	-1.68	-0.11	Probable protein arginine N-methyltransferase 3	histone arginine methylation (GO:0034969);
ANE-Chitin	GLYMA11G19480	Q8LJS2	-1.63	0.15	-1.65	-0.41	Histone deacetylase HDT1	chromatin modification (GO:0016568);
ANE-Chitin	GLYMA15G03710	P68428	-1.33	0.22	-1.12	-0.48	Histone H3.2	No-GOBP
ANE-Chitin	GLYMA15G34870	P68428	-2.03	0.00	-1.25	-0.48	Histone H3.2	No-GOBP
ANE-Chitin	GLYMA20G33320	Q1S9I9	-2.30	-0.12	-2.26	-0.97	Probable histone H2B.1	No-GOBP
ANE-Chitin	GLYMA13G16500	Q70Z17	-1.39	0.00	-1.49	-0.87	Nucleosome assembly protein 1;3	nucleosome assembly (GO:0006334);

ANE-Chitin	GLYMA17G06170	Q70Z17	-1.41	-0.10	-1.36	-0.93	Nucleosome assembly protein 1;3	nucleosome assembly (GO:0006334);
ANE-Chitin	GLYMA08G36280	Q8VZ79	-2.14	-0.82	-1.66	-0.78	Protein INVOLVED IN DE NOVO 2	DNA methylation (GO:0006306);
ANE-Chitin	GLYMA13G26240	Q6YSJ5	-1.21	-0.02	-1.68	-0.94	Protein argonaute 16	gene silencing by RNA (GO:0031047);
ANE-Chitosan	GLYMA03G30440	O65759	-1.81	0.16	-1.00	-1.02	Histone H2AX	No-GOBP
ANE	GLYMA02G38921	P0CG89	-1.03	0.00	-0.65	-0.13	Histone H4	nucleosome assembly (GO:0006334);
ANE	GLYMA10G03640	Q84MP7	-2.32	0.24	-0.96	-0.34	Probable histone H2A variant 3	chromatin silencing (GO:0006342);
ANE	GLYMA11G13940	P68428	-2.00	0.19	-0.95	-0.57	Histone H3.2	No-GOBP
ANE	GLYMA11G15497	P0CG89	-1.26	0.00	-0.59	-0.20	Histone H4	nucleosome assembly (GO:0006334);
ANE	GLYMA11G24323	P0CG89	-1.30	0.00	-0.84	-0.46	Histone H4	nucleosome assembly (GO:0006334);
ANE	GLYMA12G09000	Q8LJS2	-1.37	0.04	-0.99	-0.54	Histone deacetylase HDT1	chromatin modification (GO:0016568);
ANE	GLYMA17G29925	P0CG89	-1.25	0.08	-0.67	-0.51	Histone H4	nucleosome assembly (GO:0006334);
ANE	GLYMA18G50120	P68428	-1.29	0.23	-0.62	-0.82	Histone H3.2	No-GOBP
ANE	GLYMA19G42240	Q94F49	-2.04	0.00	-0.75	-0.35	Probable histone H2A.5	chromatin silencing (GO:0006342);
ANE	GLYMA20G37630	Q8RWL4	-1.48	-0.68	-0.19	-0.95	Increased DNA methylation 2	DNA demethylation (GO:0080111);

Table 44: ANE down-regulated genes in phytohormone-regulated processes.

Treatments	Gene IDs	Uniprot IDs	ANE	Alginate	Chitin	Chitosan	Annotations	Biological processes
ANE-Alginate-Chitin-Chitosan	GLYMA15G00450	Q93ZB2	-1.83	-1.54	-3.29	-3.15	Ent-kaurene oxidase, chloroplastic	ent-kaurene oxidation to kaurenoic acid (GO:0010241); gibberellin biosynthetic process (GO:0009686);
ANE-Alginate-Chitin	GLYMA12G30770	Q05999	-1.50	-2.09	-1.18	-0.57	Serine/threonine-protein kinase D6PKL3	auxin-activated signaling pathway (GO:0009734); basipetal auxin transport (GO:0010540);
ANE-Alginate-Chitin	GLYMA17G37750	P46689	-1.53	-2.48	-3.50	0.70	Gibberellin-regulated protein 1	response to abscisic acid, brassinosteroid, gibberellin (GO:0009739); unidimensional cell growth (GO:0009826);
ANE-Chitin-Chitosan	GLYMA16G08070	Q17TI5	-1.52	0.00	-1.13	-2.11	Protein BREVIS RADIX	auxin-activated signaling pathway (GO:0009734); cytokinin-activated signaling pathway (GO:0009736); response to abscisic acid (GO:0009737);
ANE-Chitin-Chitosan	GLYMA13G01870	Q9SYT0	-2.57	0.12	-2.62	-1.69	Annexin D1	cellular oxidant detoxification (GO:0098869); potassium ion export (GO:0071435); response to abscisic acid (GO:0009737); response to cold (GO:0009409); (GO:0006970); response to salt stress (GO:0009651);

ANE-Chitin	GLYMA12G03990	P33081	-1.71	-0.51	-1.15	0.70	Auxin-induced protein 15A	auxin-activated signaling pathway (GO:0009734);
ANE-Chitin	GLYMA03G28740	Q6NMQ7	-2.49	-0.50	-3.28	-0.43	Gibberellin-regulated protein 6	gibberellic acid mediated signaling pathway (GO:0009740); response to sugar
ANE-Chitin	GLYMA04G33610	P46690	-1.53	-0.62	-2.16	1.31	Gibberellin-regulated protein 4	cell redox homeostasis (GO:0045454); gibberellic acid mediated signaling pathway (GO:0009740); response to gibberellin (GO:0009739);
ANE-Chitin	GLYMA06G20830	Q6NMQ7	-1.47	-0.71	-1.88	1.01	Gibberellin-regulated protein 6	gibberellic acid mediated signaling pathway (GO:0009740); response to sugar
ANE-Chitosan	GLYMA17G05760	Q9ZRA4	-2.17	0.18	-0.99	-2.06	Auxin-binding protein ABP19a	auxin-activated signaling pathway (GO:0009734);
ANE-Chitosan	GLYMA02G42990	Q9FLB1	-1.25	-0.43	-0.48	-1.17	Abscisic acid receptor PYL5	abscisic acid-activated signaling pathway (GO:0009738);
ANE	GLYMA02G16890	P33491	-1.02	0.00	-0.60	-0.90	Auxin-binding protein T92	auxin-activated signaling pathway (GO:0009734);
ANE	GLYMA04G40170	Q9SG92	-2.03	-0.47	0.00	-0.27	Methylesterase 17	indoleacetic acid conjugate metabolic process (GO:0033473);
ANE	GLYMA02G05530	P32295	-1.10	0.00	-0.71	-0.35	Indole-3-acetic acid-induced protein ARG7	response to auxin (GO:0009733);
ANE	GLYMA14G15820	Q9LFM5	-1.47	0.00	-0.75	-0.43	Probable indole-3-pyruvate monooxygenase YUCCA4	auxin biosynthetic process (GO:0009851); cotyledon development (GO:0048825); inflorescence development (GO:0010229); oxidation-reduction process (GO:0055114); positive regulation of flower development (GO:0009911);
ANE	GLYMA16G09760	Q94BT2	-2.10	-0.73	-0.41	0.21	Auxin-induced in root cultures protein 12	auxin-activated signaling pathway (GO:0009734); oxidation-reduction process (GO:0055114);
ANE	GLYMA01G35180	Q17TI5	-1.76	-0.42	-0.22	-0.80	Protein BREVIS RADIX	auxin-activated signaling pathway (GO:0009734); cytokinin-activated signaling pathway (GO:0009736); response to abscisic acid (GO:0009737);
ANE	GLYMA17G16420	Q944A7	-1.10	-0.08	-0.65	-0.03	Probable serine/threonine-protein kinase	brassinosteroid mediated signaling pathway (GO:0009742); response to abscisic acid (GO:0009737);
ANE	GLYMA18G49400	P46690	-2.10	-0.90	1.01	1.91	Gibberellin-regulated protein 4	response to gibberellin (GO:0009739);
ANE	GLYMA09G37290	Q6NMQ7	-2.20	-0.66	-0.66	3.49	Gibberellin-regulated protein 6	gibberellic acid mediated signaling pathway (GO:0009740); response to sugar
ANE	GLYMA07G37240	P26987	-2.56	-0.82	0.84	0.00	Stress-induced protein SAM22	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE	GLYMA07G37265	P26987	-2.13	-0.84	0.61	0.00	Stress-induced protein SAM22	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE	GLYMA07G37270	P26987	-2.22	-0.24	0.69	0.24	Stress-induced protein SAM22	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952); response to biotic stimulus (GO:0009607);
ANE	GLYMA17G03365	P26987	-1.08	-0.27	1.08	0.00	Stress-induced protein SAM22	abscisic acid-activated signaling pathway (GO:0009738); defense response (GO:0006952); response to biotic stimulus (GO:0009607);