

CHARACTERIZATION OF A LIPASE IN ARABIDOPSIS DEFENSE

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ABSTRACT

Plant defense responses are constitutively activated in the *Arabidopsis thaliana* *ssi2* mutant plant. In addition, the *ssi2* mutant allele confers a dwarf phenotype. The *SSI2* gene encodes a stearoyl-ACP-desaturase, which converts stearic acid (18:0) to oleic acid (18:1), suggesting a role for lipids in plant defense. Microarray analysis identified several genes which encode putative acyl hydrolases/lipases that are expressed at elevated levels in the leaves of *ssi2*, in comparison to the wild type plant. One gene in particular, At5g14180, was expressed at 60-fold greater level in *ssi2* than in the wild type plant. To study the involvement of At5g14180 in plant defense and lipid metabolism, two transgenic lines containing T-DNA insertions within the At5g14180 gene were identified. These two T-DNA insertional alleles of the At5g14180 gene attenuate the *ssi2*-conferred heightened resistance to a virulent strain of *Pseudomonas syringae* pv. *maculicola* in the *ssi2* At5g14180 double mutant plant. Furthermore, pathogen growth was enhanced in the At5g14180 single mutant plants, as compared to the wild type plant. Profiling of lipid composition in leaf tissue identified changes in the lipid composition between the At5g14180 mutant and wild type plants, suggesting that the At5g14180 encoded protein may impact lipid metabolism in *Arabidopsis* leaves.

TABLE OF CONTENTS

Table of contents.....	iii
List of figures.....	iv
List of tables.....	vi
Acknowledgements.....	vii
Chapter I. Introduction and literature review.....	1
Chapter II. Characterization of the At5g14180 T-DNA insertion mutation	
Introduction.....	15
Materials and Methods.....	18
Results.....	25
Discussion.....	33
Figure Legends.....	37
Figures	44
Chapter III. Quantitative analysis of Arabidopsis galactolipids and oxylipins in	
Introduction.....	55
Materials and Methods.....	60
Results	63
Discussion.....	66
Figure Legends.....	69
Figures	72
Chapter IV. Conclusions and future directions.....	78
References.....	81
Appendix.....	89

LIST OF FIGURES

Chapter II

Figure 2 – 1. Genes encoding lipases of poorly defined metabolic function are expressed at elevated levels in the <i>ssi2</i> mutant.....	44
Figure 2 – 2. At5g14180 protein sequence.....	45
Figure 2 – 3. PCR based strategy for identification of plants with T-DNA insertion in At5g14180 gene.....	46
Figure 2 – 4. PCR products confirming homozygosity of At5g14180 T-DNA insertion mutant lines.....	47
Figure 2 – 5. Systemic Acquired Resistance is not impaired in At5g14180 T-DNA insertion mutant lines.....	48
Figure 2 – 6. Growth of Psm in At5g14180 T-DNA insertion mutants.....	49
Figure 2 – 7. At5g14180 expression is induced upon <i>P. syringae</i> pv. <i>maculicola</i> infection.....	50
Figure 2 – 8. Response to SA is not impaired in At5g14180 T-DNA insertion mutants.....	51
Figure 2 – 9. At5g14180 expression is induced upon SA treatment.....	52
Figure 2 – 10. PCR products confirming homozygosity of Salk_101919 <i>ssi2</i> double mutant.....	53
Figure 2 – 11. Growth of <i>P. syringae</i> pv. <i>maculicola</i> in Salk_101919 <i>ssi2</i> double mutant.....	54
Figure 2 – 12. At5g14180 is constitutively expressed in the <i>ssi2</i> mutant.....	55

Chapter III

Figure 3 – 1. Profile of 16:0 free fatty acid content in At5g14180 T-DNA insertion mutant lines.....	72
Figure 3 – 2. Profile of 18:0 free fatty acid content in At5g14180 T-DNA insertion mutant lines.....	73

Figure 3 – 3. Profile of 18:1 free fatty acid content in At5g14180 T-DNA insertion mutant lines.....	74
Figure 3 – 4. Profile of 18:2 free fatty acid content in At5g14180 T-DNA insertion mutant lines.....	75
Figure 3 – 5. Profile of jasmonate content in At5g14180 T-DNA insertion mutant lines.....	76
Figure 3 – 6. Ratio of 36 and 34 carbon molecular species of PC and PE in At5g14180 T-DNA insertion lines.....	77

LIST OF TABLES

Table A – 1. Oxylipin Profile – GC/MS Experiment 1.....	90
Table A – 2. Oxylipin Profile – GC/MS Experiment 2.....	91
Table A – 3. Oxylipin Profile – GC/MS Experiment 3.....	92
Table A – 4. Total lipid species – ESI MS/MS Experiment 1.....	93
Table A – 5. Total lipid species – ESI MS/MS Experiment 1.....	116
Table A – 6. Total lipid species – ESI MS/MS Experiment 1.....	140
Table A – 7. PCR Primers.....	164

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CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW

Plants are constantly exposed to the threat of consumption and habitation by a wide variety of organisms including bacteria, viruses, fungi, insects and herbivores. In response to this threat, plants have evolved sophisticated defense mechanisms. The specific response to biotic stress has been characterized in the genetic model plant *Arabidopsis thaliana* and is characterized by specific biological responses and the involvement of a variety of phytohormone signaling pathways (Pieterse et. al., 2001).

Defense Mechanisms in Plants

Plants possess several pre-formed barriers that serve as a passive first line of defense against pathogen infection. The extracellular matrix of the epidermis in plants is covered with a hydrophobic cuticle, which protects the plant from the outside environment and serves as a barrier to pathogen infection (Nawrath, 2006). The waxy layer covering the plant cuticle contains some anti-microbial activity, inhibiting pathogen infection (Nawrath, 2006). The plant cell wall also functions as a preformed barrier to infection, as it must be penetrated in order for infection to occur (Jones and Takemoto, 2003). Upon pathogen infection plants initiate active defense responses of two types: localized innate immunity and systemic immunity. Localized innate immunity consists of the local defense response at the site of attack or infection. In order for the plant defense response to occur, recognition of the invading species must occur. The plant is able to recognize the invading pathogen by at least two molecular methods. One method, classified as gene-for-gene resistance of one or more plant *Resistance (R)* gene(s)

corresponding to an *Avirulence (Avr)* gene(s) expressed by the pathogen (Tang et al., 1996; Nimchuk et al., 2001; Ausubel, 2005). Several of these *Avr*-encoded proteins contribute to pathogenicity in susceptible host. Avr recognition is mediated by the *R* gene product (Ausubel, 2005). Recognition can also operate by means of Pathogen (or Pattern) Recognition Receptors (PRRs) that recognize Pathogen-Associated Molecular Patterns (PAMPs) (Ausubel, 2005). PAMPs include lipopolysaccharide, peptidoglycan and bacterial flagellin (Ausubel, 2005). Often, this recognition results in activation of the hypersensitive response (HR), characterized by tissue collapse, oxidative burst and programmed cell death at the site of pathogen ingress. The HR retards spread of the pathogen from the initial infection site (Hammond-Kosack and Jones, 1996). Subsequently, in many cases of plant-pathogen interaction, defenses are activated throughout the plant resulting in systemic immunity to subsequent attack by pathogen (Métraux et al., 2002; Durrant and Dong, 2004).

Systemic immunity encompasses the induced defenses activated in uninfected plant tissues. This induced defense, commonly referred to as systemic acquired resistance (SAR) confers broad spectrum disease resistance throughout the plant (Hunt and Ryals, 1996; Durrant and Dong, 2004). SAR activation is accompanied by the induction of *PATHOGENESIS-RELATED (PR)* genes, which encode PR proteins, some of which possess anti-microbial activities (Hunt and Ryals, 1996). The expression of these PR genes serves as a reliable marker to assess the activation of SAR (Ward et al., 1991; Yalpani et al., 1991). SAR activation is accompanied by an increase in levels of salicylic acid (SA) at the initial site of pathogen infection, as well as throughout systemic

tissues (Rasmussen et al., 1991). The application of SA or benzothiadiazole, a synthetic functional analog of SA has been shown to produce SAR-like induced resistance to the tissue to which it is applied (Hunt and Ryals, 1996). SAR can also be induced chemically by 2,6-dichloroisonicotinic acid (INA), another functional analog of SA (Métraux et al., 1991; Oostendorp et al., 2001; Dong, 2004). SA accumulation in the distal organs is required for expression of SAR (Friedrich et al., 1995). By expressing the bacterial *nahG* gene encoding salicylate hydroxylase, an enzyme which catalyzes the conversion of SA to catechol, in transgenic tobacco plants, the accumulation of free SA was restricted to very low levels and the plants were shown to be defective in SAR (Friedrich et al., 1995). Similarly, SAR was compromised in the *Arabidopsis sid2* mutant, which encodes a key SA biosynthetic enzyme (Nawrath et al., 1999; Wildermuth et al., 2001). Although SA is required for the expression of SAR grafting experiments with the NahG tobacco plants have suggested that SA is not the long distance signal that is transmitted from the pathogen inoculated leaf to the distal organs where SAR is expressed (Friedrich et al., 1995).

In addition to the systemic immunity of SAR, two other types of induced resistance have been characterized in plants. These include Wound Inducible Resistance (WIR) (Kessler and Baldwin, 2002) and Induced Systemic Resistance (ISR) (Van Loon et al., 1998). WIR is induced by wounding associated with herbivore attack (Kessler and Baldwin, 2002). ISR is induced by nonpathogenic rhizosphere bacteria colonizing plant roots (Van Loon et al., 1998). Contrary to the involvement of salicylic acid as a signaling molecule in SAR, ISR and WIR alternatively utilize jasmonic acid (JA) and ethylene in

defense signaling (Van Loon et al., 1998; Kessler and Baldwin, 2002). In the WIR response, predation by insects induces the accumulation of JA (Creelman et al., 1992; Kessler and Baldwin, 2002). This JA accumulation induces genes involved in plant defense including genes encoding protease inhibitors, which help protect the plant from insect damage (Ranjan and Lewark, 1992) and the *CHS*, *PAL*, *HMGR* genes involved in phytoalexin biosynthesis (Creelman et al., 1992). JA is structurally related to prostaglandins, which produce tissue inflammation at sites of infection or tissue injury in mammals (Straus and Glass, 2001). Studies with the *Arabidopsis fad3-2 fad7-2 fad8* triple mutant plant, which are defective in the synthesis of linolenic acid, the precursor for JA, demonstrated an essential role for JA in WIR (McConn et al., 1997). These mutants produced negligible levels of JA and were shown to suffer from high mortality rates as compared to wild type plants when exposed to the larvae of a saprophagous fungal gnat, *Bradysia impatiens* (McConn et al., 1997). Application of methyl jasmonate to the *fad3-2 fad7-2 fad8* mutants restored resistance against *Bradysia impatiens* (McConn et al., 1997).

ISR is activated by strains of rhizosphere bacteria collectively termed Plant Growth Promoting Rhizobacteria (PGPR) whose application to plant roots can promote plant growth and improve plant health during periods of stress by suppressing activity of pathogens (Van Loon et al., 1998). The ISR mechanism operates independently of SA signaling and is not accompanied by elevated expression of *PR* genes (Pieterse et al., 1998). The ISR independence of the SA pathway was shown using transgenic *Arabidopsis* plants expressing the bacterial *nahG* gene, which catalyzes the conversion of

SA to catechol (Pieterse et al., 1998). ISR was not compromised in NahG plants. In contrast, ISR was compromised in JA signaling mutants (Pieterse et al., 1998). The *jar1* mutant, which has lowered sensitivity to methylJA, while able to activate SAR, was unable to induce rhizobacteria-mediated ISR (Pieterse et al., 1998).

JA- and ethylene-dependent defense responses have also been implicated in plant defense against necrotrophic pathogens, specifically *Botrytis cinerea*. Infection of the *Arabidopsis coi1* (*coronitine insensitive*) mutant, which is deficient in JA signaling, exhibited enhanced susceptibility to *B. cinerea* infection (Thomma et al., 1998). Similarly enhanced susceptibility to *B. cinerea* was observed in *Arabidopsis ein2-1* (*ethylene insensitive2*) mutants defective in ethylene signal transduction (Thomma et al., 1999). Further, neither the *coi1* nor the *ein2* mutants were able to induce expression of the JA- and ethylene-dependent antimicrobial Defensin gene *PDF1.2* (*PLANT DEFINSINI.2*) in response to *B. cinerea* infection (Thomma et al., 1998, 1999).

Together with SAR, WIR and ISR compose plant systemic induced resistance (SIR). The SIR response comprises an intricate signaling network involving SA, JA and ethylene (Spoel et al., 2003). Although each pathway is involved in separate defense mechanisms, the SA-dependent and JA-dependent pathways have been shown to cross-communicate in order to regulate defense responses to best suit the needs of the plant (Feys and Parker, 2000; Pieterse et al., 2001). The simultaneous induction of SAR and ISR has been shown to have an additive effect on the level of induced defense (Van Wees et al., 2000). SA and JA have been shown to have an antagonistic relationship, also

(Pena-Cortes et al., 1993; Doares et al., 1995; Felton et al., 1999). Application of SA or its derivatives, aspirin, for example, blocked JA biosynthesis and the action of JA in wound signaling (Pena-Cortez et al., 1993; Doares et al., 1995). Further, this antagonism has been shown *in vivo* using transgenic tobacco plants with silenced expression or overexpression of phenylalanine ammonialyase (PAL), an enzyme that catalyzes the first committed step of phenylpropanoid synthesis, from which SA is derived (Felton et al., 1999). PAL-silenced transgenic tobacco plants showed higher levels of JA induced by wounding than in the PAL-overexpression line and in PAL-modified lines, constitutive JA levels correlated inversely with constitutive SA levels (Felton et al., 1999). JA antagonism to SA was shown using *Arabidopsis* mutants blocked in JA signaling (Kloek et al., 2000). Infection of the *coi1* mutant with *Pseudomonas syringae* resulted in hyperactivation of SA signaling (Kloek et al., 2000).

A key regulatory protein, NON-EXPRESSOR OF PR1 (NPR1), also known as NIM1 (NON-INDUCIBLE IMMUNITY1) and SAI1 (SALICYLIC ACID-INSENSITIVE1) that modulates SA signaling was identified via genetic screens for mutants compromised in SA signaling (Cao et al., 1994; Delaney et al., 1995; Glazebrook et al., 1996; Shah et al., 1997). Mutant *npr1* plants were able to accumulate SA in response to pathogen infection, but were insensitive to SA, unable to express *PR* genes and were deficient in SAR defense activity. Ectopic expression of the wild type *NPR1* gene in the *npr1* mutant background, resulted in heightened resistance to *Pseudomonas syringae* (Cao et al., 1997) thus confirming that *NPR1* encodes a positive regulator of SA signaling. In addition, ectopic overexpression of *NPR1* in *Arabidopsis* enhances

resistance to *Pseudomonas syringae*, the oomycete *Hyaloperonospora parasitica* and the fungal pathogen *Fusarium graminearum* (Cao et al., 1998; Makandar et al., 2006). Cloning of the *NPR1* gene established that it encodes a novel protein containing two protein-protein interaction domains, a BTB/POZ domain and an ankyrin-repeat domain (Cao et al., 1997). Upon induction of SAR, the *NPR1* protein is translocated to the nucleus (Kinkema et al., 2000). Once *NPR1* is localized in the nucleus, it interacts with a subset of the TGA2 family of bZIP transcription factors to transduce the SA signal required for *PR* gene activation (Fan and Dong, 2002). Consequently, *NPR1* function is required for the transduction of the SA signal and *PR* gene activation in SAR.

In addition to its function in SAR, *NPR1* modulates the cross-talk between the SA- and JA-dependent pathways (Spoel et al, 2003) and is required for the function of ISR induced resistance (Pieterse et al., 1998). While *NPR1* translocated to the nucleus regulates SA signaling and SAR induction, *NPR1* in the cytosol negatively regulates JA-responsive gene expression (Spoel et al., 2003). Although the mechanism by which this regulation occurs is still unknown, it is hypothesized that it occurs either by *NPR1* inhibition of positive regulators of JA responsive genes or alternatively by facilitating the transport of negative regulators of JA responsive genes into the nucleus (Spoel et al., 2003). *NPR1* necessity in the ISR defense response was demonstrated by the lack of *P. fluorescens* WCS417r-mediated ISR activity in *npr1* mutant plants (Pieterse et al., 1998). Accordingly, *NPR1* has been shown to have a significant role in plant systemic induced resistance and has a vital central role in the cross-talk and interconnectivity of plant signaling pathways. Ectopic expression of Arabidopsis *NPR1* in rice, tomato and wheat

promotes resistance against a variety of pathogens, suggesting the *NPR1*-modulated pathway is functional in plants other than *Arabidopsis* (Chern et al., 2005; Lin et al., 2004; Makandar et al., 2006).

Although the *NPR1* gene has been shown to be required for SAR related defense activity and SA induced expression of *PR* genes, an *NPR1*-independent defense pathway has been shown to exist (Shah et al., 2001). In order to identify other components of this *NPR1*-independent defense pathway, the *npr1-5* mutant plant was mutagenized with Ethyl Methylsulfonate (EMS) and screened for mutants exhibiting constitutive *PR* gene expression, leading to the discovery of the *ssi* (*suppressor of SA-insensitivity 1*) mutants *ssi1*, *ssi2* and *ssi4* (Shah et al., 1999, 2001; Shirano et al., 2002). In addition to the constitutive expression of the *PR* genes, the mutant plants constitutively accumulate elevated levels of SA and exhibit heightened resistance to a variety of pathogens including *Pseudomonas syringae* and *Peronospora parasitica* (Kachroo et al., 2001, 2003, 2004; Nandi et al., 2003; Shah et al., 1999, 2001). In addition, the *ssi2* mutant also exhibits heightened resistance to Cucumber mosaic virus and the phloem-feeding insect, green peach aphid (Pegadaraju, 2005; Sekine et al., 2004), displays a dwarf phenotype and spontaneously exhibit lesions and cell death (Shah et al., 2001). Evidence of an *NPR1*-independent signaling pathway was illustrated by studies of the *ssi1 npr1* and *ssi2 npr1* double mutant plant (Shah et al., 1999, 2001). The *ssi1* and *ssi2* mutant containing the wild type *NPR1* allele accumulated greater levels of *PR1* gene transcripts than *ssi1 npr1* and *ssi2 npr1* double mutant plants, respectively, indicating the function of an

NPR1-dependent pathway functioning additively with the NPR1-independent pathway (Shah et al., 1999, 2001).

Lipids in Plant Defense

Cloning of the *SSI2* gene revealed that it encodes a plastid-localized stearoyl-acyl-carrier-protein desaturase, which converts stearic acid (18:0) to oleic acid (18:1) (Kachroo et al., 2001). This stearoyl-ACP-desaturase is a member of the soluble fatty acid desaturase family, a family of enzymes that function as key regulators of fatty acid desaturation (Kachroo et al., 2001). Consequently, gas chromatography-mass spectrometry (GC-MS) revealed that the leaves of *ssi2* mutant plants contain elevated levels of 18:0 fatty acids and reduced level of 18:1 in comparison to the wild type plant (Kachroo et al., 2001). Stearoyl-ACP-desaturase catalyzes the first step in the pathway producing linolenic acid, a precursor for the JA defense-signaling molecule (Kachroo et al., 2001). Accordingly, the activation of JA-inducible defense responses was assessed in the *ssi2* mutant, and JA-mediated resistance to *B. cinerea* as well as the JA-defense pathway associated expression of *PDF1.2* was found to be impaired in the *ssi2* mutant (Kachroo et al., 2001). A contributory role for SA in antagonizing the JA-defense pathway resulting in susceptibility to *B. cinerea* was ruled out by the preservation of enhanced susceptibility to *B. cinerea* and the inability to express *PDF1.2* in *ssi2 nahG* double mutants (Kachroo et al., 2001; Shah et al., 2001). However, the JA-dependent expression of *PDF1.2* in the *ssi2* mutant was restored by co-application of 18:1, suggesting the involvement of an 18:1-derived factor in the JA-inducible *PDF1.2* expression (Kachroo et al., 2001).

Genetic studies in *Arabidopsis* have provided further support for the role of lipids in plant defense against biotic stress. For example, loss-of-function mutations in the *SFD1 (SUPPRESSOR OF FATTY ACID DESATURASE DEFICIENCY1)* gene compromised the activation of SAR in response to inoculation with an avirulent bacterial pathogen (Nandi et al., 2004). SAR conferred accumulation of SA, elevated expression of *PRI* and heightened resistance to pathogen were attenuated in the distal leaves of *sfd1* mutants that were locally inoculated with avirulent pathogen. However, basal resistance to avirulent and virulent strains of *P. syringae* was not compromised and the locally inoculated leaves exhibit elevated levels of SA and expression of *PRI* (Nandi et al., 2004). Furthermore, application of the SA analog BTH was able to enhance disease resistance in the *sfd1* mutant, indicating that *sfd1* is not defective in SA signaling *per se* (Nandi et al., 2004). Cloning of *SFD1* revealed it encodes a putative dihydroxyacetone phosphate (DHAP) reductase involved in glycerolipid metabolism (Nandi et al., 2004). In the *sfd1* mutant plant, plastid glycerolipid composition was altered; *sfd1* leaves had lower content of plastid-synthesized MGDG (monogalactosyldiacylglycerol) and DGDG (digalactosyldiacylglycerol), suggesting *SFD1* involvement in lipid metabolism and implicating the involvement of *SFD1* derived lipids in the activation of the SAR induced defense response (Nandi et al., 2004).

Further support for the involvement of a lipid-based signaling molecule in SAR comes from studies involving the *Arabidopsis dir1(defective in induced resistance 1)* mutant. Like *sfd1*, the *dir1* mutant is unable to develop SAR associated heightened

resistance to bacterial or oomycete pathogens, but does exhibit local resistance to avirulent and virulent strains of *P. syringae* (Maldonado et al., 2002). Petiole exudates from *dir1* leaves challenged with avirulent pathogen are unable to elicit *PR1* expression in wild-type leaves, but petiole exudates from wild-type plants challenged with avirulent pathogen are sufficient to induce *PR1* expression in *dir1* plants, indicating *dir1* is likely defective in the production or transmission of an essential mobile signal (Maldonado et al., 2002). *DIR1* encodes a putative apoplastic lipid transfer protein and it is likely that *DIR1* promotes long distance signaling via interaction with a lipid-derived molecule (Maldonado et al., 2002).

In *Arabidopsis*, glycerolipid biosynthesis occurs through two distinct pathways: the prokaryotic pathway, located on the plastid inner envelope and the eukaryotic pathway, localized on the endoplasmic reticulum (Somerville et al., 2000). Both pathways are initiated by the formation of phosphatidic acid that requires two acylation reactions transferring fatty acids from acyl-ACP or acyl-CoA to glycerol-3-phosphate, by the prokaryotic and eukaryotic pathways, respectively. In the prokaryotic pathway, PA is further channeled into the synthesis of other chloroplast lipids, including phosphatidylglycerol (PG), monogalactosyldiacylglycerol (MGDG), digalactosyldiacylglycerol (DGDG) and sulfoquinovosyldiacylglycerol (SQDG). PA is used to synthesize PG or can be converted to diacylglycerol (DAG) by means of a phosphatidic acid-phosphatase (Somerville et al., 2000). DAG serves as the precursor for the synthesis of MGDG, DGDG and SQDG.

In the eukaryotic pathway, fatty acids exported from the chloroplast are used to synthesize PA. This ER-derived PA can be further channeled to synthesis of phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylinositol (PI) and phosphatidylserine (PS). Some PC or PC-derived products are transported back to the plastid where they are used to synthesize some species of plastid-localized lipids – MGDG, DGDG and SQDG using an acyl-glycerol component (Mongrand et al., 2000). Examples of modified PC plastid-localized lipids include 36:6 MGDG and 36:6 DGDG (Mongrand et al., 2000).

Lipid Modifying Enzymes

The expanding role of lipids in plant defense has focused some attention on the role of lipid-modifying enzymes. In particular, phospholipsases, enzymes which catalyze the hydrolysis of phospholipids into fatty acids and other lipophilic substances, have been implicated as having a role in plant response to pathogen infection. There are four major classes of phospholipases, differentiated by the type of reaction that they catalyze, designated A, B, C and D. Three classes, A, C, and D have been shown to be involved in the plant defense response (Dhondt et al., 2002; de Jong et al., 2004; de Torres et al., 2002).

Phospholipase A's catalyze the phospholipid hydrolysis to produce a free fatty acid and a lysophospholipid (Ryu, 2004). Phospholipase A involvement has been implicated in JA biosynthesis during the activation of defense gene responses by releasing a fatty acid precursor of JA (Dhondt et al., 2002). Further, in tobacco plants,

microbial elicitors and exposure to pathogen have been shown to induce the expression of 3 tobacco genes *NtPat1*, *NtPat2* and *NtPat3*, encoding putative phospholipase A enzymes in elicitor-treated or pathogen inoculated plant tissue (Dhondt et al., 2002). The expression of these putative lipase-encoding genes precedes the accumulation of JA (Dhondt et al., 2002). Additionally, the infection of tobacco plants by *Tobacco Mosaic Virus* (TMV) was shown to activate Phospholipase A activity (Dhondt et al., 2002).

Phospholipase C releases a diacylglycerol and a phosphate containing head group by catalyzing a phosphate cleavage reaction. DAG can further be processed by a DAG kinase to yield PA. Phospholipase C activity was shown to be involved in short-term accumulation of PA during an incompatible gene interaction involving the Cf4 tomato gene and the *Cladosporium fulvum* AVR4 elicitor (de Jong et al., 2004). In this incompatible interaction, phospholipase C activity was shown to act upstream of the oxidative burst (de Jong et al., 2004).

Phospholipase D catalyzes the release of PA from phospholipids. Phospholipase D activity has been shown to be stimulated in response to *Pseudomonas* challenge in Arabidopsis (de Torres et al., 2002), and by the addition of microbial elicitors to tomato suspension cells (Van der Luit et al., 2000) and to rice suspension cells (Yamaguchi et al., 2003). Moreover, phospholipase D enzymatic activity is stimulated in response to H₂O₂ and oleic acid, which modulate defense responses in Arabidopsis (Zhang et al., 2003). Additionally, a mutant lacking in phospholipase D activity was shown to have a

greater sensitivity to H₂O₂ activity and cell death than the wild-type plant (Zhang et al., 2003)

In addition, two genes, *PAD4* (*PHYTOALEXIN-DEFICIENT4*) and *EDS1* (*ENHANCED DISEASE SUSCEPTIBILITY1*), which are required for activation of SAR, encode proteins with sequence similarity to putative acyl hydrolases (Feys et al., 2001). However, the biochemical function of *PAD4* and *EDS1* is not known.

CHAPTER TWO

Characterization of the At5g14180 T-DNA insertion mutation effect on defense-related activity

INTRODUCTION

Plants have evolved sophisticated defense mechanisms to combat invading pathogens. Upon pathogen infection, the plant is able to recognize the threat and initiate the local defense response associated with the hypersensitive response (HR) at the site of infection, characterized by tissue collapse, oxidative burst and programmed cell death (Hammond-Kosak and Jones, 1996). As a consequence of this recognition and response, systemic defense mechanisms are activated (Uknes et al., 1992). This induced systemic resistance, commonly referred to as systemic acquired resistance (SAR) confers broad spectrum resistance throughout the plant (Hunt and Ryals, 1996). SAR activation is associated with an increase in levels of salicylic acid (SA) (Rasmussen et al., 1991) and is accompanied by the induction of *PATHOGENESIS-RELATED (PR)* genes, which encode PR proteins, some of which possess anti-microbial activities (Hunt and Ryals, 1996).

Although the SAR defense mechanism has been well characterized, the exact signal involved in SAR activation has not been identified. Furthermore, how this signal is perceived by leaves is poorly understood. This signal, generated in the pathogen-infected plant organs, is believed to move through the phloem to distal plant tissues, where it activates SAR activity (Durrant and Dong, 2004; Chaturvedi and Shah, 2006). Recently it has been implicated that lipids may be involved in SAR signaling and

activation (Durrant and Dong, 2004; Chaturvedi and Shah, 2006). For example, loss-of-function mutations in the *SFD1* (*SUPPRESSOR OF FATTY ACID DESATURASE DEFICIENCY1*) gene compromised SAR activation in response to inoculation with avirulent bacterial pathogen (Nandi et al., 2004). The *sfd1* mutant although capable of accumulating SA in the pathogen inoculated organ, does not accumulate SA in the distal organs of plants that have been locally inoculated with pathogen. However, *sfd1* is responsive to the SA analog BTH. These results suggest that *SFD1* is required for either the synthesis or translocation of a mobile signal from the site of pathogen infection (Nandi et al., 2004). The *SFD1* gene encodes a putative dihydroxyacetone phosphate reductase involved in glycerophospholipid metabolism (Nandi et al., 2004).

Similarly, a loss-of-function mutation in the *DIR1* (*DEFECTIVE IN INDUCED RESISTANCE1*) gene, results in a plant which is unable to express SAR, but is able to exhibit local resistance to *P. syringae* (Maldonado et al., 2002). Due to the fact that *DIR1* encodes a putative apoplastic lipid transfer protein and *dir1* mutant petiole exudates are not sufficient to elicit *PR1* expression, it is likely that *DIR1* promotes long distance signaling via interaction with a lipid derived molecule (Maldonado et al., 2002). Lipid binding was confirmed in structural studies with *DIR1* (Maldonado et al., 2002). In addition, two other genes, *PAD4* (*Phytoalexin-deficient4*) and *EDS1* (*enhanced disease susceptibility1*), which are required for activation of SAR encode proteins with sequence similarity to putative acyl hydrolases (Feys et al., 2001).

The *ssi2* (*suppressor of SA-insensitivity 2*) mutant, further establishes a role for lipids in plant defense. These mutants constitutively express PR genes, constitutively accumulate high levels of SA and exhibit heightened disease resistance to a variety of pathogens including *Pseudomonas syringae*, *Peronospora parasitica* and *Cucumber mosaic virus* (Kachroo et al., 2001, 2003, 2004; Nandi et al., 2003; Sekine et al., 2004; Shah et al., 1999, 2001). The *SSI2* gene encodes a plastid-localized stearoyl-acyl-carrier-protein desaturase, a member of the soluble fatty acid desaturase family, a family of enzymes that function as key regulators of fatty acid desaturation, which converts stearic acid (18:0) to oleic acid (18:1) (Kachroo et al., 2001). This suggests a role for lipids in the *ssi2*-conferred constitutive systemic defense activation phenotype. Lipases have previously been implicated as having a role in the plant defense response (Dhondt et al., 2002; de Jong et al., 2004; Zhang et al., 2003). This study focused on the At5g14180 gene, which encodes a protein with homology to acyl hydrolases/lipases that is expressed at elevated levels in the *ssi2* mutant plant. Here we show that T-DNA insertional mutations in the At5g14180 gene, which encodes a putative lipase and is expressed at elevated levels in the *ssi2* mutant plant, suppresses the *ssi2*-conferred heightened resistance in the *ssi2* At5g14180 double mutant plant. Furthermore, in comparison to the wild type plant, growth of the virulent pathogen *Pseudomonas syringae* pv. *maculicola* was enhanced in the mutant plants.

MATERIALS AND METHODS

Plant Growth Conditions

Arabidopsis plants were grown by sowing seeds in standard soil mix, placing two seeds in opposite corners of a 3-inch square pot. Pots were then covered with plastic and kept in a 4°C chamber for 72h. Plants were then transferred to a growth chamber programmed for 14 hours light and 10 hours dark with an average temperature of 22°C. Plants continued to be covered for one week with clear plastic in order to maintain high humidity. Plants were fertilized using Scotts fertilizer (The Scotts Company, Marysville, OH) according to manufacturer's instructions every two weeks.

Arabidopsis Mutants

The transgenic Arabidopsis lines containing a T-DNA insertion (SALK collection <http://signal.salk.edu>) in the At5g14180 gene used in this study were obtained from the Ohio State University Stock Center. The Salk lines, Salk_101919 and Salk_082589 are in the ecotype Columbia background. The *ssi2* mutants are in the Nössen background (Kachroo et al. 2001). Salk_101919 *ssi2* double mutants were obtained by pollinating flowers of an *ssi2* plant with pollen from the Salk_101919 mutant plant. F1 generation was screened for cross success by confirming homozygosity of the *SSI2* gene and At5g14180 gene using DNA extraction and PCR.

DNA Extraction for PCR analysis

DNA was extracted for PCR analysis in the previously described method (Konieczny and Ausubel, 1993). One medium-sized plant leaf of approximately 50mg was placed in a 1.5ml microfuge tube and frozen in liquid nitrogen for at least 10 minutes. The frozen samples were then ground with a plastic pestle in the microfuge tube. While still frozen, 200 μ l of extraction buffer (200mM Tris-HCl pH 7.5, 250mM NaCl, and 25mM EDTA pH 8.0, 0.5% SDS). To this extract 200 μ l of Tris-saturated Phenol:Chloroform (1:1) solution was added and vortexed. The mixture was then centrifuged at 13,000 rpm for 15 min. The upper aqueous layer was transferred to a fresh micro centrifuge tube. DNA was precipitated from the solution using an equal amount of isopropanol. The precipitated DNA was then washed with 70% ethanol and suspended in sterile distilled water.

DNA from the T-DNA insertion lines, salk_101919 and salk_082589 was collected using this method and used to screen for homozygous lines using PCR analysis. To assess the homozygosity of the mutant lines, two separate screens were used. For the Salk_101919 line, gene specific primers were used to check for the presence or absence of the wild type At5g14180 gene, 101919-F (5'-GGTAAATTAGATAATGGTTGCCCA-3') and 101919-R (5'-GGCTATATGCCTTAAAGCGGG-3') were used. To check for the presence of the T-DNA insertion, the 101919-R primer was used along with the T-DNA left border primer (5'-GCGTGGACCGCTTGCTGCAAC-3'). PCR was performed under the following

conditions for both sets of primers: 95°C for 5 min followed by 30 cycles of 95°C for 30 s, 65°C for 30 s and 72°C for 2 min with a final extension of 72°C for 5 min.

For the Salk_082589 line, gene specific primers were used to check for the presence or absence of the At5g14180 gene, 082589-F (5'-GGTAAATTAGATAATGGTTGCCCA-3') and 082589-R (5'-GGCTATATGCCTTAAAGCGGG-3') were used. To check for the presence of the T-DNA insertion, the 082589-F primer was used along with the T-DNA left border primer. PCR was performed under the following conditions for both sets of primers: 95°C for 5 min followed by 30 cycles of 95°C for 30 s, 65°C for 30 s and 72°C for 2 min with a final extension of 72°C for 5 min. The resulting amplified fragments were resolved on a 1.0% agarose gel, stained with ethidium bromide and visualized using the Gel Doc UVP BioDoc-ItTM system.

DNA from the Salk_101919 *ssi2* double mutant was collected using this method and used to screen the segregating F2 population to identify plants homozygous for both the T-DNA insertion knockout line and the *ssi2* mutant allele. The homozygosity of the mutant line was assessed using the primers and PCR conditions previously described. To differentiate between the *ssi2* mutant and the wild type *SSI2* alleles, a derived-cleaved amplified polymorphic sequence (dCAPS) was used. Primers *ssi2dCAPS*-F (5'-TTGTTTGTTGGGGACATGATCACACAGAAGGTGCA-3') and *ssi2dCAPS*-R (5'-TCGATCTGCCTCATGTCAACAGG-3') were used in the PCR reaction under the following conditions: 95°C 5 min followed by 35 cycles of 95°C for 45 s, 65°C for 45 s

and 72°C for 45 s with a final extension of 72°C for 5 min. The resultant amplicon was 200bp. Wild-type DNA contains an Apal1 site (New England Biolabs, MA). Upon digestion with the Apal1 restriction enzyme, wild type amplicons yield two fragments of 175bp and 25bp, while *ssi2* mutant amplicons yield one 200bp fragment. The resulting restriction digested amplification product was resolved on a 2.5% agarose gel, stained with ethidium bromide and visualized using the Gel Doc UVP BioDoc-It™ system.

RNA extraction and RT-PCR analysis

Leaf material was quick frozen in liquid nitrogen and RNA was extracted in the method previously described (Chomczynski and Sacchi, 1987). While still frozen, plant tissue is ground with a plastic pestle and suspended in 1ml of guanidine-phenol-acetate solution. Sample is then allowed to sit at room temperature for 10min. 200µl of CHISM (24:1 mixture of chloroform: isomyl alcohol) was then added and samples were then vortexed vigorously for 10 – 20s, kept at room temperature for 5min and centrifuged at 13,000 rpm for 15min. From the supernatant, RNA is precipitated using 500µl isopropanol and centrifuged for 10min at 15,000 rpm. Isolated RNA was then purified using the RNeasy Mini kit (Stratagene, CA). RNA was quantified using a spectrophotometer at 260nm.

RT-PCR reactions were carried out using a two-step procedure. The first step, generation of cDNA was performed using oligo-dT primer (Promega) and MMLV Reverse Transcriptase (Promega) along with RNA, heated at 37°C for 1 hour.

The second step uses the cDNA produced in step one in a PCR reaction using Actin-F (5'-ATGAAGATTAAGGTCGTGGCA-3') and Actin-R (5'-TCCGAGTTGAAGAGGGCTAC-3'), PR1-F (5'-ATGAATTTACTGGCTATTC-3') and PR1-R (5'-ATGAATTTACTGGCTATTC-3'), At5g14180-F (5'-GGCCATGGATATGGTCAAAC-3') and At5g14180-R (5'-ATCCAGCGGATCAAAATCTG-3') gene specific primers used for amplification of Actin (control), PR-1 and At5g14180. PCR was performed under the following conditions: 95°C 5min followed by 25 cycles of 94°C for 35 sec, 50°C for 35 sec and 72°C for 90 sec with a final extension of 72°C for 10 min. The resulting amplified fragments were resolved on a 1.0% agarose gel, stained with ethidium bromide and visualized using the Gel Doc UVP BioDoc-It™ system.

Bacterial growth conditions

P.syringae pv tomato DC3000 containing *avrRpt2* was grown overnight at 28°C in King's B medium (King et al, 1954) containing 50mg/mL kanamycin and 25mg/mL rifampicin. *P.s. pv. maculicola* ES4326 was grown overnight at 28°C in King's B medium containing 100μg/mL streptomycin.

Bacterial infection of plants

For basal pathogen growth experiments, *P. syringae* pv. *maculicola* ES4326 was suspended in 10mM MgCl₂ (OD₆₀₀ = 0.0001). This suspension was then infiltrated in the abaxial surface of four-week-old plant leaves using a 1ml plastic syringe without the needle. Plants were then covered with clear plastic for 12 h. After 72 h, leaf discs of the

same size (0.283 cm^2) were taken from four Psm-infected leaves and ground with a plastic pestle in 1mL of 10mM MgCl₂. From these suspensions, serial dilutions were made in 1:100, 1:1000 and 1:10000 concentrations. From each dilution, 10µl was plated on 1/3 area of a King's B medium agar plate containing 100µg/ml streptomycin. Plates were allowed to incubate for 48 h at 28°C and bacterial colony growth numbers were determined each sample.

For systemic acquired resistance experiments, four leaves from each plant were inoculated with either 10mM MgCl₂ or the avirulent bacterial pathogen *P. syringae* pv tomato DC3000 suspended in 10 mM MgCl₂ ($\text{OD}_{600} = 0.01$) infiltrated into the abaxial surface of four-week old plants using a 1ml plastic syringe without the needle. After 72h, distal leaves of these same plants (4-6 leaves per plant) were then inoculated with the virulent pathogen *P. syringae* pv. *maculicola* ES4326 suspended in 10mM MgCl₂ ($\text{OD}_{600} = 0.00025$) infiltrated into the abaxial surface using a 1ml plastic syringe without the needle. Plants were then covered with clear plastic for 12 h. After 72 h, leaf discs of the same size were taken from four *P. syringae* pv. *maculicola*-infected leaves and ground with a plastic pestle in 1ml of 10mM MgCl₂. From these suspensions, serial dilutions were made in 1:100, 1:1000 and 1:10000 concentrations. From each dilution, 10µl was plated on 1/3 area of a King's B medium agar plate containing 100µg/ml streptomycin. Plates were allowed to incubate for 48 h at 28°C and bacterial colony numbers determined.

For salicylic acid (SA) treatment followed by bacterial infection, four-week old plants were sprayed and subirrigated with a 0.05 mM Salicylic acid solution and covered with clear plastic dome for 12 h, after which the plastic dome was removed. Plants similarly treated with water provided the controls. 24 h subsequent to application of SA, 6-8 leaves from each treated plant were infiltrated with *P. syringae* pv. *maculicola* ES4326 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.0001). The bacterial suspension was infiltrated into the abaxial surface of plant leaves using a 1ml plastic needleless syringe. Plants were then covered with clear plastic dome for 12h to maintain high humidity and facilitate the infection process, after which the dome was removed. 72 h later, leaf discs of the same size (0.283 cm²) were taken from four *P. syringae* pv. *maculicola*-infected leaves and ground with a plastic pestle in 1mL of 10mM MgCl₂. From these suspensions, serial dilutions were made in 1:100, 1:1000 and 1:10000 concentrations. From each dilution, 10µl was plated on 1/ area of a King's B medium agar plate containing 100µg/ml streptomycin. Plates were allowed to incubate for 48 hours at 28°C and bacterial colony growth was counted for each sample. For each treatment, three such samples containing four leaf discs each were used.

RESULTS

Genes encoding lipases of poorly defined metabolic function are expressed at elevated levels in the *ssi2* mutant. The *ssi2* mutant plant constitutively exhibits a SAR-like defense mechanism, conferring enhanced resistance to a wide variety of pathogens (Kachroo et al., 2001, 2003, 2004; Nandi et al., 2003; Sekine et al., 2004; Shah et al., 1999, 2001). The *SSI2* gene encodes a stearoyl-acyl-carrier-protein desaturase, which converts steric acid (18:0) to oleic acid (18:1) (Kachroo et al., 2001), thus implicating a role for lipids in plant defense. Microarray analysis of the *ssi2* mutant plant revealed 12 genes predicted to encode putative lipases/esterases, expressed at high levels in the *ssi2* mutant as compared to the wild type plant (Fig. 2-1). The At5g14180 gene expression is 60-fold higher in the *ssi2* mutant plant than in the wild type plant (Fig 2 – 1).

At5g14180 protein sequence. The presence of the SER ASP HIS catalytic triad in the protein sequence of the At5g14180 designates the function of At5g14180 as a putative lipase (Fig. 2 – 2). Additionally, the At5g14180 protein sequence contains a signal peptide localizing it to the vacuole.

PCR based strategy for identification of plants with T-DNA insertion in At5g14180 gene. At5g14180 gene specific primers, represented here by L and R, were designed such that a short PCR amplification time would yield a gene specific PCR product only in the presence of the wild type At5g14180 allele. To assess the presence of the T-DNA insertion in the Salk_101919 and Salk_082589 mutant lines, a gene specific primer was

used along with primer specific to the left border of the T-DNA insertion. In the Salk_082589 line, the T-DNA insertion is oriented such that amplification using the 082589-F primer along with the T-DNA left border primer will amplify a product validating the presence of the T-DNA insertion (Fig 2 – 3). The Salk_101919 T-DNA insertion is orientated such that amplification with the 101919-R primer and the T-DNA left border primer will amplify a product validating the presence of the T-DNA insertion in Salk_101919 mutant lines (Fig. 2 – 3).

PCR products confirming homozygosity of At5g14180 T-DNA insertion mutant lines. To assess the function of the At5g14180 gene in the *ssi2*-conferred phenotype and in plant defense, two transgenic SALK lines (Salk_101919 and Salk_082589) that contain T-DNA insertions within the At5g14180 gene (<http://signal.salk.edu>), were obtained. Salk_101919 and Salk_082589 were obtained from the Ohio State University Stock Center. To confirm that these Salk lines were homozygous for the T-DNA insertion, therefore knocking-out the At5g14180 gene, DNA extraction and PCR analysis was performed on the Salk lines. To confirm the absence of the At5g14180 gene in the T-DNA insertion mutants, gene specific primers, Salk_101919-F and Salk_101919-R; Salk_082589-F and Salk_082589-R, specific to each Salk line, were used to amplify DNA from wild type, Salk_101919 and Salk_082589 plants. In gel lanes 1 and 5, the wild type gene-specific PCR product is shown, with an absence of product for Salk_101919 and Salk_082589 in gel lanes 2 and 6 (fig 2 – 4). To confirm the presence of the T-DNA insertion in each of the Salk lines, the gene specific Salk_101919-R and Salk_082589-R primers were used along with primer specific to the left-border of the T-

DNA insertion. In gel lanes 4 and 8, the T-DNA insertion PCR product is shown for the Salk_101919 and Salk_082589 lines, with an absence of the T-DNA insertion product for wild type, gel lanes 3 and 7 (Fig 2 – 4). These products, taken together, confirm the homozygosity of the Salk_101919 and Salk_082589 mutant lines.

Systemic Acquired Resistance is not impaired in At5g14180 T-DNA insertion mutants. To test the involvement of the At5g14180 gene in the SAR defense response, Wild type and the Salk_101919 and Salk_082589 homozygous transgenic plants were infiltrated with either 10mM MgCl₂ (mock) or *P. syringae* pv *tomato* DC 3000 *AvrRpt2* avirulent pathogen (SAR). After 72 h, the distal leaves of these same plants were inoculated with the virulent pathogen *P. syringae* pv. *maculicola* ES326. Bacterial colony growth was assessed for mock and SAR treated plants and expressed as colony forming units (CFU) per cm² of leaf area (Fig 2 – 5). SAR activity was assessed by comparing bacterial growth in SAR treated plants to bacterial growth in mock treated plants. In the wild type and the Salk_101919 and Salk_082589 plants, bacterial growth was reduced on plants primed with *P. syringae* pv *tomato* DC3000 *AvrRpt2* as compared to plants primed with 10mM MgCl₂, indicative of SAR defense activity induction (Fig 2 – 5). This gives evidence that the Salk_101919 and Salk_082589 mutant plants are SAR competent. Therefore, mutations in the At5g14180 gene do not impair SAR related defense activity.

Growth of *P. syringae* pv. *maculicola* in At5g14180 T-DNA insertion mutants.

In the above experiments to study SAR, the mock-treated Salk_101919 and Salk-082589 mutant plants appeared to be more susceptible to *P. syringae* pv. *maculicola* ES4326, as shown by the higher level of bacterial growth in mock treated wild type plants (Fig. 2 – 6). To determine if the mutants are indeed more susceptible to the virulent pathogen, the wild type and the Salk_101919 and Salk_082589 plants were inoculated with the virulent pathogen *P. syringae* pv. *maculicola* ES4326. The basal resistance was assessed by counting the bacterial colony growth and expressing it as colony forming units (CFU) per cm² of leaf area (Fig 2 – 6). Comparison of bacterial growth on the Salk_101919 and Salk_0282589 mutant plants with the bacterial growth on the wild type plant indeed gave evidence to support slight enhanced susceptibility of the Salk_101919 and Salk_082589 mutant plants to the virulent pathogen *P. syringae* pv. *maculicola* ES4326 (Fig 2 – 6). This approximately 2-3 fold difference in growth between wild type and mutant plants was consistently observed over four independent experiments.

At5g14180 expression is induced upon *P. syringae* pv. *maculicola* infection. The At5g14180 gene was expressed in wild type plants 24h and 48h after infection with the virulent pathogen *P. syringae* pv. *maculicola* ES4326, but expression was not induced in the Salk_101919 and Salk_082589 mutant plants after *P. syringae* pv. *maculicola* infection (Fig. 2 – 7), confirming knock-out of At5g14180 expression. Additionally, *PR1* expression was induced in the wild type and in the Salk_101919 and Salk_082589 mutant plants 24h and 48h after infection with *P. syringae* pv. *maculicola* (Fig. 2 – 7).

During pathogenesis, response to SA is not impaired in the At5g14180 T-DNA insertion mutants. An increase in plant SA levels is associated with SAR defense related activity and SA is constitutively expressed in the *ssi2* mutant. Additionally, treatment with SA can activate the systemic defense response and decrease plant susceptibility to pathogen. Therefore, SA response was assessed in the Salk_101919 and Salk_082589 mutant plants. Wild type, Salk_101919 and Salk_082589 plants were sprayed and subirrigated with a 0.05 mM salicylic acid solution (SA) or as a control, with water (mock). After 24 h, these SA- or mock-treated plants were then inoculated with the virulent pathogen *P. syringae* pv. *maculicola* ES4326. Bacterial colony growth was assessed for SA and mock treated plants and expressed as colony forming units (CFU) per cm² of leaf area (Fig 2 – 8). Response to SA was assessed by comparing bacterial growth of SA-treated plants to that of mock-treated plants. In the wild type, Salk_101919 and Salk_082589 plants, bacterial growth was reduced on SA-treated plants, as compared to mock-treated plants, indicative of SA systemic defense activity induction (Fig 2 – 8). Furthermore, as expected, mock-treated Salk_101919 and Salk_082589 plants again exhibited slight enhanced susceptibility to the virulent pathogen *P. syringae* pv *maculicola* ES4326, as compared to growth on the mock-treated wild type plant (Fig 2 – 8). Thus, SA treatment was able to induce enhanced defense response in Salk_101919 and Salk_082589 mutant plants, as compared to mock-treated Salk_101919 and Salk_082589 mutant plants, therefore, the response to SA is not impaired in the At4g14180 T-DNA insertion mutants.

At5g14180 expression is induced upon SA treatment. RNA extracted from wild type and the Salk_101919 and Salk_082589 mutant plants after treatment with water (mock) or a 0.05 mM salicylic acid solution. At5g14180 gene expression was induced in wild type plants 12h and 24h after treatment with SA, while mock-treated wild type plants did not express the At5g14180 gene (Fig. 2 – 9). At5g14180 gene expression was not induced in the Salk_101919 and Salk_082589 mutant plants under any treatment conditions (Fig. 2 – 9). PR1 gene expression was observed in the wild type and in the Salk_101919 and Salk_082589 mutant plants 24h and 48h after SA treatment (Fig. 2 – 9).

PCR products confirming homozygosity of Salk_101919 ssi2 double mutants.

To evaluate the effect of the T-DNA insertion mutations in the At5g14180 gene on the *ssi2* mutant phenotype and constitutive enhanced defense mechanism, Salk_101919 *ssi2* double mutants were created by pollinating flowers of an *ssi2* plant with pollen from the Salk_101919 mutant plant. The resulting double mutant displayed a dwarf phenotype as compared to the wild type plant and Salk_101919 mutant plant, but as compared to the *ssi2* mutant plant, were slightly larger (Fig 2 – 10(A)).

Salk_101919 *ssi2* double mutants were confirmed in the F2 generation by DNA extraction and PCR analysis. To confirm the absence of the At5g14180 gene in the Salk_101919 *ssi2* double mutant, gene specific primers were used to amplify DNA from wild type and the Salk_101919 and Salk_082589 mutant plants. In Figure 2 – 10(B), gel lane 1, the wild type product of amplification by the gene specific primers 101919-F and 101919-R is present, while in gel lane 2 and lane 3 the Salk_101919 and Salk_101919

ssi2 product of amplification with gene specific primers is absent. To confirm the presence of the T-DNA insertion in the Salk_101919 *ssi2* double mutant, the Salk_101919-R gene specific primer and the T-DNA left border primer were used to amplify DNA from wild type and Salk_101919 *ssi2* mutant plants. In figure 2 – 10(B) gel lane 4, the wild type product of amplification with the 101919-F and T-DNA left border primer is absent, while in lanes 5 and 6, the amplification product is present in the DNA from Salk_101919 and Salk_101919 *ssi2* mutant plants amplified with 101919-F gene specific and T-DNA left border primers. Taken together, the presence of the T-DNA insertion and absence of the At5g14180 gene specific band indicate the Salk_101919 *ssi2* double mutant is homozygous for the T-DNA insertion. To differentiate between *ssi2* and the wild type *SSI2* allele a derived-cleaved amplified polymorphic sequence (dCAPS) was used. Primers ssi2dCAPS-F and ssi2dCAPS-R were used to amplify DNA from wild type, *ssi2* and Salk_101919 *ssi2* double mutant plants. The resulting amplified product was then digested with the *Apal1* restriction enzyme, which restricts the *SSI2* amplicon, but not the *ssi2* amplicon. In Figure 2 – 10(B), gel lane 7, the restriction digestion of wild type PCR product with *Apal1* yielded a smaller 175 bp product, while amplification and digestion of PCR product from the *ssi2* single mutant and the Salk_101919 *ssi2* double mutant yielded a larger uncut product of 200bp, lanes 8 and 9. The presence of the uncut amplified product indicates the Salk_101919 *ssi2* double mutant is homozygous for the *ssi2* mutation.

Growth of *P. syringae* pv. *maculicola* in Salk_101919 *ssi2* double mutant. To evaluate the effect of the T-DNA insertion mutation on *ssi2*-conferred enhanced defense

phenotype, Salk_101919 *ssi2* double mutants, along with wild type (Col-0 and 1/8E) and the Salk_101919 and *ssi2* single mutant plants, were inoculated with the virulent pathogen *P. syringae* pv. *maculicola* ES4326. The basal resistance was assessed by counting the bacterial colony growth and expressing it as colony forming units (CFU) per cm² of leaf area (Fig. 2 – 11). Bacterial growth in the Salk_101919 *ssi2* double mutant was found to be greater than in the *ssi2* mutant plant, but lower than wild type and Salk_101919 single mutant (Fig 2 – 11).

At5g14180 is constitutively expressed in the *ssi2* mutant. RNA was extracted from *P. syringae* pv. *maculicola* ES4326 treated wild type (Col-0 and the 1/8E line in accession Nössen) plants and Salk_101919, Salk_101919 *ssi2* double mutant and *ssi2* single mutant plants 0h, 24h and 48h post-inoculation. As expected, At5g14180 gene was constitutively expressed in the *ssi2* mutant (Fig. 2 – 12). Expression of At5g14180 expression was higher in Col-0, 1/8E wild type plants and in the *ssi2* single mutant plant 24h and 48h after infection with *P. syringae* pv. *maculicola* (Fig 2 – 12). At5g14180 expression was not observed in Salk_101919 and Salk_101919 *ssi2* double mutant regardless of treatment. *PR1* expression was observed to be constitutively expressed in the *ssi2* mutant plant, as well as in the Salk_101919 *ssi2* double mutant plant at comparable levels (Fig. 2 – 12). Furthermore, *PR1* gene expression was induced in response to pathogen inoculation in the Salk_101919 mutant similar to levels observed in the pathogen-challenged wild type plants (Fig. 2 – 12). These results suggest that the At5g14180 gene does not have a discernable impact on the pathway leading to *PR1* gene expression.

DISCUSSION

The *ssi2* mutant has been previously shown to constitutively express *PR* genes, accumulate elevated levels of SA and exhibit heightened resistance to a variety of pathogens including *Pseudomonas syringae* pv *maculicola*, *Hyaloperonospora parasitica* and *Cucumber mosaic virus* (Kachroo et al., 2001, 2003, 2004; Nandi et al., 2003; Sekine et al., 2004; Shah et al., 1999, 2001). Additionally, these mutants display a dwarf phenotype and spontaneously exhibit lesions and cell death (Shah et al., 1999, 2001). The *SSI2* gene encodes a plastid-localized stearoyl-acyl-carrier-protein desaturase, which converts steric acid (18:0) to oleic acid (18:1) (Kachroo et al., 2001). This stearoyl-ACP-desaturase is a member of the soluble fatty acid desaturase family, a family of enzymes that function as key regulators of fatty acid desaturation (Kachroo et al., 2001). Previous studies involving two genes, *PAD4* (*Phytoalexin-deficient4*) and *EDS1* (*enhanced disease susceptibility1*), which have been shown to play a role in SAR and control SA accumulation in response to pathogen, exhibit homology to lipases/acyl hydrolases (Falk et al., 1999; Feys et al., 2001; Jirage et al., 1999). Further, loss-of-function mutations in the *pad4* and *eds1* genes suppress *ssi2*-conferred enhanced resistance (Kachroo et al., 2005; Nandi et al., 2005). These observations implicate a role for lipids in the growth and defense phenotype of the *ssi2* mutant phenotypes. Previous studies with the suppressor of fatty acid desaturase deficiency mutants, which encode genes involved in glycerolipid biosynthesis in the plastid indicated a role for plastid lipid metabolism in the growth and defense phenotypes of the *ssi2* mutant (Nandi et al., 2003, 2004).

In order to further evaluate the role of lipids in the *ssi2*-conferred phenotypes, microarray analysis was used to identify genes involved in lipid metabolism that are expressed at elevated levels in the *ssi2* plant. Microarray analysis revealed 12 genes that encode putative lipases/acyl hydrolases, which are expressed at elevated levels in the *ssi2* mutant plant, as compared to the wild type plant (Fig 2 – 1). One gene in particular, At5g14180, was found to be induced 60-fold in the *ssi2* mutant, as compared to wild type (Fig 2 – 1). The At5g14180 protein sequence contains the SER ASP HIS catalytic triad (Fig. 2 – 2), characteristic of required for lipase catalytic function, implicating its function as being a putative lipase. Previous studies had identified two other genes, *PAD4* (*PHYTOALEXIN-DEFICIENT4*) and *EDS1* (*ENHANCED DISEASE SUSCEPTIBILITY1*), that exhibit sequence similarities to lipases and are involved in *ssi2*-conferred phenotypes and in plant defense. Loss-of-function mutations *pad4* and *eds1* mutant alleles attenuated the *ssi2*-conferred *PR1* expression and enhanced disease resistance phenotype (Kachroo et al., 2005; Nandi et al., 2005). In addition, SAR and SA accumulation in response to pathogen infection were also compromised by the *pad4* and *eds1* mutant plants (Falk et al., 1999; Feys et al., 2001; Jirage et al., 1999).

To assess the role and involvement of At5g14180 in plant defense and in *ssi2*-mediated phenotypes, T-DNA insertion mutants were obtained, Salk_101919 and Salk_082589. In this chapter, we show that the presence of the Salk_101919 allele compromised *ssi2*-conferred enhanced resistance to the virulent pathogen *P. syringae* pv. *maculicola* ES4326, as shown by greater bacterial growth seen in the Salk_101919 *ssi2* double mutant as compared to the *ssi2* mutant plant (Fig. 2 – 11). However, the

Salk_101919 *ssi2* double mutant maintained the dwarf stature associated with the *ssi2* allele, albeit the double mutant being slightly larger in size than the *ssi2* mutant (Fig. 2 – 10A). This is similar to the dwarf phenotype of the *ssi2 pad4*, *ssi2 nahG* and *ssi2 eds5* plants, all of which are compromised in *ssi2*-conferred enhanced resistance (shah et al., 2001; Nandi et al., 2005). The *PAD4*, *EDS5* and *nahG* transgene impact SA signaling/synthesis (Durrant and Dong, 2004; Chaturvedi and Shah, 2006). However, unlike the *PAD4*, *EDS5* and *nahG* transgenes, *ssi2*-contributed *PR1* expression was not attenuated in the Salk_101919 *ssi2* double mutant plant, suggesting that the At5g14180 gene affects a different mechanism that is hyperactive in the *ssi2* mutant.

SAR was not compromised in the Salk_101919 and Salk_082589 T-DNA insertion mutant lines (Fig. 2 -5, 2 – 8), which were also responsive to exogenously applied SA. However, basal resistance to pathogen was compromised in the Salk_101919 and Salk_082589 mutant plants (Fig 2 – 6). The weak effect of these mutant alleles on basal resistance in the single mutant plants as opposed to the strong effect of the Salk_101919 insertion on the *ssi2*-conferred heightened resistance phenotype could be due to the fact that the At5g14180 gene may be involved in the NPR1-independent mechanism, which is also hyperactivated in the *ssi2* mutant. This would also explain why SA application enhanced resistance in the Salk_101919 and Salk_082589 single mutant plants to levels comparable to that in wild type plant, in which the NPR1 dependent pathway has a larger contribution than the NPR1-independent pathway in SA signaling. Moreover, this would also explain why SAR was not compromised in the Salk_101919 and Salk_082589 single mutant plant as the

manifestation of SAR is dependent on the NPR1 dependent pathway. Future studies of At5g14180 *npr1* double mutant plant and plants ectopically expressing At5g14180 will aid in testing the involvement of At5g14180 in an NPR1-independent mechanism.

Alternatively, the absence of a strong defense phenotype in the Salk_101919 and Salk_082589 mutant plants could be due to genetic redundancy due to presence of other lipases that may substitute in the absence of a functional At5g14180 allele in the Salk_101919 and Salk_08289 single mutant plants. However, in the *ssi2* mutant, since basal expression of the At5g14180 gene is very high (Fig. 2 – 12), this high level expression of At5g14180 contributes to the *ssi2*-conferred resistance to *P. syringae pv maculicola* ES4326.

The *ssi2* mutant also exhibits heightened resistance to the phloem feeding insect, green peach aphid (*Myzus persicae* Sölzer) (Pegadaraju et al., 2005). The At5g14180 gene is required for this *ssi2*-conferred defense phenotype as well (Vijay Singh, Joe Louis and J. Shah, personal comm.). Furthermore, in comparison to the wild type plant, green peach aphid population was larger on the Salk_101919 and Salk_082589 mutant plants (Vijay Singh, Joe Louis and J. Shah, personal comm.), providing further evidence for an important role for this putative lipase gene in plant defense against biotic stress.

FIGURE LEGENDS

Fig. 2 – 1. Genes encoding lipases of poorly defined metabolic function are expressed at elevated levels in the *ssi2* mutant. Microarray analysis of the *ssi2* mutant plant revealed several genes encoding putative lipases are expressed at elevated levels as compared to the wild-type plant. In particular, the At5g14180 Arabidopsis gene displayed a fold induction 60-times higher in the *ssi2* mutant plant, as compared to the wild-type plant.

Fig. 2 – 2. At5g14180 protein sequence. The presence of the SER ASP HIS catalytic triad in the protein sequence of At5g14180 designates the function of At5g14180 as a putative lipase. Additionally, the At4g14180 protein sequence contains a signal peptide localizing it to the vacuole.

Fig. 2 – 3. PCR based strategy for identification of plants with T-DNA insertion in At5g14180 gene. At5g14180 gene specific primers, represented here by L and R, were designed such that a short PCR amplification time would yield a gene specific PCR product only in the presence of the wild type At5g14180 allele. To assess the presence of the T-DNA insertion in the Salk_101919 and Salk_082589 mutant lines, a gene specific primer was used along with primer specific to the left border of the T-DNA insertion. In the Salk_082589 line, the T-DNA insertion is oriented such that amplification using the 082589-F primer along with the T-DNA left border primer will amplify a product validating the presence of the T-DNA insertion. The Salk_101919 T-DNA insertion is

orientated such that amplification with the 101919-R primer and the T-DNA left border primer will amplify a product validating the presence of the T-DNA insertion in Salk_101919 mutant lines.

Fig. 2 –4. PCR products confirming homozygosity of At5g14180 T-DNA insertion mutant lines. DNA from Salk_101919 and Salk_082589 mutant lines was extracted and PCR was performed to assess the presence of a T-DNA insertion and absence of the At5g14180 gene. Gel lanes 1 and 2 represent WT and Salk_101919 DNA amplification with the gene specific primers, 101919-F (5'-GGTAAATTAGATAATGGTTGCCCA-3') and 101919-R (5'-GGCTATATGCCTTAAAGCGGG-3'). Gel lanes 3 and 4 represent WT and Salk_101919 DNA amplification with the 101919-R primer and the T-DNA left border primer (5'-GCGTGGACCGCTTGCTGCAAC-3'). Gel lanes 5 and 6 represent WT and Salk_082589 DNA amplification with the gene specific primers 082589-F (5'-GGTAAATTAGATAATGGTTGCCCA-3') and 082589-R (5'-GGCTATATGCCTTAAAGCGGG-3'). Gel lanes 7 and 8 represent WT and Salk_082589 DNA amplification with the 082589-F primer and the T-DNA left border primer.

Fig. 2 – 5. Systemic Acquired Resistance is not impaired in At5g14180 T-DNA insertion mutant lines. Four leaves from each plant of wild-type (col-0), salk_101919 and salk_082589 were inoculated with 10 mM MgCl₂ (mock) or P. syringae pv tomato DC3000 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.01) by infiltration into the abaxial surface using a 1-ml plastic syringe. After 72 hours, distal leaves of the same plants (4-6

leaves per plant) were then inoculated with *P. syringae* strain *P.s. maculicola* ES4326 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.00025) by infiltration into the abaxial surface using a 1-ml plastic syringe. After 72 hours, leaf discs of the same size (0.0283 cm²) were taken from four *P.s. maculicola*-infected leaves and ground in MgCl₂. Serial dilutions were made and plated on King's with streptomycin. Bacterial numbers were titered. The bacterial numbers are represented as the number of colony forming units per cm².

Fig. 2 – 6. Growth of *P. syringae* pv. *maculicola* in At5g14180 T-DNA insertion mutants. *P. syringae* pv. *maculicola* ES4326 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.0001) was infiltrated into the abaxial surface of leaves from wild-type (col-o), salk_101919 and salk_082589 plants using a 1-ml plastic syringe without the needle. After 72 hours, leaf discs of the same size (0.0283 cm²) were taken from four Psm-infected leaves and ground in MgCl₂. Serial dilutions were made and plated on King's with streptomycin. Bacterial numbers were titered. The bacterial numbers are represented as the number of colony forming units per cm². Bacterial numbers represent the mean of 5 samples ±SE. Asterisks above the bars indicate values are different from the WT control with a confidence of 95% with the Student's *t*-test.

Fig. 2 – 7. At5g14180 expression is induced upon *P. syringae* pv. *maculicola* infection. RNA was extracted from the above experiment, from wild type and the Salk_101919 and Salk_082589 mutant plants. Expression of the wild type At5g14180 gene was assessed in untreated plants (0hr) and plants treated with *P. syringae* pv.

maculicola ES4326, 24h and 48h post inoculation. RNA was used to create cDNA, which was amplified using PCR by Actin primers, Actin-F (5'-ATGAAGATTAAGGTCGTGGCA-3') and Actin-R (5'-TCCGAGTTGAAGAGGCTAC-3') as a control for RNA quality and At5g14180-F (5'-GGCCATGGATATGGTCAAAC-3') and At5g14180-R (5'-ATCCAGCGGATCAAAATCTG-3') gene specific primers, used to identify the expression of the At5g14180 wild type gene. PR1 gene expression was also evaluated, using the PR1-F (5'-ATGAATTACTGGCTATT-3') and PR1-R (5'-ATGAATTACTGGCTATT-3') primers to amplify expression of the PR1 gene.

Fig. 2 – 8. Response to SA is not impaired in the At5g14180 T-DNA insertion mutants. Wild-type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with a 0.05 mM salicylic acid solution. After 24 hours, 6-8 leaves from each treated plant were inoculated with *P. syringae* strain *P.s. maculicola* ES4326 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.0001), infiltrated into the abaxial surface with a 1-ml plastic syringe without the needle. After 72 hours, leaves of the same size were taken from four psm-infected leaves and ground in MgCl₂. Serial dilutions were made and plated on King's with streptomycin. Bacterial numbers were titered. The bacterial numbers are represented as the munber of colony forming units per cm². Bacterial numbers represent the mean of 15 plants ±SE. Results were concluded with data obtained from 3 independent experiments. Asterisks above the bars indicate values are different from the mock control for each sample with a confidence of 95% with the Student's *t*-test.

Fig. 2 – 9. At5g14180 expression is induced upon SA treatment. RNA was extracted from the above experiment, from wild type and the Salk_101919 and Salk_082589 mutant plants. Expression of the wild type At5g14180 gene was assessed in untreated plants (0hr) and plants treated with either water (mock) or a 0.05 mM salicylic acid solution (SA), at 12 h and 24h timepoints. RNA was used to create cDNA, which was amplified using PCR by Actin primers, Actin-F (5'-ATGAAGATTAAGGTCGTGGCA-3') and Actin-R (5'-TCCGAGTTGAAGAGGCTAC-3') as a control for RNA quality and At5g14180-F (5'-GCCATGGATATGGTCAAAC-3') and At5g14180-R (5'-ATCCAGCGGATCAAAATCTG-3') gene specific primers, used to identify the expression of the At5g14180 wild type gene. PR1 gene expression was also evaluated, using the PR1-F (5'-ATGAATTACTGGCTATT-3') and PR1-R (5'-ATGAATTACTGGCTATT-3') primers to amplify expression of the PR1 gene.

Fig. 2 – 10. PCR products confirming homozygosity of Salk_101919 ssi2 double mutant. (A) Photograph of Salk_101919, WT, Salk_101919 ssi2 and *ssi2* plants. (B) DNA from salk_101919 ssi2 double mutant plants was extracted and PCR was performed to assess the absence of the At5g14180 gene and the SSI2 gene. Gel lanes 1, 2 and 3 represent WT, Salk_101919, and Salk_101919 ssi2 DNA amplification with the gene specific primers, 101919-F (5'-GGTAAATTAGATAATGGTTGCCCA-3') and 101919-R (5'-GGCTATATGCCTTAAAGCGGG-3'). Gel lanes 4, 5 and 6 represent WT, Salk_101919 and Salk_101919 ssi2 DNA amplification with the 101919-R primer and the T-DNA left border primer (5'-GCGTGGACCGCTTGCTGCAAC-3'). Gel lanes

7, 8 and 9 represent WT, *ssi2* and Salk_101919 *ssi2* DNA amplification with *ssi2dCAPS-F* (5'-TTGTTTGGTGGGGACATGATCACACAGAAGGTGCA-3') and *ssi2dCAPS-R* (5'-TCGATCTGCCTCATGTCAACAGG-3') and subsequent digestion with Apal1 restriction enzyme (New England Biolabs, MA).

Fig. 2 – 11. Growth of *P. syringae* pv. *maculicola* in Salk_101919 *ssi2* double mutant.

P. syringae pv. *maculicola* ES4326 suspended in 10mM MgCl₂ (OD₆₀₀ = 0.0001) was infiltrated into the abaxial surface of wild-type (Col-0 and 1/8E), salk_101919, *ssi2* and 101919 *ssi2* plants using a 1-ml plastic syringe without the needle. After 72 hours, leaf discs of the same size (0.0283 cm²) were taken from four *Psm*-infected leaves and ground in MgCl₂. Serial dilutions were made and plated on King's with streptomycin. Bacterial numbers were titered. The bacterial numbers are represented as the number of colony forming units per cm². Bacterial numbers represent the mean of 15 plants ±SE. Results were concluded with data obtained from 3 independent experiments. Asterisks above the bars indicate values are different from the WT and *ssi2* control with a confidence of 95% with the Student's *t*-test.

Fig. 2 – 12. At5g14180 is constitutively expressed in the *ssi2* mutant. RNA was extracted from wild type (col-0 and 1/8E) and the Salk_101919 *ssi2* double mutant and *ssi2* single mutant plants, after treatment with *P. syringae* pv. *maculicola*. Expression of the wild type At5g14180 gene was assessed in samples taken at 0h, 24h and 48h time points. RNA was used to create cDNA, which was amplified using PCR by Actin primers, Actin-F (5'-ATGAAGATTAAGGTCGTGGCA-3') and Actin-R (5'-

TCCGAGTTGAAGAGGCTAC-3') as a control for RNA quality and At5g14180-F (5'-GGCCATGGATATGGTCAAAC-3') and At5g14180-R (5'-ATCCAGCGGATCAAAATCTG-3') gene specific primers, used to identify the expression of the At5g14180 wild type gene. *PR1* gene expression was also evaluated, using the PR1-F (5'-ATGAATTACTGGCTATT-3') and PR1-R (5'-ATGAATTACTGGCTATT-3') primers to amplify expression of the *PR1* gene.

Fig. 2 - 1

Fold Induction

Locus	<i>ssi2</i>	Homology
At2g39400	3.221	Putative Phospholipase
At5g18630	2.867	Triacylglycerol lipase-like protein
At5g14180	66.017	Lipase
At1g31480	4.399	Unknown; similar to phospholipase
At2g39410	2.918	Putative Phospholipase
At1g28670	2.676	Lipase
At2g39420	2.649	Putative Phospholipase
At5g11650	2.835	Lysophospholipase-like protein
At3g62860	2.125	Putative protein lysophospholipase
At1g28580	5.253	Putative Lipase
EDS1	N/A	Lipase
PAD4	N/A	Lipase

Fig. 2 - 2

1 MAGSV MVPSV SIGLA LSVLI FFALS LKTLE ARGTF GRLAG
41 QPPQR TAAAGG ICASS VHIFG YKCEE HDVVT QDGYI LNMQR
81 IPEGR AGAVA GDGGK RQPVL IQHGI LVDGM SWLLN PADQN
121 LPLIL ADQGF DVWMG NTRGT RFSRR HKYLN PSQRA FWNWT
161 WDELV SYDLP AMFDH IHGLT GQKIH YLGHS LGTLI GFASF
201 SEKGL VDQVR SAAML SPVAY LSHMT TVIGD IAAKT FLAEA
241 TSILG WPEFN PKSGL VGDFI KAICL KAGID CYDLV SVITG
281 KNCCL NASTI DLFLA NEPQS TSTKN MIHLA QTVRD KELRK
321 YNYGS SDRNI KHYGQ AIPPA YNISA IPHEL PLFFS YGGLD
361 SLADV KDVEF LLDQF KYHDI DKMVN QFVKD YAHAD FIMGV
401 TAKDV VYNQV ATFFK RQA

GxSxG motif 1

Catalytic triad: Ser, Asp, His

Fig. 2 – 3

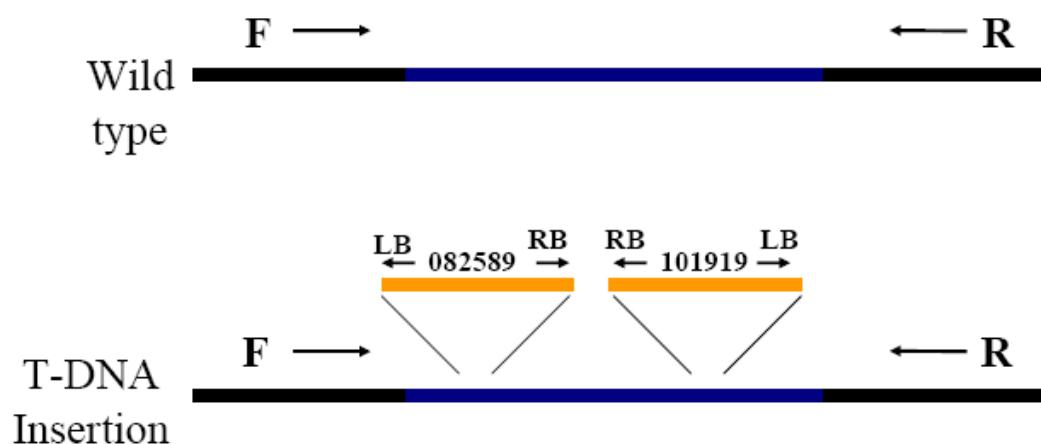


Fig. 2 – 4

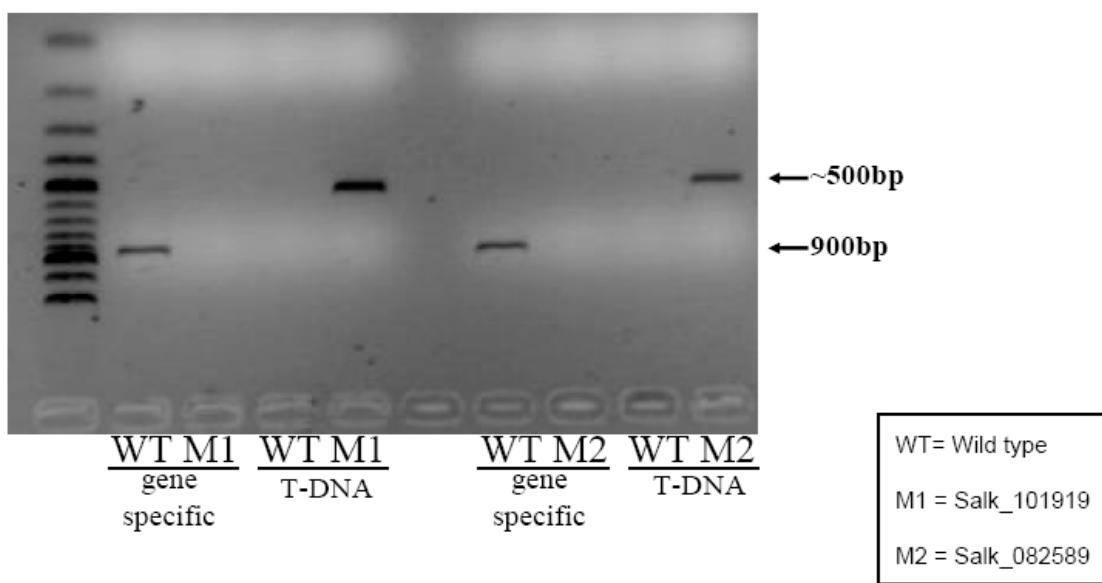


Fig. 2 - 5

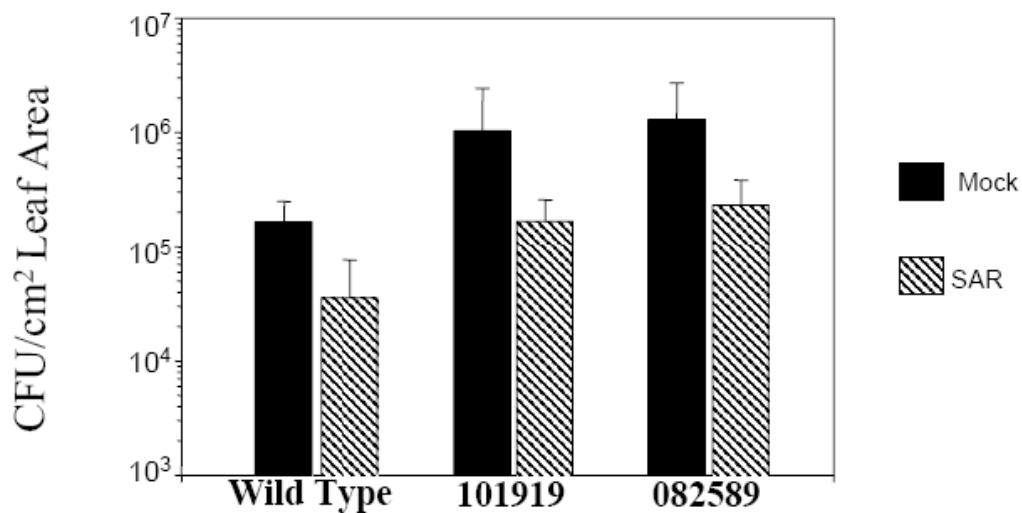


Fig. 2 - 6

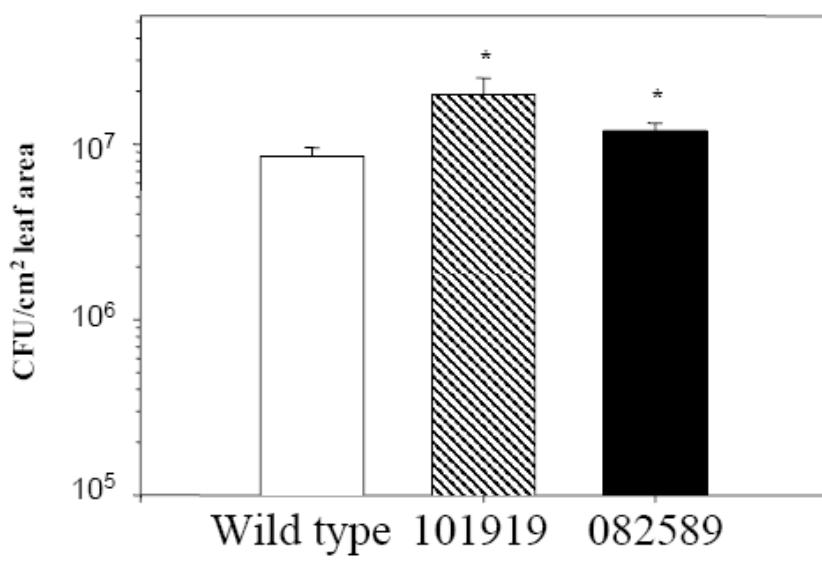


Fig. 2 – 7

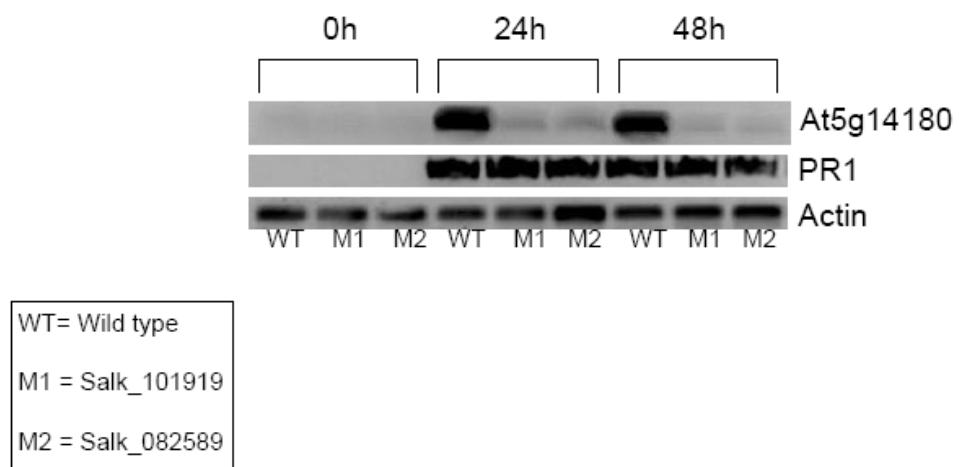


Fig. 2 - 8

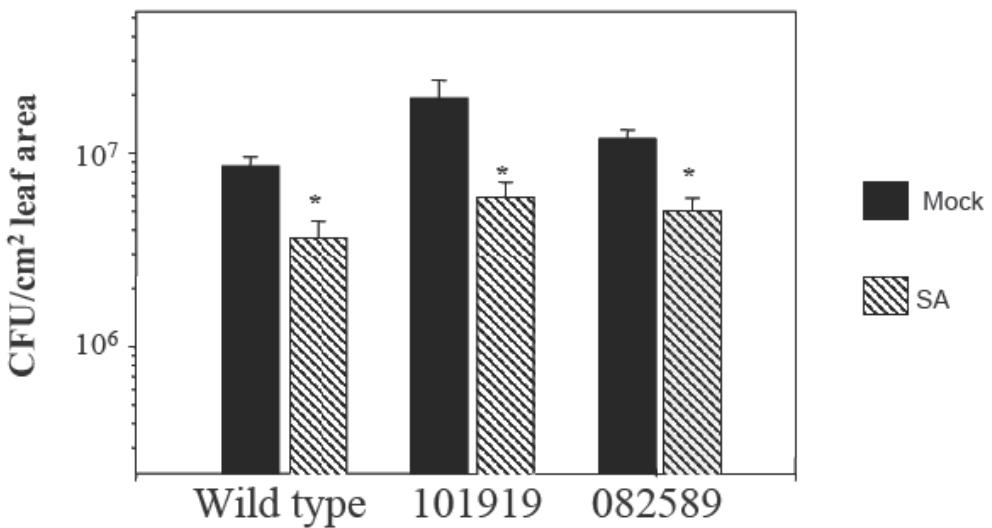
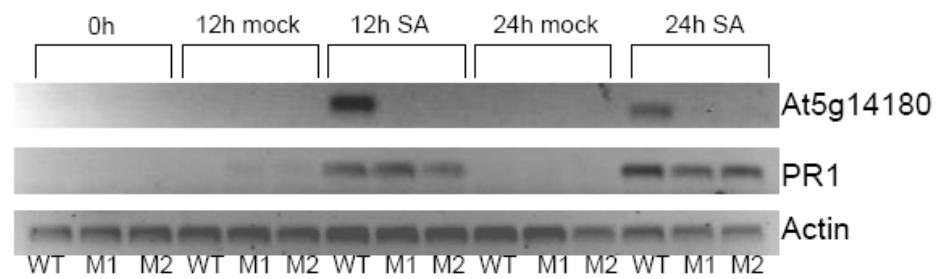


Fig. 2 – 9



WT= Wild type
M1 = Salk_101919
M2 = Salk_082589

Fig. 2 - 10

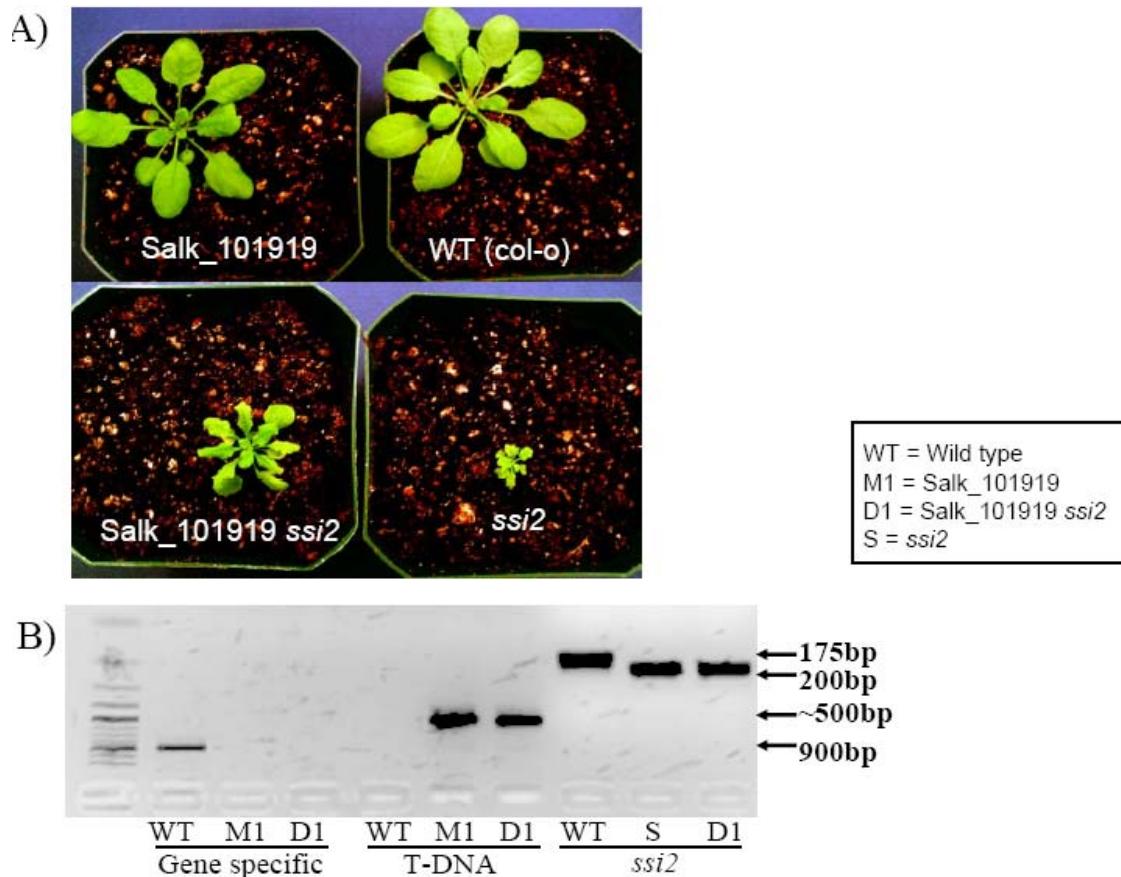


Fig. 2 – 11

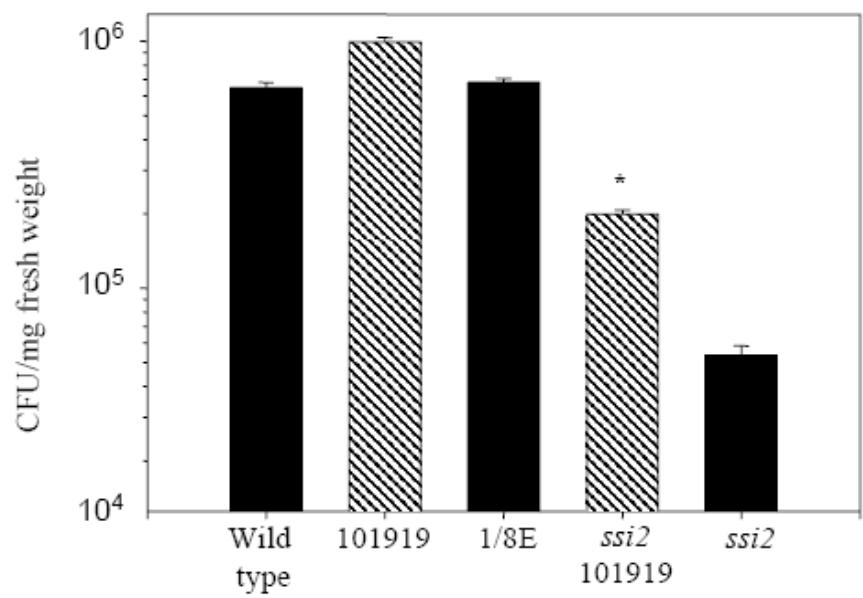
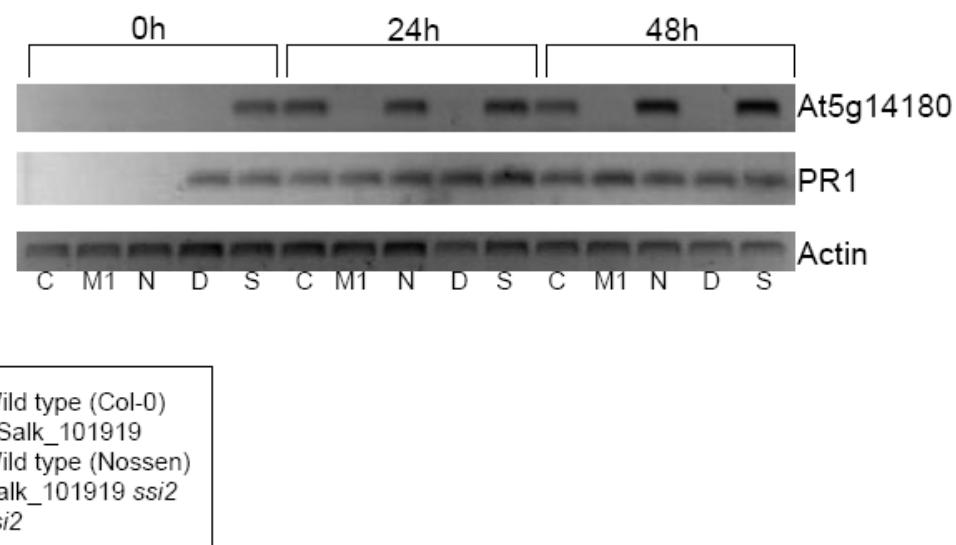


Fig 2 – 12



CHAPTER THREE

Quantitative analysis of *Arabidopsis galactolipids and oxylipins in At5g14180 T-DNA insertion mutation lines*

INTRODUCTION

In higher plants, fatty acid synthesis occurs in the plastid, resulting in the synthesis of palmitic acid (16:0), stearic acid (18:0) and oleic acid (18:1)-ACP (Somerville et al., 2000). These fatty acids can then enter the prokaryotic pathway, localized in the plastidic inner envelope or can enter the eukaryotic pathway by exportation into the cytoplasm as CoA thioesters (Somerville, 2000). Both pathways are initiated by the formation of phosphatidic acid, occurring via two acylation reactions transferring fatty acids from acyl-ACP or acyl-CoA to glycerol-3-phosphate, by the prokaryotic and eukaryotic pathways, respectively. In the prokaryotic pathway, chloroplast lipids are synthesized, including phosphatidylglycerol (PG), monogalactosyldiacylglycerol (MGDG), digalactosyldiacylglycerol (DGDG) and sulfoquinovosyldiacylglycerol (SQDG). PA is used to synthesize PG or can be converted to diacylglycerol (DAG) by means of phosphatidic acid-phosphatases (Somerville et al., 2000). DAG serves as the precursor for the synthesis of MGDG, DGDG and SQDG.

In the eukaryotic pathway, fatty acids exported from the chloroplast are used to synthesize PA. This ER-derived PA can further be channeled for synthesis of phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylinositol (PI) and phosphatidylserine (PS), by addition of the respective head groups. Some PC or PC-derived products are transported back to the plastid where they are used to synthesize

some species of plastid-localized lipids – MGDG, DGDG and SQDG using an acyl-glycerol component (Mongrand et al., 2000). Examples of these modified PC plastid-localized lipids include 36:6 MGDG and 36:6 DGDG (Mongrand et al., 2000).

Oxylipins, a group of biologically active compounds, are synthesized from polyunsaturated fatty acids (Howe and Shilmiller, 2002). Jasmonates, including jasmonic acid (JA), are oxylipins well-characterized for their role in plant defense signaling. JA synthesis and accumulation occurs in response to wounding associated with herbivore attack (Kessler and Baldwin, 2002) and in response to the systemic resistance induced by nonpathogenic rhizosphere bacteria colonizing plant roots (Van Loon et al., 1998). JA accumulation induces the expression of genes involved in plant defense, including genes encoding protease inhibitors, which help protect the plant from insect damage (Ranjan and Lewark, 1992) and genes involved in phytoalexin biosynthesis – Chs, Pal, HMGR (Creelman et al., 1992). JA also contributes to plant defense against the necrotrophic pathogen *Botrytis cinerea* (Thomma et al., 1998). JA accumulation also has an antagonistic effect on SA levels (Kunkel and Brooks, 2002). Jasmonates are synthesized from linolenic acid (Creelman and Mullet, 1997). Linolenic acid is converted to 13-hydroperoxylinolenic acid by lipoxygenase and subsequently produced by an allene oxide synthase (AOS) and an allene oxide cyclase (AOC) – dependent pathway (Creelman and Mullet, 1997). These early steps occur in the plastid, resulting in the synthesis of the intermediate compound OPDA. OPDA is then exported out of the plastids into the peroxisomes where it is further acted upon by OPDA reductase, followed by three rounds

of deacetylation to yield JA. JA can further be converted into its methyl ester and amino acid esters.

Three classes of phospholipases, A, C, and D have been shown to be involved in the plant defense response (Dhondt et al., 2002; de Jong et al., 2004; de Torres et al., 2002; Van der Luit et al., 2000; Viehweger et al., 2002; Yamaguchi et al., 2003 and Zhang et al., 2003). Phospholipase A involvement has been implicated in JA biosynthesis during the activation of defense gene responses by releasing a fatty acid precursor of JA (Dhondt et al., 2002). Phospholipase C activity was shown to be involved in short-term accumulation of PA during an incompatible gene interaction of the Cf4 tomato gene and by the *Cladosporium fulvum* AVR4 elicitor (de Jong et al., 2004). Phospholipase D activity has been shown to be stimulated in response to *Pseudomonas* challenge in Arabidopsis (de Torres et al., 2002), and by the addition of microbial elicitors to tomato suspension cells (Van der Luit et al., 2000) and to rice suspension cells (Yamaguchi et al., 2003). Additionally, two genes, *PAD4* (Phytoalexin-deficient4) and *EDS1* (enhanced disease susceptibility1), which are required for activation of SAR encode proteins with sequence similarity to putative acyl hydrolases (Feys et al., 2001).

The *ssi2* mutant plant, as discussed in chapter two, constitutively expresses *PR1* genes, accumulates high levels of SA and exhibits enhanced resistance to both bacterial and oomycete pathogens (Shah et al., 2001). Additionally, the *ssi2* mutant displays a dwarf phenotype compared to wild type plants and exhibit spontaneous cell death and lesions (Kachroo et al., 2001; Shah et al., 2001). *SSI2* encodes a plastid-localized steroyl-

ACP desaturase, that catalyzes the desaturation of stearic acid (18:0)-ACP to oleic acid (18:1)-ACP (Kachroo et al., 2001), one of the key steps in the fatty acid biosynthesis pathway regulating levels of unsaturated fatty acids in the cells. The desaturation of stearic acid (18:0) to oleic acid (18:1) is the first step in producing the JA precursor, linolenic acid (18:3) (Farmer and Ryan, 1992). In the *ssi2* mutant, fatty acid content and complex lipid composition is altered (Kachroo et al., 2001; Nandi et al., 2003).

Compared to the lipid profile of the wild type plant, the *ssi2* mutant plant exhibits elevated levels of 18:0 membrane lipids, but reduced levels of 16:3, 18:1 and 18:2 membrane lipid content (Kachroo et al., 2001; Nandi et al., 2003). Thus, the involvement of lipids and/or lipid composition is implicated in plant defense and SA signaling.

In this chapter, the lipid profile of the At5g14180 T-DNA insertion lines, Salk_101919 and Salk_082589 was analyzed and compared to that of wild type. Both galactolipid and oxylipin profiles were characterized using ESI MS/MS and GC/MS respectively. The Salk_101919 and Salk_082589 mutants were characterized as having elevated levels of 16:0, 18:0, 18:1, and 18:2 molecular species, as compared to the wild type plant. Further, the Salk_101919 and Salk_082589 mutant lines exhibited elevated levels of jasmonates, as compared to the wild type plant. Additionally, the Salk_101919 and Salk_082589 mutant plants exhibited a decrease in the ratio of 36:6/36:4 species in both PC and PE, caused by a decrease in 36:6 PC and PE levels and an increase in 36:4 PC and PE levels, as compared to the wild type plant. Likewise, the ratio of 34:3/34:2 PC and PE levels, due to an increase in the levels of 34:2 in the mutant plants, as compared to wild type.

MATERIALS AND METHODS

Plant Growth Conditions

Arabidopsis plants were grown by sowing seeds in standard soil mix, placing two seeds in opposite corners of a 3-inch square pot. Pots were then covered with plastic and kept in a 4°C chamber for 72 hours. Plants were then transferred to a growth chamber programmed for 14 hours light and 10 hours dark with an average temperature of 22°C. Plants continued to be covered for one week with clear plastic in order to maintain high humidity. Plants were fertilized using Scotts fertilizer (The Scotts Company, Marysville, OH) according to manufacturer's instructions every two weeks.

Arabidopsis Mutants

The transgenic Arabidopsis lines containing a T-DNA insertion in the At5g14180 gene used in this study were obtained from the Ohio State University Stock Center from the Salk collection (<http://www.signal.salk.edu>). The Salk lines, Salk_101919 and Salk_082589 are in the ecotype Columbia background and are described in chapter two.

Salicylic Acid Treatment

Wild type, Salk_101919 and Salk_082589 plants were treated by spraying and subirrigation with either a 0.05 mM salicylic acid solution (SA) or water (mock). Samples were taken at 0 hrs, 12 hrs and 24 hrs for lipid extraction and ESI-MS/MS analysis or oxylipin extraction and analysis.

Lipid Extraction and ESI-MS/MS Analysis

Four-week old plants were treated with either water (mock) or 0.05mM salicylic acid solution and sampled at 0 hr, 12 hr and 24 hr time points. Lipid extraction on these plant tissues was performed according to the method previously described (Welty et al., 2002). From each plant, 3 – 5 leaves were collected and immersed in 75°C isopropanol with 0.01% butylated hydroxytoluene for 15 minutes. Following the isopropanol extraction, multiple extractions using chloroform/methanol (2:1) were performed until the plant tissue was translucent and colorless. The extracted solvent was washed once with 1 ml of 1 M KCl and once with 2 ml water. The solvent was then evaporated and redissolved in exactly 1 ml of chloroform. The remaining plant tissue was heated overnight at 105°C and weighed to produce the plant dry weight, minus lipid.

From the 1 ml extract, 20 μ L of plant extract was combined with phospholipid and galactolipid internal standards, along with solvents. Phospholipid standard is composed of 0.660 nmol LysoPC 13:0, 0.660 nmol Lyso PC 19:0, 0.630 PC 28:0, 0.544 nmol PC 48:2, 0.378 nmol LysoPE 14:0, 0.344 nmol LysoPE 18:0, 0.381 nmol PE 28:0, 0.309 nmol PE 48:2, 0.352 nmol lyso PG 14:0, 0.347 nmol lyso PG 18:0, 0.313 nmol PG 28:0, 0.233 nmol PG 48:2, 0.302 nmol PA 28:0, 0.317 nmol PA 40:0, 0.228 nmol PS 28:0 and 0.230 nmol PS 40:0. Galctolipid standard is composed of 2.008 nmol MGDG 34:0, 0.392 nmol MGDG 36:0, 0.494 nmol DGDG 34:0 and 0.706 nmol DGDG 36:0. Solvent ratio was 300:665:35 chloroform/methanol/ammonium acid.

The samples were then analyzed using a “triple” quadrupole tandem mass spectrometer (API – 400 Applied Biosystems, Foster City, CA) equipped for electrospray ionization.

Oxylipin Extraction and Analysis

Samples were prepared by the method previously described (Schmelz et al., 2004). Plant leaf tissue was weighed, frozen in liquid nitrogen and placed in 2 ml screw cap Fast Prep® tubes containing 1g Zirmil® beads (1.1 mm; SEPR ceramic beads and Powders, Mountainside, NJ, USA) along with DhJA and isotopically labeled internal standards (100 ng each in 5µl EtOH) and 300 µl of 1-propanol:H₂O:HCl (2:1:0.005) extraction buffer. Samples were then shaken for 20 s with a FastPrep® FP 120 tissue homogenizer (Obiogene). 1 ml CHCl₃ was added and samples were again shaken for 20s and centrifuged at 11,300 for 30s. The bottom layer containing CHCl₃:1-propanol was pippetted and transferred to a 4 ml glass vial sealed with a teflon lined screw cap (SUN SRI; Wilmington, NC, USA). Samples were then vapor phase extracted and analyzed by chemical ionization gas chromatography/mass spectrometry as previously described (Schmelz et al., 2004).

RESULTS

The wild type At5g14180 gene is predicted to encode a putative lipase, based on the presence of the Serine-Aspartic Acid-Histidine (Ser Asp His) catalytic triad, which is required for hydrolytic activity in other lipases, in the At5g14180 encoded protein sequence (Fig. 2 – 2). Furthermore, the wild type At5g14180 gene contains a putative signal peptide targets the product to the vacuole (Fig. 2 – 2). In order to determine the effect of the loss of the At5g14180 gene activity on lipid composition of the Salk_101919 and Salk_082589 mutant plants, as compared to wild type and possibly identify a substrate for lipase activity, oxylipin and complex lipid quantification and analysis was performed.

Oxylipin analysis using GC/MS characterized the free fatty acid and phytohormone content of the Salk_101919 and Salk_082589 mutant plants as compared to the wild type plant under normal conditions and after treatment with water (mock) or 0.05mM Salicylic acid solution (SA) (Appendix, table A – 1, table A – 2, table A – 3). In the Salk_101919 and Salk_082589 mutant plants, levels of 16:0 free fatty acids (Fig. 3 – 1), 18:0 free fatty acids (Fig. 3 – 2), 18:1free fatty acids (Fig. 3 – 3) and 18:2 free fatty acids (Fig. 3 – 4) were shown to be elevated in the Salk_101919 and Salk_082589 mutant plants, as compared to the wild type control, regardless of treatment type. Additionally, in the Salk_101919 and Salk_082589 mutants, displayed elevated levels of jasmonates, under all treatment conditions, in relation to levels of jasmonates in the wild type plant (Fig. 3 – 5).

Quantification and analysis of complex lipid species using ESI MS/MS, characterized the lipid profile of the major lipid classes from the prokaryotic pathway, including phosphatidylglycerol (PG), monogalactosyldiacylglycerol (MGDG), digalactosyldiacylglycerol (DGDG) as well as the eukaryotic pathway classes, phosphatidic acid (PA), phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylinositol (PI) and phosphatidylserine (PS). The lipid profile was analyzed under control or basal conditions and after treatment with either water (mock) or 0.05 mM salicylic acid solution (SA). The lipid profile of the Salk_101919 and Salk_082589 mutants, as compared to the wild type plant, did not appear to differ significantly from wild type in total mol% of each of the major lipid classes, under control or basal conditions or after treatment (Appendix: Chart A – 4, Chart A – 5 and Chart A – 6). However, analysis of the individual molecular species identified changes in the mol% ratios of 36:6/36:4 PC, 34:3/34:2 PC, 36:6/36:4 PE and 34:3/34:2 PE (Fig. 3 – 6).

The decrease in the mol% ratio of 36:6/36:4 PC was the result of a decrease in mol% of 36:6 (di18:3) PC and an increase in mol% of 36:4 (di18:2) PC in the Salk_101919 and Salk_082589 mutant plants, as compared to the wild type plant (Fig 3 – 6). Similarly, the decreased mol% ratio of 36:6/36:4 PE was the result of a decrease in the mol% of 36:6 (di18:3) PE and an increase in mol% of 36:4 (di18:2) PE in the Salk_101919 and Salk_082589 mutant plants, as compared to the wild type plant (Fig. 3 – 6).

The decrease in the mol% ratio of 34:3/34:2 PC in the Salk_101919 and Salk_082589 mutant plants as compared to the wild type plant, was the result of decreased 34:3 (16:0-18:3) PC in the Salk_101919 and Salk_082589 mutant plants with little change in the amount of 34:2 (16:0-18:2) PC (Fig 3 – 6). Likewise, the decrease in the mol % ratio of 34:3/34:2 PE in the Salk_101919 and Salk_082589 mutant plants, in relation to wild type levels, resulted from decreased 34:3 (16:0-18:3) PE in the mutant plants, with little change in the mol % of 34:2 (16:0-18:2) PE (Fig. 3 – 6).

DISCUSSION

Changes in the lipid profile of the Salk_101919 and Salk_082589 mutant lines appeared to occur in species produced by the eukaryotic pathway, which functions outside of the chloroplast. The observed changes in PC and PE species give further evidence for the functioning of the putative lipase encoded by the At5g14180 wild type gene in a compartment outside of the chloroplast. The changes in 36:6/36:4 mol % ratios of PC and PE, in the Salk_101919 and Salk_082589 mutant plants in relation to wild type plants (Fig. 3 – 6) could likely be the result of a reduction in the rate of steps leading from 36:4 PC and PE to 36:6 PC and PE. Similarly, the changes in the mol % 34:3/34:2 ratio of PC and PE in the Salk_101919 and Salk_082589 mutant plants in relation to wild type ratios (Fig. 3 – 6), might also be caused by a reduction in the rate of steps leading to synthesis of 34:3 PC and PE from 34:2 PC and PE. According to the current dogma, desaturation of 18:1 to 18:2 and of 18:2 to 18:3 occurs in complex lipids. Hence, a relative reduction in the activity of the *FAD3* encoded desaturase, which catalyzes the desaturation of dienoic fatty acids in phospholipids to trienoic fatty acids could result in increased content of dienoic fatty acid containing 36:4 and 36:2 PC and PE species. Alternatively, the changes in the levels of 36:4 and 34:2 could be the result of reduced recycling of 36:4 and 34:2 products, resulting in the relative reduction (mol%) in levels of 36:6 and 34:3 PC and PE. For example, the At5g14180 encoded protein may be involved in the replacement of dienoic fatty acids by a trienoic fatty acid resulting in the synthesis of 36:6 PC and PE from 36:4 PC and PE. Thus, loss of At5g14180 activity in the Salk_101919 and Salk_082589 mutant plants is expected to result in the build up in levels of 36:4 and 34:2 PC and PE in comparison to the wild type plant.

Changes in the levels of 36:6, 36:4, 34:3 and 34:2 species of PC and PE as determined by the ESI MS/MS analysis of the major lipid classes (Fig 3 – 6) correlated with the changes in levels of 16:0, 18:1 and 18:2 free fatty acids, as determined by the GC-MS analyses. The increase in levels of 16:0, 18:0, 18:1 and 18:2 free fatty acids in the At5g14180 mutants as compared to the wild type plant, could result from increased synthesis of these free fatty acids in the mutant plants, presumably due to increased flux of C through the fatty acid biosynthesis pathway in the mutant plants. Alternatively, the increase in levels of these free fatty acids could be explained by a higher rate of release of these fatty acids from phospholipids, presumably due to an involvement of the At5g14180 encoded protein in deacylation/reacylation of phospholipids. For example, if the At5g14180 protein functions as an acyl transferase then loss of this activity would be expected to decrease the reincorporation of free fatty acids into phospholipids. However, if this is the case, then we expect that the At5g14180 encoded protein has a preference for dienoic over trienoic fatty acids since, the level of free 18:3 was not affected in the mutant plants.

The increased level of jasmonates in the Salk_101919 and Salk_082589 mutant plants, in relation to wild type plants (Fig 3 – 5), could result from an overall increase in levels of free 16C and 18C fatty acids in the mutant plant, creating a greater pool of free fatty acids for jasmonate production in the mutant. The major substrate of jasmonate biosynthesis is linolenic acid (18:3), which is produced from stearic acid (18:0) via oleic acid (18:1) and octadecadienoic acid (18:2) (Creelman and Mullet, 1997). Increase flux

of fatty acids through this pathway may result in increased availability of 18:3, which could then be converted into jasmonates. Alternatively, the higher levels of jasmonates may result from increased sensitivity of the At5g14180 mutant plant to some unknown stress; jasmonate levels in Arabidopsis are known to change in response to both, biotic and abiotic stress.

Although the lipid profiles of the Salk_101919 and Salk_082589 mutant lines got from the ESI-MS/MS and GC-MS analysis do not provide any clear indication of the biochemical function of the At5g14180 gene, the changes in the lipid and oxylipin profiles of the Salk_101919 and Salk_082589 mutant lines as compared to the wild type plant, indicate the At5g14180 gene does serve some role in lipid metabolism and composition in the wild type plant. However, the exact function of the gene and protein product cannot be determined without cloning and expression of the protein product to determine enzymatic activity.

FIGURE LEGENDS

Fig. 3 – 1. Profile of 16:0 free fatty acid content in At5g14180 T-DNA insertion mutant lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with either water (mock) or a 0.05 mM Salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points. Samples were prepared by the extraction method described in Schmelz et al., 2004. Samples were quantified and analyzed by GC/MS. Specifically, the 16:0 fatty acid profile is shown. Salk_101919 and Salk_081589 mutants appear to have an increased 16:0 fatty acid content, as compared to wild type, regardless of treatment type, expressed in ng/g of fresh weight.

Fig. 3 – 2. Profile of 18:0 free fatty acid content in At5g14180 T-DNA insertion mutant lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with either water (mock) or a 0.05 mM Salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points after treatment. Samples were prepared by the extraction method described in Schmelz et al., 2004. Samples were quantified and analyzed by GC/MS. Specifically, the 18:0 fatty acid profile is shown. Salk_101919 and Salk_081589 mutants appear to have increased 18:0 fatty acid content, as compared to wild type, regardless of treatment type, expressed in ng/g fresh weight.

Fig. 3 – 3. Profile of 18:1 free fatty acid content in At5g14180 T-DNA insertion mutant lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed

and subirrigated with either water (mock) or a 0.05 mM salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points after treatment. Samples were prepared by the extraction method described in Schmelz et al., 2004. Samples were quantified and analyzed by GC/MS. Specifically, the 18:1 fatty acid profile is shown. Salk_101919 and Salk_081589 mutants appear to have an increased fatty acid content, as compared to wild type, regardless of treatment type, expressed in ng/g fresh weight.

Fig. 3 – 4. Profile of 18:2 free fatty acid content in At5g14180 T-DNA insertion mutant lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with either water (mock) or a 0.05 mM salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points after treatment. Samples were prepared by the extraction method described in Schmelz et al., 2004. Samples were quantified and analyzed by GC/MS. Specifically, the 18:2 fatty acid profile is shown. Salk_101919 and Salk_081589 mutants appear to have an increased 18:2 fatty acid content, as compared to wild type, regardless of treatment type, expressed in ng/g fresh weight.

Fig. 3 – 5. Profile of jasmonate content in At5g14180 T-DNA insertion mutant lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with either water (mock) or a 0.05 mM Salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points. Samples were prepared by the extraction method described in Schmelz et al., 2004. Samples were

quantified and analyzed by GC/MS. Specifically, the jasmonate profile is shown. Salk_101919 and Salk_081589 mutants appear to have an increased jasmonate content, as compared to wild type, regardless of treatment type, expressed in ng/g fresh weight.

Fig. 3 – 6. Ratio of 36 and 34 carbon molecular species of PC and PE in At5g14180

T-DNA insertion lines. Wild type (Col-0), Salk_101919 and Salk_082589 plants were sprayed and subirrigated with either water (mock) or a 0.05 mM Salicylic acid solution (SA). Leaves from mock and SA treated plants were then sampled at 0h, 12h and 24h time points after treatment. Galactolipid species were extracted and analyzed using a “triple” quadrupole tandem mass spectrometer (API – 4000, Applied Biosystems, Foster City, CA) equipped for electrospray ionization, as described in Welti et al., (2002). Ratios of 36:6/36:4 phosphatidylcholine, 34:3/34:2 phosphatidylcholine, 36:6/36:4 phosphatidylethanolamine, and 34:3/34:2 phosphatidylethanolamine lipid profiles, expressed in mol%.

Fig. 3 – 1

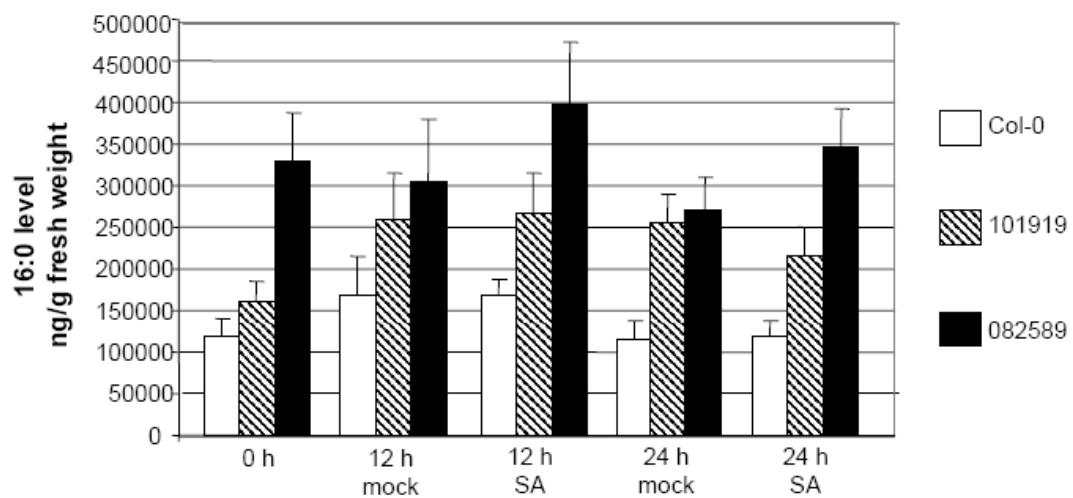


Fig. 3 – 2

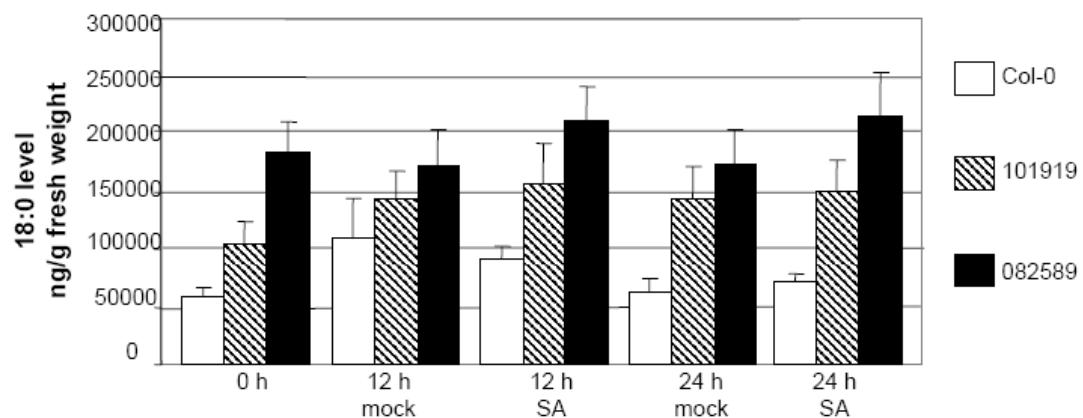


Fig. 3 – 3

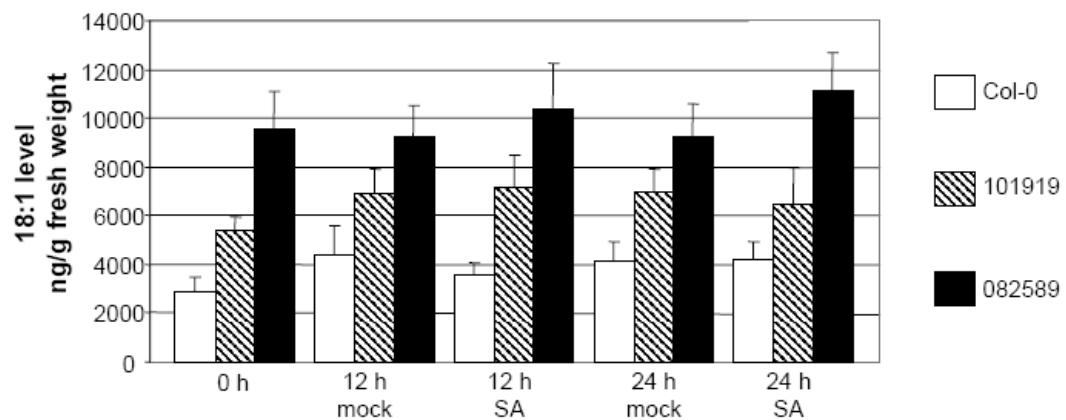


Fig. 3 – 4

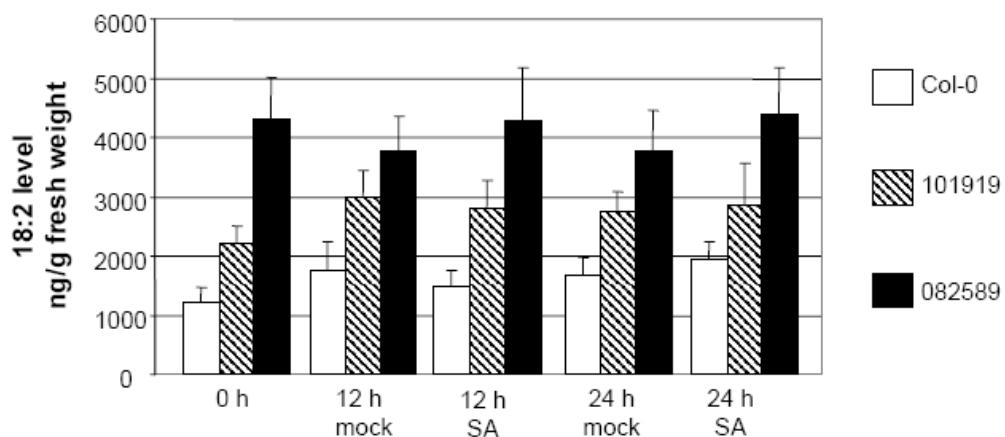


Fig. 3 – 5

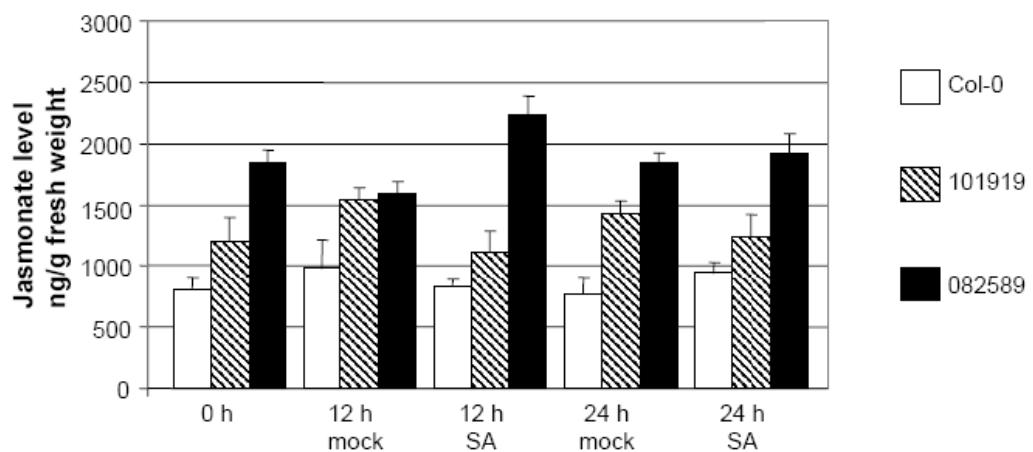
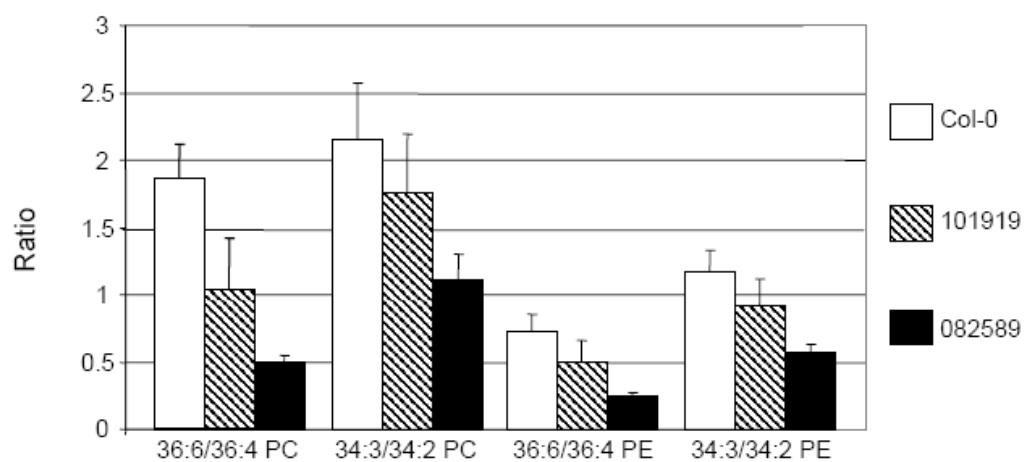


Fig. 3 – 6



CHAPTER FOUR

Conclusions and Future Directions

In this study, the function of the At5g14180 gene, which is expressed at elevated levels in the *ssi2* mutant plant (Fig. 2 – 1) and encodes a putative lipase (Fig 2 – 2), was characterized in *Arabidopsis* defense and the impact of this gene on lipid composition characterized by comparing lipid composition between the Salk_101919 and Salk_082589 T-DNA insertion mutations in At5g14180 and the wild type plant. The Salk_101919 and Salk_082589 T-DNA mutants were found to not be impaired in SAR activity (Fig. 2-5) and were able to respond to the application of exogenously applied SA (Fig. 2 – 8), suggesting that this gene does not have a role in SAR. However, basal resistance to the virulent pathogen *P. syringae ps. maculicola* was found to be impaired in the Salk_101919 and Salk_082589 mutant lines as compared to the wild type plant (Fig. 2 – 6). Furthermore, the presence of the Salk_101919 allele attenuated the *ssi2*-conferred enhanced resistance to the virulent pathogen *P. syringae* pv. *maculicola* ES4326, as shown by greater bacterial grown seen in the Salk_101919 *ssi2* double mutant as compared to the *ssi2* mutant plant (Fig. 2 – 11).

GC/MS analysis of lipid composition in the Salk_101919 and Salk_082589 T-DNA insertion mutants showed increased levels of free 16:0 (Fig. 3 – 1), 18:0 (Fig 3 – 2), 18:1 (Fig. 3 – 3) and 18:2 (Fig. 3 – 4) fatty acid species, in the mutant as compared to the wild type plant. Additionally, the Salk_101919 and Salk_082589 mutant lines contained an increase in the level of jasmonates (Fig. 3 – 5), as compared to the wild type plant. Quantification and analysis of complex lipid species using ESI MS/MS, demonstrated

changes in the major lipid classes, phosphatidylcholine (PC) and phosphatidylethanolamine (PE), in the Salk_101919 and Salk_082589 mutant plants as compared to the wild type plant (Fig. 3 – 6). Specifically, the ratio of 36:6/36:4 PC and PE as well as 34:3/34:2 PC and PE, was shown to be decreased in the Salk_101919 and Salk_082589 mutant plants (Fig. 3 – 6).

The weak effect of the Salk_101919 and Salk_082589 mutant alleles on pathogen growth in the single mutant, could be explained by the increase in jasmonates shown by lipid profiling. JA and SA have been shown to have an antagonistic effect towards the other (Felton et al., 1999), therefore increased JA levels may decrease SA levels and therefore increase the susceptibility to the virulent pathogen *P. syringae* pv. *maculicola*. The weak effect of these mutant alleles on pathogen growth in the single mutant plants as opposed to the strong effect of the Salk_101919 insertion on the *ssi2*-conferred heightened resistance phenotype could be due to the fact that basal expression of the At5g14180 gene is very high in the *ssi2* mutant plant and relatively poor in the wild type plant (Fig. 2 – 12). The Salk_101919 *ssi2* double mutant maintained the dwarf stature associated with the *ssi2* allele, albeit the double mutant being slightly larger in size than the *ssi2* mutant (Fig. 2 – 10A). This is similar to the dwarf phenotype of the *ssi2 pad4*, *ssi2 nahG* and *ssi2 eds5* plants, all of which are compromised in *ssi2*-conferred enhanced resistance (Shah et al., 2001; Nandi et al., 2005). The *PAD4*, *EDS5* and *nahG* transgene impact SA signaling/synthesis (Durrant and Dong, 2004; Chaturvedi and Shah, 2006). This high level expression of At5g14180 most likely has an important contribution to the *ssi2*-conferred resistance to the virulent pathogen *P. syringae* pv. *maculicola* ES4326.

The At5g14180 gene may be associated with the NPR1-independent mechanism, which is overshadowed by the NPR1-dependent pathway in wild type plants. However, the NPR1-independent pathway does contribute significantly to the *ssi2*-conferred resistance (Shah et al., 2001), thereby explaining the more pronounced effect of the Salk_101919 and Salk_082589 mutant alleles on the *ssi2*-conferred resistance. Alternatively, although not exclusively, in the wild type plant there are likely redundant activities contributed by other lipases that may substitute in the absence of At5g14180 single mutant plants, Salk_101919 and Salk_08289, than in the *ssi2* mutant, in which the At5g14180 gene is expressed at elevated levels.

Changes in the lipid profile of the Salk_101919 and Salk_082589 mutant plants, specifically in the major lipid classes, PC and PE, indicate that the product encoded by the At5g14180 gene is serving some role in lipid metabolism and composition. These changes are seen specifically in species produced by the eukaryotic pathway, which functions outside of the chloroplast. The protein sequence of the At5g14180 gene contains a putative signal peptide for vacuolar localization (Fig. 2 – 2), so these changes indicate the At5g14180 encoded protein does have a role in lipid metabolism, functioning at some point within the eukaryotic pathway.

The changes in 36:6/36:4 mol% ratios of PC and PE, in the Salk_101919 and Salk_082589 mutant plants in relation to wild type plants (Fig. 3 – 6) could likely be the result of a relative reduction in flux leading from 36:4 PC and PE to 36:6 PC and PE. Alternatively, the changes in the levels of 36:4 and 36:6 could be the result of increased

production and recycling of a 36:4 product, leaving less 36:4 PC and PE present to produce the 36:6 PC and PE product. The changes in the mol% 34:3/34:2 ratio of PC and PE in the Salk_101919 and Salk_082589 mutant plants in relation to wild type ratios (Fig. 3 – 6), might also be caused by a block in the pathway leading to synthesis of 34:3 PC and PE from 34:2 PC and PE. Again, this ratio change could alternatively be explained by increased production and recycling of a 34:2 PC and PE product, leading to the decreased production of the 34:3 PC and PE products.

To fully establish and characterize the function of the At5g14180 gene and gene-related product, further studies must be carried out. The At5g14180 gene should be cloned and expressed in *E. coli* or other expression system to characterize the nature and function of the protein product. Furthermore, the At5g14180 gene should be overexpressed and the basal defense response to *P. syringae ps maculicola* ES4326 should be assessed for enhanced resistance. In addition, an At5g14180 *npr1* double mutant plant would provide further information on possible involvement of the At5g14180 gene in the *NPR1*-independent mechanism. Finally, computational search of the *Arabidopsis* genome database for other genes that encode proteins similar to At5g14180 combined with RNAi or T-DNA insertional lines could be used to study lipid-metabolic proteins similar to that produced by the At5g14180 gene to assess the extent of genetic redundancy and its contribution to *Arabidopsis* defense.

References

- Aubel FM (2005). Are innate immune signaling pathways in plants and animals conserved? *Nat. Immunol.* 6: 973 – 979.
- Cao H, Bowling SA, Gordon AS and Dong X (1994). Characterization of an Arabidopsis mutant that is nonresponsive to inducers of systemic acquired resistance. *Plant Cell* 6: 1583 – 1592
- Cao H, Glazebrook J, Clarke JD, Volko S and Dong X (1997). The arabidopsis NPR1 gene that controls systemic acquired resistance encodes a novel protein containing ankyrin repeats. *Cell* 88: 57 – 63
- Cao H, Li X and Dong X (1998). Generation of broad-spectrum disease resistance by overexpression of an essential regulatory gene in systemic acquired resistance. *Proc Natl Acad Sci.* 95: 6531 – 6536
- Chaturvedi R and Shah J (2006). Salicylic Acid in Plant Disease Resistance. *Salicylic Acid: A Plant Hormone* pp. 335 – 370.
- Chern M, Fitzgerald H, Canlas P, Navarre D and Ronald P (2005). Overexpression of a Rice NPR1 homolog leads to constitutive activation of defense response and hypersensitivity to light. *Molecular Plant-Microbe Interactions* 18: 511 - 520
- Chomczynski P and Sacchi N (1987). Single-step method for RNA isolation by acid guanidinium thiocyanate-phenol-chloroform extraction. *Anal. Biochem* 162: 156 – 159
- Creelman RA, Tierny ML, and Mullet JE (1992). Jasmonic acid/methyl jasmonate accumulate in wounded soybean hypocotyls and modulate wound gene expression. *Proc. Natl. Acad. Sci. USA* 89: 4938 – 4941
- Creelman R and Mullet J (1997). Biosynthesis and action of jasmonates in plants. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* 48: 355 – 381
- De Jong C, Laxalt A, Barmann B, De Wit P, Joosten M and Munnik T (2004). Phosphatidic acid accumulation is an early response in the Ct4/Avr4 interaction. *Plant J.* 39: 1 – 12
- Delaney TP, Friedrich L and Ryals JA (1995). Arabidopsis signal transduction mutant defective in chemically and biologically induced disease resistance. *Proc. Natl. Acad. Sci. USA* 92: 6602 – 6606
- Dhondt S, Gouzerh G, Miller A, Legrand M and Heitz T (2002). Spatio-temporal expression of patatin-like lipid acyl hydrolases and accumulation of jasmonates in

elicitor-treated tobacco leaves are not affected by endogenous levels of salicylic acid.
Plant J. 32: 749 – 762

Doares SH, Narvaez-Vasquez J, Conconi A and Ryan CA (1995). Salicylic acid inhibits synthesis of protease inhibitors in tomato leaves induced by systemin and jasmonic acid.
Plant Physiol. 108: 1741 – 1746

Dong X (2004) Systemic Acquired Resistance. Annual Review of Phytopathology. 42: 185 – 209.

Durrant WE and Dong X (2004) Systemic Acquired Resistance. Annu Rev. Phytopathol. 42: 185 – 209

Fan W and Dong X (2002). In vivo interaction between NPR1 and transcription factor TGA2 leads to salicylic acid mediated gene activation in arabidopsis. Plant Cell 14: 1377 – 1389

Falk A, Feys B, Frost L, Daniels M and Parker J (1999). EDS1, an essential component of R-gene mediated disease resistance in Arabidopsis has homology to eukaryotic lipases. Proc. Natl. Acad. Sci. 96: 3292 – 3297

Farmer E and Ryan C (1992). Octadecanoid precursors of jasmonic acid activate the synthesis of woud-inducible proteinase inhibitors. Plant Cell 4: 129 – 134
Felton GW, Korth KL, Bi JL, Wesley SV, Huhman DV, Matthews MC, Murphey JB, Lamb C, and Dixon RA (1999). Inverse relationship between systemic acquired resistance of plants to microorganisms and to insect herbivory. Current Biology 9: 317 – 320

Feys B, Moisan L, Newman M, and Parker J (2001). Direct interaction between the Arabidopsis disease resistance signaling proteins, EDS1 and PAD4. EMBO J. 20: 5400 – 5411

Feys B and Parker J (2000). Interplay of signaling pathways in plant disease resistance. Trends Genet. 16: 449 – 455

Friedrich L, Vernooij B, Gaffney T, Morse A and Ryals J (1995). Characterization of tobacco plants expressing a bacterial salicylate hydroxylase gene. Plant Molecular Biology 29: 959 – 968

Glazebrook J, Rogers E and Ausubel F (1996). Isolation of Arabidopsis mutants with enhanced disease susceptibility by direct screening. Genetics 143: 973 – 982

Hammond-Kosack KE and Jones JDG (1996). Resistance gene-dependent plant responses. Plant cell 8: 1773 – 1791

Howe G and Schilmiller A (2002). Oxylipin metabolism in response to stress. Current Opinion in Plant Biology 5: 230 – 236

Hunt M and Ryals J (1996). Systemic acquired resistance signal transduction. Crit. Rev. Plant Sci. 15: 583 – 606

Jirage D, Tootle T, Reuber T, Frost L, Feys B, Parker J, Ausubel F and Glazebrook J (1999). *Arabidopsis thaliana* PAD4 encodes a lipase-like gene that is important for salicylic acid signaling. Proc. Nat. Acad. Sci. 96: 13583 – 13588

Jones D and Takemoto D (2003). Plant innate immunity direct and indirect recognition of general and specific pathogen associated molecules. Current Opinion in Immunology. 16: 48 – 62.

Kachroo A, Lapchyk L, Fukushige H, Hildebrand D, Klessig D and Kachroo P (2003). Plastidial fatty acid signaling modulates salicylic acid- and jasmonic acid-mediated defense pathways in the *Arabidopsis ssi2* mutant. Plant Cell 15: 2952 – 2965

Kachroo A, Srivathsac C, Lapchyk L, Fallone D, Hildebrand D, Kachroo P (2004). Oleic Acid levels regulated by glycerolipid metabolism modulate defense gene expression in *Arabidopsis*. Plant J. 40: 647 – 659.

Kachroo P, Shanklin J, Shah J, Whittle E, Klessing D (2001) A fatty acid desaturase modulates the activation of defense signalling pathways in plants. PNAS 98 (16): 9448 – 9453

Kachroo P, Srivathsac C, Navarre D, Lapchyk L, Kachroo A (2005). Role of salicylic acid and fatty acid desaturation pathways in *ssi2*-mediated signaling. Plant Physiol. 139: 1717 – 1735

Kessler A and Baldwin IT (2002). Plant responses to insect herbivory: the emerging molecular analysis. Annu Rev Plant Biol 53: 299 – 328

King EO, Ward MK and Raney DE (1954). Two simple media for the demonstration of phycocyanin and fluorescin. J lab. Clin. Med. 44: 301-307

Kinkema M, Fan W, and Dong X (2000). Nuclear localization of NPR1 is required for activation of PR gene expression Plant Cell 12: 2339 – 2350

Konieczny A, Ausubel, FM (1993) A Procedure for mapping *Arabidopsis* mutations using co-dominant ecotype specific PCR-based markers. Plant J 4: 403-410

Kunkel B and Brooks D (2002). Cross talk between signaling pathways in plant defense. Curr. Opin. Plant Biol. 5: 325 – 331

Maldonado A, Doerner P, Dixon R, Lamb C, Cameron R. (2002). A putative lipid transfer protein involved in systemic resistance signaling in *Arabidopsis*. *Nature* 419: 399 – 403.

Lin W, Lu C, Wu J, Cheng M, Lin Y, Yang N, Black L, Green S, Wang J and Cheng C (2005). Transgenic tomato plants expressing the *Arabidopsis* NPR1 gene display enhanced resistance to a spectrum of fungal and bacterial diseases. *Transgenic Research* 13: 567 – 581

Makandar R, Essig J, Schapaugh M, Trick H and Shah J (2006). Genetically engineered resistance to Fusarium head blight in wheat by expression of *Arabidopsis* NPR1. *Mol. Plant-Microbe Interact.* 19: 123 – 129

Marashige T and Skoog F (1962). A revised media for rapid growth and bioassay with tobacco tissue culture. *Physiol. Plant* 15: 473 – 497.

McConn J, Creelman RA, Bell E, Mullet JE, and Browse J (1997). Jasmonate is essential for insect defense in *arabidopsis*. *Proc. Natl. Acad. Sci. USA* 94: 5473 – 5477

Metraux JP, Ahl-Goy P, Staub T, Speich J, Steinemann A, Ryals J and Ward E (1991). Induced resistance in cucumber in response to 2,6-dichloroisonicotinic acid and pathogens. *Advances in Molecular Genetics of Plant-Microbe Interactions*, Vol 1, H Hennecke and DPS Verma, eds (Dordrecht, The Netherlands Kluher Academic Publishers) pp 432 – 439

Metraux JP, Nawrath C. and Genoud T. (2002). Systemic acquired resistance. *Euphytica.*, 124: 237 – 243

Mongrand S, Cassagne C and Bessoule J (2000). Import of lyso-phosphatidyl-choline into chloroplasts likely at the origin of eukaryotic plastidial lipids. *Plant Physiol.* 22: 845 - 852

Nandi A, Krothapalli K, Buseman C, Li M, Welti R, Enyedi A and Shah J. (2003). The *Arabidopsis thaliana* Sfd mutants affect plastidic lipid composition and suppress dwarfing, cell death and the enhanced disease resistance phenotypes resulting from the deficiency of a fatty acid desaturase. *Plant Cell* 15 : 2383 – 2398

Nandi A, Welti R, and Shah J (2004). The *Arabidopsis thaliana* Dihydroxyacetone Phosphate Reductase Gene SUPPRESSOR OF FATTY ACID DESATURASE DEFICIENCY 1 is required for glycerolipid metabolism and for activation of systemic acquired resistance. *The Plant Cell* 16: 465 – 477

Nandi A, Moeder W, Kachroo P, Klessig DF, and Shah J (2005). *Arabidopsis* ssi2-conferred susceptibility to *Botrytis cinerea* is dependent on EDS5 and PAD4. *Mol. Plant-Microbe Interact.* 18: 363 – 370

Nawrath C (2006). Unraveling the complex network of cuticular structure and function. *Current Opin. In Plant Biology* 9: 281 – 287

Nawrath C and Metraux JP (1999). Salicylic acid induction-deficient mutants of *Arabidopsis* express PR-2 and PR-5 and accumulate high levels of camalexin after pathogen inoculation. *Plant Cell* 11: 1393 – 1404

Nimchuk Z, Rohmer L, Chang J and Dangl J (2001). Knowing the dancer from the dance: R-gene products and their interactions with other proteins from host and pathogen. *Current Opinion in Plant Biology.* 4: 288 – 294

Oostendorp M, Kunz W, Dietrich B, Staub T (2001). Induced disease resistance in plants by chemicals. *European Journal of Plant Pathology.* 107: 19 – 28

Pegadaraju P, Knepper C, Reese J and Shah J (2005). Premature leaf senescence modulated by the *Arabidopsis* PHYTOALEXIN DEFICIENT4 gene is associated with defense against the phloem-feeding green peach aphid. *Plant Physiol* 139: 1927 – 1934

Pena-Cortes H, Albrecht T, Prat S, Weller EW, and Willmitzer L (1993). Aspirin prevents wound-induced gene expression in tomato leaves by blocking jasmonic acid biosynthesis. *Planta* 191: 123 – 128

Pieterse CMJ, Van Wees SCM, Van Pelt JA, Knoester M and Laan R (1998). A novel signaling pathway controlling induced systemic resistance in *arabidopsis*. *Plant cell* 10: 1571 – 1580

Pieterse C, Ton J and Van Loon L (2001) Cross-talk between plant defense signaling pathways: boost or burden? AgBiotechNet 3

Pieterse CM and Van Loon LC (2004). NPR1: the spider in the web of induced resistance signaling pathways. *Curr. Opin. Plant Biol.* 7: 456 – 464

Ranjan R, and Lewark S (1992). Jasmonic acid promotes germination and lipase activity in nonstratified apple embryos. *Physiol. Plant.* 86: 335 – 339

Rasmussen JB, Hammerschmidt R, and Zook MN (1991). Systemic induction of salicylic acid accumulation in cucumber after inoculation with *Pseudomonas syringae*. *Plant Physiol* 97: 1342 – 1347

Ryu SB (2004). Phospholipid-derived signaling mediated by phospholipase A in plants. *Trends Plant Sci.* 9: 229 – 235

Schmelz E, Engelberth J, Tumlinson J, Block A, and Alborn H (2004). The use of vapor phase extraction in metabolic profiling of phytohormones and other metabolites. *The Plant Journal* 39: 790 – 808

Sekine K, Nandi A, Ishihara T, Hase S, Ikegami M, Shah J and Takahashi H (2004). Enhanced resistance to cucumber mosaic virus in the *Arabidopsis thaliana* ssi2 mutant is mediated via an SA-independent mechanism. *Mol. Plant-Microbe Interactions.* 17: 623 – 632

Shah J, Tsui F, and Klessig DF (1997). Characterization of a salicylic acid-insensitive mutant (sai1) of *arabidopsis thaliana* identified in a selective screen utilizing the SA inducible expression of the tms2 gene. *Mol. Plant-Microbe Interact.* 10: 69 – 78

Shah J, Kachroo P and Klessig D (1999). The *Arabidopsis* ssi1 mutation restores pathogenesis-related gene expression in npr1 plants and renders defensin gene expression salicylic acid dependent. *Plant Cell* 11: 191 – 206.

Shah J, Kachroo P, Nandi A and Klessig D (2001). A recessive mutation in the *Arabidopsis* ssi2 gene confers SA- and NPR1-independent expression of PR genes and resistance against bacterial and oomycete pathogens. *Plant J.* 25: 563 – 574

Shirano Y, Kachroo P, ShahJ and Klessig D (2002). A gain of function mutation in an *Arabidiopsis* To11 Interleukin receptor-nucleotide binding site-leucine-rich repeat type R gene triggers defense responses and results in enhanced disease resistance. *Plant Cell* 14: 3149 – 3162

Spoel S, Koornneef A, Claessens S, Korzelius J, Van Pelt J, Mueller M, Buchala A, Metraux JP, Brown R, Kazan K, Van Loon LC, Dong X and Pieterse C (2003). NPR1 modulates cross-talk between salicylate-and jasmonate-dependent pathways through a novel function in the cytosol. *The Plant Cell* 15: 760 – 770

Straus D and Glass C (2001). Cyclopentenone prostaglandins: new insights on biological activities and cellular targets. *Medicinal Research Reviews* 21: 185 – 210

Somerville C, Browne J, Jaworski JG and Ohrologge JB (2000). Lipids. In *Biochemistry and Molecular Biology of Plants*. B Buchanan, W Gruissem and R Jones eds (Rockville, MD: American Society of Plant Biologists) pp 456 – 527

Tang X, Frederick RD, Zhou J, Halterman DA, Jia Y, and Martin GB (1996). Initiation of plant disease resistance by physical interaction of AvrPto and the Pto kinase. *Science* 274: 2060 – 2063

Thomma B, Eggermont K, Pennincky I, Mauch-Mani B, Vogelsang R, Cammue B and Broekaert W (1998). Separate jasmonate-dependent and salicylate-dependent defense pathways in *Arabidopsis* are essential for resistance to distinct microbial pathogens. *Proc Natl Acad Sci* 95: 15107 – 15111

Thomma B, Eggermont K, Tierens K and Broekaert W (1999). Requirement of functional Ethylene-insensitive2 gene for efficient resistance of *Arabidopsis* to infection by *botrytis cinerea*. *Plant Physiol* 121:1093 – 1101

De Torres M, Fernandez-Delmond I, Niittyla T, Sanchez P and Grant M (2002). Differential expression of genes encoding Arabidopsis phospholipases after challenge with virulent or avirulent pseudomonas isolates. *MPMI* 15: 808 – 816.

Uknes S, Mauch-Mani B, Moyer M, Potter S, Williams S, Dincher S, Chandler D, Slusarenko A, Ward E, and Ryals J (1992). Acquired resistance in arabidopsis. *Plant cell* 4: 645 – 656

Van der Luit A, Piatti T, van Doorn A, Musgrave A, Felix G, Boller T and Mumik T (2000). Elicitation of suspension-cultured tomato cells triggers the formation of phosphatidic acid and diacylglycerol phosphate. *Plant Physiol.* 123: 1507 - 1516

Van Loon LC, Bakker PA, and Pieterse Cm (1998). Systemic resistance induced by rhizosphere bacteria. *Annu Rev Phytopathol* 36: 453 – 483

Van Wees S, Swart E, Van Pelt J, Van Loon L and Pieterse C (2000). Enhancement of induced disease resistance by simultaneous activation of salicylate- and jasmonate-dependent defense pathways in *Arabidopsis thaliana*. *Proc Natl Acad Sci* 97: 8711 - 8716

Viehweger K, Dordschbal B, and Roos W (2002). Elicitor-activated phospholipase A₂ generates lysophosphatidylcholines that mobilize the vacuolar H⁺ pool for pH signaling via the activation of Na⁺ dependent proton fluxes. *Plant Cell* 14: 1509 – 1525

Welti R, Li W, Li M, Sang Y, Biesiada H, Zhou H-E, Rajashekhar CB, Williams TD, and Wang X (2002). Profiling membrane lipids in plant stress response. Role of Phospholipase D alpha in freezing-induced lipid changes in *Arabidopsis*. *J. Biol.Chem.* 277: 31994 – 32002

Wildermuth MC, Dewdney J, Wu G and Ausubel FM (2001). Isochorismate synthase is required to synthesize salicylic acid for plant defense. *Nature* 414: 562 – 565

Yamaguchi T, Minami E and Shibuya N (2003). Activation of phospholipases by N-acetylchitooligosaccharide elicitor in suspension-cultured rice cells mediates reactive oxygen generation. *Physiol. Plant* 118: 361 – 370

Zhang W, Wang , Qin C, Wood T, and Olafsdotir G (2003). The oletae-stimulated phospholipase D, PLD δ and phosphatidic acid decrease H₂O₂-induced cell death in *Arabidopsis*. *Plant Cell* 15: 2285 – 2295

APPENDIX

TABLE A – 1 OXYLIPN PROFILE GC/MS EXPERIMENT 1

		O hr	12 hr mock	12 hr SA	24 hr mock	24 hr SA		
	avg	stdev	avg	stdev	avg	stdev	avg	stdev
JA	Col-0	719.13	140.83	724.77	136.00	643.03	52.28	947.58
	101919	1146.91	118.79	1340.31	231.56	930.84	131.72	1246.32
Benzoinic a.	082589	1495.51	149.79	1671.15	182.97	1341.79	161.08	1900.19
	Col-0	795.41	117.31	838.89	77.43	826.84	130.11	1428.18
OPDA	101919	1635.70	245.91	1710.79	232.62	1321.16	202.11	1628.71
	082589	1902.25	320.16	2068.42	345.78	1569.44	355.96	2847.18
SA	Col-0	43.85	8.11	49.42	7.65	54.43	9.29	44.97
	101919	50.22	9.22	51.36	13.61	49.75	9.19	62.95
OPDA	082589	55.98	28.71	6.54	6.54	73.29	12.31	81.67
	Col-0	983.26	109.88	2095.06	461.91	44932.82	3644.43	2436.65
SA	101919	907.30	127.89	2226.25	323.19	40873.01	2714.82	2865.29
	082589	1146.82	135.74	3133.32	586.43	54179.33	13533.5	2328.79
16:0	Col-0	93513.10	6368.75	81380.01	13018.15	94325.21	5802.52	123434.75
	101919	177304.01	39331.68	164181.74	23399.40	133487.63	20512.45	171334.67
18:3	082589	214674.26	40491.65	178126.31	18726.71	244592.87	55535.67	300712.98
	Col-0	1171.09	166.84	3645.09	1894.91	2290.06	988.72	4046.94
18:2	101919	2217.03	902.12	3190.67	1687.12	1682.19	264.28	1350.37
	082589	2132.25	503.12	3056.44	1718.73	3471.41	1745.92	2732.51
18:0	Col-0	1189.72	167.64	1677.97	125.53	1333.53	73.18	1563.04
	101919	1890.90	366.37	2332.13	466.48	1773.94	30618	1886.71
18:1	082589	1990.76	333.07	2250.32	435.41	3000.24	755.53	2681.77
	Col-0	2491.06	612.01	3721.56	287.69	2727.39	501.99	2845.52
18:0	101919	2867.35	333.93	5822.71	1130.03	3683.55	792.69	2490.22
	082589	3396.35	593.67	6260.39	1245.07	6925.41	1504.68	4436.65

TABLE A – 2 OXYLIPN PROFILE GC/MS EXPERIMENT 2

	0 hr		12 hr mock		12 hr SA		24 hr mock		24 hr SA		
	avg	stdev	avg	stdev	avg	stdev	avg	stdev	avg	stdev	
JA	Col-0	838.31	61.41	997.18	103.18	838.84	155.21	1003.97	104.79	1007.75	74.29
	101919	1200.05	205.48	1095.76	144.23	1172.77	146.78	846.79	182.53	1343.69	110.33
Benzoic a.	082589	1712.56	326.38	2062.05	124.29	1997.25	252.19	1772.57	298.88	1750.93	177.16
	Col-0	1138.07	131.14	990.97	105.17	717.13	152.21	1113.76	165.81	1150.93	150.94
OPDA	101919	1653.98	293.37	1135.34	225.08	1284.90	192.17	1053.06	129.95	1299.18	348.28
	082589	2143.39	218.81	2460.48	392.04	1822.03	574.44	1740.63	398.62	1738.17	229.73
OPDA	Col-0	76.67	12.03	86.32	15.38	88.72	17.72	86.06	29.63	80.42	16.79
	101919	58.88	23.17	99.91	23.28	77.78	11.37	124.95	22.11	85.03	12.45
SA	082589	59.49	17.54	124.61	22.12	104.99	19.32	80.42	16.79	113.39	9.22
	Col-0	666.18	110.29	2312.74	1225.15	51998.43	3549.98	2123.15	579.34	33833.08	5744.77
SA	101919	987.75	144.03	2733.58	1169.14	50869.52	3406.81	3144.69	1272.22	31138.32	3962.53
	082589	1255.52	107.69	3688.92	604.33	55265.68	4604.63	1984.19	144.48	25341.59	3878.59
16:0	Col-0	126830.46	5380.63	120444.61	12871.47	111499.79	18587.38	150090.78	18272.15	138486.43	20193.38
	101919	172215.07	27523.99	158558.84	27172.60	156490.50	24054.62	155030.13	23045.37	216800.87	31550.53
18:3	082589	266925.82	31072.37	32574.62	37211.62	218636.44	31539.07	307765.02	23681.54	261449.43	36966.61
	Col-0	1445.88	113.74	1601.68	244.76	1180.51	108.11	3510.62	1037.67	1537.54	202.57
18:2	082589	7820.08	5884.16	2238.46	191.20	1545.89	164.10	4553.57	2223.89	1674.27	291.05
	Col-0	1872.27	216.66	2112.64	611.39	1400.65	264.63	2630.86	551.15	2026.07	281.51
18:1	101919	2037.77	265.35	1629.68	290.17	1405.10	314.52	2068.32	566.23	2954.89	619.09
	082589	3339.39	1285.48	2861.63	139.18	2015.52	472.58	3402.41	660.91	2506.49	594.17
18:0	Col-0	4172.21	612.01	3669.85	944.62	2461.88	771.11	5524.44	958.68	4798.64	708.13
	101919	6204.85	1577.27	2043.06	252.06	3137.24	889.37	6309.91	1585.87	7783.95	1480.58
18:0	082589	7691.49	3420.46	6439.38	1764.37	7842.13	1550.39	9882.79	2118.87	6039.38	1473.28
	Col-0	58322.51	8398.27	69700.26	4697.21	63018.72	12678.32	95168.23	12278.59	88548.07	5272.18
	101919	102104.85	12219.02	79018.24	11427.31	89457.98	18645.33	103529.53	21620.33	149835.09	23733.86
	082589	160501.43	21652.07	150495.84	12720.84	126172.94	19707.39	191494.99	9634.68	151808.96	17877.21

TABLE A - 3 OXYLIPN PROFILE GC/MS EXPERIMENT 3

	0 hr		12 hr mock		12 hr SA		24 hr mock		24 hr SA		
	avg	stdev	avg	stdev	avg	stdev	avg	stdev	avg	Stdev	
JA	Col-0	813.51	98.16	980.09	148.53	827.92	64.50	769.84	142.22	946.73	75.95
	101919	1205.01	196.12	1541.47	325.04	1116.71	165.54	1433.85	95.65	1234.26	187.90
Benzoinic a.	082589	1832.30	109.42	1588.05	87.95	2225.94	159.37	1830.65	88.83	1921.24	164.28
	Col-0	674.33	107.69	935.90	226.40	752.78	153.54	910.18	57.06	943.86	108.00
OPDA	101919	1001.76	238.45	1357.64	103.18	991.54	149.76	1683.42	177.35	1278.92	210.05
	082589	1987.42	216.58	1494.53	107.43	2454.61	289.86	1403.69	243.69	1811.21	219.84
SA	Col-0	55.66	22.56	112.50	59.15	50.12	13.39	131.90	28.29	158.84	44.51
	101919	56.19	17.89	97.64	15.31	79.85	7.67	126.38	40.61	135.55	29.29
16:0	082589	101.88	41.23	142.23	19.78	124.47	57.52	174.92	84.31	130.23	45.67
	Col-0	671.48	84.99	1723.20	314.01	46746.69	8466.29	2326.93	667.84	37763.30	6915.80
18:3	101919	1053.59	211.32	3766.05	1014.49	26289.86	113.37	3566.63	1079.08	23567.34	3855.78
	082589	1822.65	466.93	2159.19	487.97	28558.84	4301.15	2294.62	576.45	17971.75	1476.55
Col-0	120963.46	19924.17	168305.88	45452.62	167856.24	18977.67	114042.26	23865.75	119404.50	19290.84	
	101919	158995.99	25240.11	259467.30	56646.05	266795.59	49056.43	255174.88	33808.10	214208.64	34549.13
Col-0	082589	330286.05	58457.41	306057.64	74734.32	397565.91	74822.20	269711.98	39466.39	346468.59	46246.72
	1049.41	198.94	1388.02	314.34	1165.31	129.19	2147.13	260.19	2209.79	438.79	
Col-0	101919	1611.45	178.48	2717.48	841.98	2442.09	503.48	1788.03	249.96	1963.09	310.43
	082589	3625.57	1430.82	4055.03	1735.69	2217.09	398.42	2259.45	330.18	2823.13	440.30
Col-0	1230.81	244.58	1755.26	497.74	1505.52	248.58	1696.87	288.17	1954.99	290.38	
	101919	2224.99	279.74	2981.88	467.49	2786.46	485.77	2737.57	350.39	2856.01	711.25
Col-0	082589	4306.31	700.39	3778.99	594.03	4300.28	870.11	3795.12	671.20	4378.84	791.08
	2906.49	571.88	4354.83	1255.84	3548.37	542.26	4147.24	788.39	4204.81	708.13	
Col-0	101919	5404.14	579.44	6909.81	1006.99	7198.01	1305.94	6998.23	908.25	6468.32	1480.58
	082589	9603.68	1492.82	9237.05	1293.42	10376.39	1918.52	9212.33	1383.11	11109.34	1567.79
Col-0	58332.51	8398.27	109508.72	34933.36	91682.21	11244.78	63072.24	12237.72	71401.89	7123.85	
	101919	103488.85	20631.02	143995.86	24650.05	157932.59	34174.09	144443.11	27954.18	150150.44	27216.56
Col-0	082589	184156.43	25959.12	172408.37	31886.13	211529.52	30062.74	172866.76	30836.75	215782.48	37962.18

TABLE A – 4. TOTAL LIPID SPECIES – ESI MS/MS EXPERIMENT 1

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	Stdev
DGDG 34:6	1.118986	0.097751	1.293635	0.144785	1.072353	0.140867
DGDG 34:5	0.144561	0.011412	0.185189	0.032773	0.191758	0.022152
DGDG 34:4	0.09946	0.010783	0.112762	0.014145	0.110665	0.013065
DGDG 34:3	3.288272	0.14073	2.964665	0.280843	2.445199	0.130912
DGDG 34:2	0.543981	0.043538	0.470316	0.05718	0.414086	0.027578
DGDG 34:1	0.12521	0.010462	0.146124	0.025677	0.158668	0.021132
DGDG 36:6	11.08261	0.486922	9.983793	1.311141	7.859936	0.475655
DGDG 36:5	1.04882	0.138589	0.701567	0.206355	0.443833	0.093063
DGDG 36:4	0.263044	0.021717	0.205583	0.028903	0.158962	0.018437
DGDG 36:3	0.158728	0.020271	0.117583	0.017275	0.118364	0.008697
DGDG 36:2	0.013319	0.002258	0.008312	0.000896	0.011764	0.002225
DGDG 36:1	0.000569	0.000574	0.001069	0.000676	0.002144	0.001994
DGDG 38:6	0.03657	0.007513	0.0252	0.007321	0.026102	0.007484
DGDG 38:5	0.006679	0.001456	0.004629	0.00211	0.006597	0.002334
DGDG 38:4	0.003804	0.001274	0.002964	0.001211	0.002307	0.001008
DGDG 38:3	0.001988	0.000803	0.000684	0.000696	0.000609	0.000878
Total DGDG	17.9366	0.802799	16.22408	1.947617	13.02335	0.776117
Col-o 12hr mock 101919 12hr mock 082589 12hr mock						
Sample description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.212806	0.104136	1.154005	0.104717	1.056774	0.068704
DGDG 34:5	0.13655	0.012958	0.175472	0.010528	0.201108	0.013308
DGDG 34:4	0.112282	0.018266	0.100932	0.014291	0.114309	0.021127
DGDG 34:3	3.342314	0.198073	2.753837	0.229692	2.407396	0.153095
DGDG 34:2	0.579049	0.066071	0.422524	0.051776	0.411473	0.014126
DGDG 34:1	0.124836	0.018143	0.127626	0.00917	0.140878	0.023427
DGDG 36:6	11.21324	0.408567	9.250201	0.834972	8.095682	0.353684
DGDG 36:5	1.037081	0.117565	0.548677	0.203999	0.507756	0.085972
DGDG 36:4	0.259481	0.027613	0.175586	0.046023	0.155358	0.022567
DGDG 36:3	0.141884	0.012782	0.114037	0.004706	0.119688	0.013705
DGDG 36:2	0.01475	0.001382	0.010112	0.002248	0.011105	0.002241
DGDG 36:1	0.00119	0.000493	0.000499	0.000883	0.001601	0.001274
DGDG 38:6	0.030148	0.004124	0.034514	0.008439	0.031362	0.008192
DGDG 38:5	0.006935	0.00113	0.006164	0.001663	0.004708	0.002019
DGDG 38:4	0.002967	0.00108	0.003397	0.001527	0.002488	0.001207
DGDG 38:3	0.001506	0.001333	0.000904	0.000671	0.000701	0.000329
Total DGDG	18.21702	0.715264	14.87849	1.369785	13.26239	0.614403

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.158199	0.12777	1.083433	0.095425	1.115775	0.056968
DGDG 34:5	0.14281	0.006334	0.147361	0.01526	0.202562	0.027258
DGDG 34:4	0.105528	0.003506	0.099458	0.006064	0.12256	0.01015
DGDG 34:3	3.367974	0.283955	2.882852	0.154526	2.423552	0.092945
DGDG 34:2	0.573776	0.103326	0.457778	0.027614	0.401704	0.013212
DGDG 34:1	0.124078	0.023491	0.118825	0.011528	0.165464	0.017978
DGDG 36:6	11.53445	0.332059	10.40288	0.311931	8.411293	0.486999
DGDG 36:5	1.067372	0.181282	0.745857	0.079831	0.542326	0.077985
DGDG 36:4	0.263352	0.038105	0.211707	0.014388	0.164516	0.013468
DGDG 36:3	0.144839	0.014522	0.133584	0.010819	0.117802	0.008007
DGDG 36:2	0.011679	0.005208	0.007079	0.005311	0.010611	0.004705
DGDG 36:1	0.000812	0.000691	0.001253	0.00095	0.002648	0.002401
DGDG 38:6	0.027294	0.00913	0.024664	0.004084	0.042366	0.007559
DGDG 38:5	0.007393	0.002053	0.005867	0.001932	0.005145	0.002368
DGDG 38:4	0.003303	0.000912	0.002639	0.000921	0.003181	0.00118
DGDG 38:3	0.002621	0.000495	0.00083	0.000632	0.000372	0.000436
Total DGDG	18.53548	0.70415	16.32607	0.326716	13.73187	0.586329
	Col-o 0hr		101919 0hr		082589 0hr	
Sample Description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	56.12877	1.232612	58.49895	2.255271	60.98628	1.070131
MGDG 34:5	1.555372	0.16533	2.0851	0.449177	2.80248	0.345369
MGDG 34:4	0.553516	0.050413	0.854309	0.236103	1.241416	0.162654
MGDG 34:3	0.497537	0.050683	0.486615	0.0722	0.582076	0.074337
MGDG 34:2	0.0833	0.014656	0.120358	0.031369	0.184519	0.039615
MGDG 34:1	0.045071	0.005016	0.064737	0.017406	0.105994	0.023204
MGDG 36:6	6.965487	0.581473	6.743837	0.416002	7.80593	0.669421
MGDG 36:5	1.036693	0.082159	0.77851	0.112708	0.725166	0.055159
MGDG 36:4	0.134737	0.009649	0.123761	0.017719	0.15405	0.011939
MGDG 36:3	0.017782	0.002373	0.01682	0.004621	0.020804	0.003803
MGDG 36:2	0.001298	0.00085	0.000563	0.000725	0.002209	0.00253
MGDG 36:1	0	0	0.000413	0.000924	0.000851	0.001213
MGDG 38:6	0.000954	0.001009	0.000505	0.000748	0.010866	0.006136
MGDG 38:5	0.000929	0.000649	0.002537	0.001197	0.005776	0.002119
MGDG 38:4	0.002623	0.001418	0.003203	0.000737	0.002864	0.000964
MGDG 38:3	0	0	0	0	0	0
Total MGDG	67.02407	1.222231	69.78022	2.45675	74.63129	1.318224

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample	ave	stdev	ave	stdev	ave	stdev
Description						
MGDG 34:6	56.24743	0.628992	61.30408	0.458956	62.38089	1.621201
MGDG 34:5	1.249615	0.110325	1.862026	0.453232	2.474826	0.416647
MGDG 34:4	0.556065	0.022596	0.76226	0.099258	0.974937	0.11047
MGDG 34:3	0.448383	0.023228	0.444426	0.018525	0.469423	0.031062
MGDG 34:2	0.061056	0.007066	0.080373	0.010865	0.122892	0.018973
MGDG 34:1	0.031811	0.010269	0.033962	0.012178	0.034394	0.012024
MGDG 36:6	7.182576	0.5819	7.391557	0.828374	7.84034	0.626822
MGDG 36:5	0.958778	0.095242	0.686717	0.079668	0.687845	0.076807
MGDG 36:4	0.1337	0.009714	0.120975	0.018442	0.131232	0.009427
MGDG 36:3	0.016484	0.004252	0.018404	0.006084	0.020302	0.005142
MGDG 36:2	0.001225	0.001023	0.001007	0.000708	0.001397	0.001313
MGDG 36:1	0.000704	0.001575	0.000821	0.000967	0	0
MGDG 38:6	0.003536	0.00216	0.007398	0.002696	0.006685	0.002257
MGDG 38:5	0.001751	0.000682	0.002215	0.00097	0.002875	0.003247
MGDG 38:4	0.002712	0.000446	0.002828	0.000848	0.002031	0.001308
MGDG 38:3	0.00019	0.000296	0.000309	0.000424	0.00101	0.001301
Total MGDG	66.89602	0.690427	72.71936	1.732191	75.15108	1.053299

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample	ave	stdev	ave	stdev	ave	stdev
Description						
MGDG 34:6	56.85268	2.393138	58.79515	1.594144	61.77192	1.557905
MGDG 34:5	1.271775	0.205362	1.507837	0.11466	2.329936	0.294624
MGDG 34:4	0.503673	0.046054	0.615691	0.049802	0.965481	0.143031
MGDG 34:3	0.465008	0.025436	0.462087	0.066349	0.463201	0.038999
MGDG 34:2	0.059774	0.006882	0.068009	0.012952	0.126546	0.029513
MGDG 34:1	0.029064	0.01035	0.045461	0.009612	0.053435	0.01586
MGDG 36:6	7.185331	1.024595	7.550969	0.64761	7.914733	0.719188
MGDG 36:5	0.917106	0.056052	0.821205	0.030375	0.674437	0.078029
MGDG 36:4	0.119949	0.008561	0.124488	0.012103	0.127429	0.008637
MGDG 36:3	0.015483	0.001965	0.014135	0.002295	0.018277	0.004729
MGDG 36:2	0.001387	0.000719	0.001372	0.001125	0.001435	0.00186
MGDG 36:1	0.001353	0.00188	0.000146	0.000326	0.000337	0.000754
MGDG 38:6	0.004419	0.003867	0.003106	0.001989	0.008564	0.004023
MGDG 38:5	0.005012	0.001013	0.004112	0.001497	0.004089	0.002637
MGDG 38:4	0.002372	0.000766	0.003281	0.001213	0.003456	0.002294
MGDG 38:3	0.000616	0.000725	0.000586	0.000428	0.000138	0.000309
Total MGDG	67.435	2.841891	70.01763	1.109256	74.46342	1.064047

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample	ave	stdev	ave	stdev	ave	stdev
Description						
MGDG 34:6	57.27557	1.775308	60.71155	1.061814	62.57051	1.51361
MGDG 34:5	1.384116	0.200233	2.090479	0.269479	2.002682	0.3808
MGDG 34:4	0.587975	0.053244	0.93768	0.117236	1.107102	0.205606
MGDG 34:3	0.433905	0.060709	0.482081	0.017991	0.529861	0.050053
MGDG 34:2	0.062748	0.004687	0.129389	0.01741	0.157015	0.038255
MGDG 34:1	0.037859	0.007088	0.075006	0.007513	0.080447	0.014952
MGDG 36:6	6.397364	0.816078	7.363661	0.340596	7.643226	0.382319
MGDG 36:5	0.929611	0.099172	0.671812	0.066317	0.554314	0.059598
MGDG 36:4	0.105777	0.017088	0.120707	0.006812	0.120396	0.00759
MGDG 36:3	0.013661	0.003959	0.017846	0.003987	0.017178	0.0039
MGDG 36:2	0.000265	0.000258	0.000939	0.001325	0.001647	0.001331
MGDG 36:1	0	0	0.000634	0.001418	0.001535	0.002174
MGDG 38:6	0.003517	0.001779	0.007202	0.001865	0.009712	0.00479
MGDG 38:5	0.001769	0.001159	0.002242	0.001156	0.005348	0.002305
MGDG 38:4	0.001951	0.001342	0.002204	0.000544	0.002311	0.00156
MGDG 38:3	0.000255	0.000352	0.00039	0.0004	0.000454	0.000623
Total MGDG	67.23635	1.220306	72.61382	0.86832	74.80374	1.894731

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample	ave	stdev	ave	stdev	ave	stdev
Description						
MGDG 34:6	57.40319	1.588423	58.71637	1.21904	60.24158	2.058066
MGDG 34:5	1.058654	0.224778	1.709905	0.408776	2.24511	0.376049
MGDG 34:4	0.508887	0.079923	0.876448	0.247336	1.076926	0.089372
MGDG 34:3	0.44093	0.021555	0.50375	0.055279	0.478269	0.013119
MGDG 34:2	0.072776	0.003943	0.130852	0.025603	0.145726	0.020061
MGDG 34:1	0.05896	0.02564	0.053458	0.014141	0.059119	0.00597
MGDG 36:6	6.235798	0.982859	7.314797	0.567148	7.054748	0.406327
MGDG 36:5	0.84505	0.069456	0.63518	0.11357	0.5739	0.13247
MGDG 36:4	0.100815	0.012178	0.122103	0.007371	0.1308	0.017994
MGDG 36:3	0.014916	0.001402	0.017235	0.004653	0.020916	0.00231
MGDG 36:2	0.000464	0.000637	1.98E-05	4.43E-05	0.000474	0.000564
MGDG 36:1	0	0	0	0	0.000224	0.000502
MGDG 38:6	0.003381	0.005903	0.005157	0.003835	0.011407	0.004297
MGDG 38:5	0.001382	0.000904	0.00052	0.000723	0.004691	0.001358
MGDG 38:4	0.002419	0.000655	0.003469	0.000516	0.001849	0.001712
MGDG 38:3	0.000108	0.000242	0	0	0.000119	0.000242
Total MGDG	66.74773	2.265487	70.08927	1.771611	72.04586	2.441568

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.188077	0.041372	0.136352	0.046753	0.036043	0.080595
PG 32:0	0.061421	0.035365	0.006855	0.009608	0	0
PG 34:4	2.09408	0.303812	2.780647	0.194953	2.189814	0.212195
PG 34:3	1.048556	0.222821	0.94033	0.217082	0.703649	0.342817
PG 34:2	0.415891	0.104033	0.556422	0.092138	0.385484	0.068876
PG 34:1	0.110125	0.067781	0.247128	0.035491	0.234466	0.114905
PG 34:0	0	0	0	0	0	0
Total PG	3.91815	0.494653	4.667734	0.195449	3.549457	0.713361

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.089446	0.081585	0.071191	0.032602	0.069633	0.067876
PG 32:0	0.014345	0.032077	0.030942	0.017759	0.003552	0.00349
PG 34:4	2.628955	0.578132	2.293304	0.40735	2.257046	0.233357
PG 34:3	1.155823	0.21511	0.786585	0.169329	0.972106	0.107126
PG 34:2	0.427267	0.159772	0.395906	0.158782	0.512774	0.078285
PG 34:1	0.092923	0.081057	0.112765	0.046688	0.086048	0.068021
PG 34:0	0	0	0.002614	0.005845	0.004074	0.00911
Total PG	4.40876	0.955083	3.693307	0.560219	3.905232	0.326377

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.161272	0.057967	0.080828	0.052891	0.093658	0.045096
PG 32:0	0.043096	0.015415	0.037782	0.02931	0.016978	0.010443
PG 34:4	2.112487	0.352686	2.511549	0.710817	2.219946	0.673647
PG 34:3	1.157981	0.413598	0.910968	0.188918	0.738428	0.166154
PG 34:2	0.445502	0.125911	0.431453	0.210851	0.453183	0.141701
PG 34:1	0.130712	0.055798	0.045101	0.060597	0.153923	0.142266
PG 34:0	0	0	0.000468	0.001046	0	0
Total PG	4.05105	0.759623	4.018149	0.960937	3.676116	0.898832

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.122828	0.060958	0.085156	0.022447	0.05749	0.024619
PG 32:0	0.010785	0.010782	0.033266	0.033689	0.013942	0.009161
PG 34:4	2.263513	0.41129	2.341609	0.277339	1.880132	0.4368
PG 34:3	1.174357	0.495304	0.794958	0.047241	0.618783	0.189438
PG 34:2	0.3551	0.102073	0.419655	0.0204	0.457019	0.075478
PG 34:1	0.088031	0.077832	0.183242	0.086585	0.1779	0.067213

PG 34:0	0	0	0	0	0	0	0
Total PG	4.014614	0.812742	3.857885	0.279396	3.205267	0.678015	
Col-o 24hr SA 101919 24hr SA 082589 24hr SA							
Sample description	ave	stdev	ave	stdev	ave	stdev	
PG 32:1	0.244777	0.035627	0.192576	0.044967	0.148174	0.022963	
PG 32:0	0.094655	0.032834	0.079056	0.032	0.046125	0.021049	
PG 34:4	1.97802	0.328034	2.072697	0.211387	2.0059	0.155991	
PG 34:3	1.007592	0.225266	0.800522	0.15481	0.807077	0.177854	
PG 34:2	0.349885	0.084209	0.393821	0.064596	0.450281	0.036868	
PG 34:1	0.164323	0.055527	0.227333	0.100182	0.233344	0.074833	
PG 34:0	0.003884	0.005014	0.000301	0.000673	0	0	
Total PG	3.843134	0.567033	3.766305	0.43358	3.690901	0.195352	

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0	0	0	0	0.001994	0.002779
lysoPG 16:0	0.000855	0.000911	0	0	0	0
lysoPG 18:3	0.001858	0.002545	0	0	0	0
lysoPG 18:2	0.001118	0.001589	0	0	0	0
lysoPG 18:1	0	0	0	0	0	0
Total lysoPG	0.003831	0.004154	0	0	0.001994	0.002779

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.002231	0.00499	0	0	0	0
lysoPG 16:0	0	0	0	0	0	0
lysoPG 18:3	0	0	0.000775	0.000787	0	0
lysoPG 18:2	0	0	0	0	0	0
lysoPG 18:1	0	0	0.000798	0.0014	0	0
Total lysoPG	0.002231	0.00499	0.001572	0.002081	0	0

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0	0	0	0	0	0
lysoPG 16:0	0.000703	0.000616	0.000411	0.00048	0.000932	0.001299
lysoPG 18:3	0.001205	0.00171	0.001073	0.001079	0.001009	0.001144
lysoPG 18:2	0.00027	0.000401	0	0	0.000238	0.000289
lysoPG 18:1	0.000166	0.000372	0.001566	0.003501	0.000845	0.001348
Total lysoPG	0.002345	0.002179	0.00305	0.004337	0.003024	0.002579

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000729	0.00163	0.000449	0.000755	0.000284	0.00042
lysoPG 16:0	0.000387	0.000742	0.00023	0.000515	0.000185	0.000415
lysoPG 18:3	0.000449	0.00077	0.000971	0.00165	0.001522	0.000743
lysoPG 18:2	0.000283	0.000634	0.000224	0.000447	0.000248	0.000555
lysoPG 18:1	0.000144	0.000322	0.000475	0.001062	0	0
Total lysoPG	0.001992	0.001672	0.002349	0.002433	0.00224	0.001452

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000366	0.000764	0	0	0.000541	0.001209
lysoPG 16:0	0.000717	0.001524	0.001089	0.002104	0.003881	0.003197
lysoPG 18:3	0.013278	0.003923	0.006722	0.006175	0.009634	0.002885
lysoPG 18:2	0.002263	0.002878	0.00042	0.00094	0.000651	0.001456
lysoPG 18:1	0	0	0	0	5.59E-05	0.000125
Total lysoPG	0.016624	0.007015	0.008231	0.006973	0.014763	0.004909

	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0	0	0	0	0	0
LysoPC 16:0	0.001983	0.00133	0.001597	0.001119	0.000396	0.000646
LysoPC 18:3	0.001852	0.000906	0.002036	0.001027	0.000319	0.00044
LysoPC 18:2	0.002082	0.000697	0.002551	0.00082	0.000909	0.000835
LysoPC 18:1	4.23E-05	9.46E-05	0.000207	0.000336	0	0
LysoPC 18:0	0	0	0.000308	0.000426	0.000655	0.000563
Total LysoPC	0.005959	0.00264	0.006698	0.002873	0.002278	0.001481

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	1.15E-05	2.57E-05	1.99E-05	4.44E-05	0	0
LysoPC 16:0	0.001233	0.000974	0.001073	0.000758	0.00068	0.000668
LysoPC 18:3	0.002737	0.000806	0.001483	0.000844	0.000856	0.000638
LysoPC 18:2	0.002407	0.000955	0.001306	0.000585	0.001085	0.001186
LysoPC 18:1	0	0	4.04E-05	5.12E-05	1.24E-05	1.85E-05
LysoPC 18:0	0.000176	0.000242	0	0	9.59E-06	2.14E-05
Total LysoPC	0.006553	0.001926	0.003903	0.001917	0.002643	0.002

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0	0	5.69E-06	1.27E-05	0	0
LysoPC 16:0	0.001353	0.001174	0.001502	0.001265	0.000947	0.001383
LysoPC 18:3	0.001691	0.00114	0.002907	0.001527	0.001096	0.000579
LysoPC 18:2	0.002121	0.00131	0.0033	0.001713	0.001962	0.0005
LysoPC 18:1	8.72E-05	0.000155	8.11E-06	1.44E-05	0	0
LysoPC 18:0	0.000222	0.000496	0.000361	0.00055	9.29E-05	0.000208
Total LysoPC	0.005485	0.003186	0.008099	0.004113	0.004098	0.001901

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0	0	0.001788	0.003998	0.000458	0.001024
LysoPC 16:0	0.001519	0.001084	0.000826	0.000705	0.001119	0.001053
LysoPC 18:3	0.003311	0.001162	0.001585	0.000623	0.001129	0.000475
LysoPC 18:2	0.001506	0.000536	0.001622	0.000676	0.001395	0.000556
LysoPC 18:1	1.04E-05	2.33E-05	6.27E-05	8.6E-05	4.96E-05	6.79E-05
LysoPC 18:0	0	0	0	0	0.000105	0.000229
Total LysoPC	0.006347	0.002459	0.004102	0.001885	0.003798	0.001183

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1						
LysoPC 16:0	0.004514	0.005391	0.006926	0.00858	0.007889	0.012344
LysoPC 18:3	0.005103	0.003214	0.004332	0.00586	0.001139	0.001562
LysoPC 18:2	0.004014	0.003775	0.002491	0.002345	0.006067	0.003045
LysoPC 18:1	0.002038	0.001899	0	0	0.000995	0.001904
LysoPC 18:0	0.002066	0.002204	0.005717	0.006509	0.001457	0.003257
Total LysoPC	0.017736	0.009819	0.021254	0.013631	0.018005	0.013937

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0	0	9.77E-05	0.000142	0	0
LysoPE 16:0	0.004293	0.002011	0.003639	0.001485	0.001431	0.001204
LysoPE 18:3	0.001866	0.001181	0.002344	0.001159	0.000374	0.000412
LysoPE 18:2	0.002258	0.000631	0.003595	0.001377	0.00205	0.000877
LysoPE 18:1	3.48E-05	7.79E-05	8.82E-05	0.000112	0	0
Total LysoPE	0.008452	0.003252	0.009764	0.003077	0.003855	0.001328

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0	0	5.69E-05	0.000127	1.27E-05	2.83E-05
LysoPE 16:0	0.005392	0.000804	0.003292	0.00211	0.00319	0.001797
LysoPE 18:3	0.002579	0.001721	0.001292	0.000845	0.000899	0.000708
LysoPE 18:2	0.0026	0.001525	0.002188	0.001451	0.001643	0.001255
LysoPE 18:1	5.81E-05	0.00013	4.57E-05	0.000102	1.72E-05	3.85E-05
Total LysoPE	0.01063	0.003064	0.006875	0.004077	0.005763	0.002995

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	6.79E-05	8.69E-05	0	0	0	0
LysoPE 16:0	0.006553	0.002203	0.003675	0.003962	0.00533	0.00306
LysoPE 18:3	0.002973	0.000682	0.001053	0.001103	0.001506	0.001061
LysoPE 18:2	0.002864	0.000968	0.001471	0.001078	0.003155	0.000941
LysoPE 18:1	9.35E-05	0.000119	3.24E-06	7.25E-06	0	0
Total LysoPE	0.012551	0.001765	0.006203	0.005673	0.009991	0.0045

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	4.55E-05	6.35E-05	5.96E-05	8.33E-05	1.44E-05	3.21E-05
LysoPE 16:0	0.006994	0.001594	0.004571	0.000584	0.00357	0.00137
LysoPE 18:3	0.002995	0.001617	0.002745	0.000906	0.001549	0.000581
LysoPE 18:2	0.003142	0.000767	0.003104	0.000731	0.002946	0.000892
LysoPE 18:1	0.000133	0.000235	0.000137	0.000115	2.44E-05	5.45E-05
Total LysoPE	0.01331	0.002668	0.010617	0.000877	0.008104	0.001266

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0	0	0	0	0	0
LysoPE 16:0	0.005812	0.00413	0.00404	0.00477	0.002109	0.002611
LysoPE 18:3	0.002347	0.001378	0.000763	0.001707	0.001238	0.00219
LysoPE 18:2	0.003299	0.001449	0.001728	0.001113	0.003063	0.001965
LysoPE 18:1	0.000137	0.000307	0.000194	0.000435	0	0
Total LysoPE	0.011594	0.005761	0.006725	0.005467	0.00641	0.00392

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.003358	0.0006	0.002458	0.000375	0.001963	0.000612
PC 34:4	0.028317	0.005593	0.026756	0.003601	0.037501	0.017465
PC 34:3	2.028287	0.334282	1.274244	0.247698	1.060387	0.31352
PC 34:2	0.967405	0.221468	0.735098	0.118683	0.95617	0.198286
PC 34:1	0.062054	0.044166	0.080155	0.020641	0.087988	0.027456
PC 36:6	0.97905	0.152075	0.639413	0.147521	0.487372	0.119491
PC 36:5	1.350422	0.095636	1.019168	0.114089	1.339839	0.434078
PC 36:4	0.529467	0.079177	0.660963	0.167467	0.965651	0.142871
PC 36:3	0.2354	0.043453	0.238593	0.075282	0.327366	0.111922
PC 36:2	0.058635	0.011018	0.072755	0.025847	0.122363	0.032897
PC 36:1	0	0	0.000376	0.000841	0	0
PC 38:6	0.005604	0.001401	0.004386	0.001067	0.004502	0.001154
PC 38:5	0.009984	0.001669	0.010869	0.003696	0.014999	0.005629
PC 38:4	0.013401	0.003374	0.019059	0.006322	0.02354	0.005202
PC 38:3	0.016543	0.003939	0.022913	0.003224	0.023226	0.008764
PC 38:2	0.007392	0.002485	0.010398	0.001304	0.009967	0.001517
PC 40:5	0.000578	0.000332	0.001159	0.00066	0.000442	0.00068
PC 40:4	0.000635	0.000351	0.001756	0.000166	0.001265	0.000816
PC 40:3	0.00168	0.000737	0.001859	0.000504	0.001336	0.000867
PC 40:2	0.001375	0.000488	0.00158	0.00047	0.000758	0.000552
Total PC	6.299588	0.797776	4.823957	0.706066	5.466636	1.266886
Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.003188	0.001315	0.001601	0.00044	0.000924	0.000796
PC 34:4	0.022738	0.002416	0.028019	0.002756	0.027664	0.005148
PC 34:3	1.750236	0.166407	1.228919	0.321875	0.882447	0.126929
PC 34:2	0.782211	0.116004	0.751864	0.068581	0.72112	0.108684
PC 34:1	0.047337	0.009099	0.04747	0.017287	0.035981	0.028487
PC 36:6	0.88722	0.139953	0.601533	0.179313	0.420544	0.062178
PC 36:5	1.104535	0.156438	1.040334	0.104971	1.025201	0.200635
PC 36:4	0.493607	0.070208	0.652894	0.040376	0.644794	0.154026
PC 36:3	0.172626	0.021866	0.157586	0.041137	0.17771	0.041262
PC 36:2	0.049368	0.007274	0.064371	0.017475	0.081448	0.020553
PC 36:1	0.000388	0.000533	0	0	0	0
PC 38:6	0.00509	0.000546	0.004448	0.001569	0.003644	0.000706
PC 38:5	0.008442	0.001355	0.011453	0.001643	0.010724	0.001798
PC 38:4	0.011164	0.002621	0.01555	0.002035	0.018618	0.004883
PC 38:3	0.015376	0.001533	0.016961	0.003359	0.018635	0.002858
PC 38:2	0.006978	0.001248	0.007147	0.003572	0.00733	0.002806
PC 40:5	0.000743	0.000407	0.000921	0.000501	0.000736	0.00058
PC 40:4	0.000655	0.000286	0.00141	0.000761	0.001493	0.000778
PC 40:3	0.001495	0.000372	0.001098	0.000651	0.001472	0.000716
PC 40:2	0.001273	0.000423	0.00088	0.000397	0.00089	0.000535
Total PC	5.364673	0.655117	4.63446	0.65205	4.081374	0.567886

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002778	0.001598	0.0021	0.000505	0.001321	0.000429
PC 34:4	0.019066	0.01075	0.022003	0.00374	0.031447	0.005205
PC 34:3	1.44756	0.817135	1.463334	0.073119	0.973836	0.244262
PC 34:2	0.563748	0.332765	0.685743	0.151307	0.686488	0.105204
PC 34:1	0.051998	0.03262	0.063061	0.014556	0.05429	0.015778
PC 36:6	0.715321	0.404297	0.725943	0.079306	0.51189	0.157198
PC 36:5	0.867509	0.490017	1.084208	0.102489	1.068358	0.084808
PC 36:4	0.404224	0.240771	0.58826	0.106764	0.643531	0.092544
PC 36:3	0.148088	0.083377	0.177543	0.038534	0.186135	0.019479
PC 36:2	0.035621	0.020889	0.048192	0.010813	0.071982	0.014013
PC 36:1	0.000752	0.00103	0.00115	0.001651	0.000276	0.000617
PC 38:6	0.004921	0.002756	0.004958	0.000617	0.003804	0.001085
PC 38:5	0.006589	0.003778	0.009137	0.001255	0.011309	0.001469
PC 38:4	0.012963	0.007346	0.017827	0.003461	0.018464	0.001364
PC 38:3	0.014444	0.008427	0.016868	0.001896	0.021089	0.005042
PC 38:2	0.007358	0.004593	0.008027	0.001683	0.011055	0.002054
PC 40:5	0.000596	0.00049	0.000474	0.000456	0.00104	0.000794
PC 40:4	0.000976	0.000908	0.000691	0.000351	0.001894	0.000702
PC 40:3	0.001455	0.000901	0.001271	0.000278	0.00154	0.000599
PC 40:2	0.000872	0.000619	0.001195	0.000518	0.001585	0.00046
Total PC	4.306838	2.443815	4.921986	0.368834	4.301334	0.574956
Col-o 24hr mock		101919 24hr mock		082589 24hr mock		
Sample description	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002941	0.000471	0.002405	0.000431	0.001812	0.000194
PC 34:4	0.028355	0.004906	0.026933	0.004455	0.023923	0.003313
PC 34:3	2.009629	0.227286	1.05154	0.072923	0.852674	0.135039
PC 34:2	0.743471	0.155206	0.633354	0.041297	0.617444	0.052987
PC 34:1	0.054098	0.007238	0.06988	0.011984	0.058526	0.009904
PC 36:6	1.024953	0.141363	0.555699	0.058379	0.408868	0.052232
PC 36:5	1.158726	0.107486	0.998921	0.099956	0.923416	0.064357
PC 36:4	0.520252	0.13965	0.611449	0.062026	0.56298	0.042806
PC 36:3	0.181201	0.013777	0.226002	0.043896	0.196933	0.024017
PC 36:2	0.0457	0.013928	0.071643	0.008402	0.073229	0.006812
PC 36:1	0.00057	0.00075	4.61E-05	0.000103	8.04E-05	0.000112
PC 38:6	0.006669	0.000773	0.004163	0.000753	0.003703	0.000825
PC 38:5	0.008845	0.001146	0.009115	0.000496	0.008925	0.000675
PC 38:4	0.013681	0.002725	0.016291	0.000909	0.015619	0.001167
PC 38:3	0.017083	0.002032	0.017708	0.002005	0.018826	0.00281
PC 38:2	0.007494	0.000721	0.009089	0.001119	0.009575	0.001116
PC 40:5	0.000885	0.000552	0.00095	0.000302	0.000934	0.000314
PC 40:4	0.000912	0.000501	0.001718	0.000395	0.001581	0.000499
PC 40:3	0.001858	0.000552	0.001735	0.000509	0.001615	0.000325
PC 40:2	0.001357	0.000663	0.001333	0.00023	0.000907	0.000277
Total PC	5.828679	0.609476	4.309974	0.324091	3.78157	0.257598

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.006328	0.005747	0.004138	0.003349	0.00395	0.003993
PC 34:4	0.020023	0.020489	0.024339	0.024904	0.054191	0.034222
PC 34:3	1.730389	0.552465	1.338534	1.114636	1.567207	0.639343
PC 34:2	0.708399	0.371867	0.86936	0.592133	0.814402	0.703785
PC 34:1	0.07932	0.094877	0.088718	0.073899	0.17751	0.016067
PC 36:6	0.725201	0.374569	0.817322	0.421818	0.535358	0.187485
PC 36:5	0.937627	0.592749	1.215323	0.544507	1.873797	0.517299
PC 36:4	0.33727	0.238146	0.942722	0.4112	1.082372	0.445244
PC 36:3	0.179352	0.198914	0.304694	0.177045	0.270221	0.202091
PC 36:2	0.045008	0.030875	0.112621	0.08335	0.1317	0.040296
PC 36:1	0.01098	0.012219	0.01975	0.030778	0.006042	0.008482
PC 38:6	0.006947	0.002876	0.008622	0.004138	0.008253	0.007638
PC 38:5	0.003382	0.002746	0.010456	0.005302	0.015369	0.008577
PC 38:4	0.011171	0.007345	0.022622	0.015758	0.025004	0.026647
PC 38:3	0.018817	0.015986	0.026433	0.013844	0.038596	0.019591
PC 38:2	0.004464	0.003363	0.019157	0.010651	0.015292	0.011362
PC 40:5	0.002922	0.003154	0.002053	0.003109	0.001917	0.000519
PC 40:4	0.001612	0.001702	0.001565	0.001294	0.000776	0.000842
PC 40:3	0.002071	0.001616	0.000868	0.001332	0.003283	0.00201
PC 40:2	0.001432	0.001701	0.001032	0.001335	0.002538	0.004795
Total PC	4.832713	1.325303	5.830328	2.298129	6.627778	2.296336

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.009207	0.000742	0.011642	0.001247	0.007676	0.000585
PE 34:3	0.933536	0.080395	0.737542	0.143975	0.450154	0.058833
PE 34:2	0.802602	0.047017	0.80425	0.102338	0.785609	0.093315
PE 34:1	0.014359	0.002385	0.017547	0.002782	0.016135	0.004241
PE 36:6	0.265534	0.034925	0.217191	0.054249	0.113755	0.009643
PE 36:5	0.562641	0.028816	0.562919	0.068165	0.448869	0.043227
PE 36:4	0.364559	0.022737	0.438572	0.056928	0.450424	0.047511
PE 36:3	0.084008	0.007463	0.101761	0.023773	0.086651	0.010422
PE 36:2	0.037374	0.002517	0.049836	0.015788	0.065171	0.009936
PE 36:1	0.001006	0.00099	0.001345	0.001334	0	0
PE 38:6	0.005592	0.000392	0.00526	0.001337	0.002935	0.000806
PE 38:5	0.006731	0.001286	0.007164	0.001546	0.006484	0.001124
PE 38:4	0.004232	0.00135	0.007197	0.002683	0.007571	0.001488
PE 38:3	0.011609	0.001036	0.012527	0.003054	0.007745	0.001865
PE 38:2	0.013879	0.002369	0.017746	0.003144	0.013387	0.002828
PE 40:3	0.010365	0.002234	0.00908	0.00189	0.006878	0.001992
PE 40:2	0.019375	0.002021	0.021904	0.005209	0.022681	0.003345
PE 42:4	0.005344	0.001589	0.005598	0.001532	0.003442	0.001347
PE 42:3	0.024628	0.003502	0.020413	0.001878	0.017848	0.002955

PE 42:2	0.020057	0.002925	0.018349	0.003767	0.01852	0.002405
Total PE	3.196638	0.101153	3.067843	0.381868	2.531935	0.23379
Sample description		Col-o 12hr mock		101919 12hr mock		082589 12hr mock
PE 34:4	0.00963	0.000961	0.009253	0.00105	0.006911	0.002699
PE 34:3	0.983476	0.018397	0.65158	0.111688	0.482148	0.031653
PE 34:2	0.84519	0.044342	0.800819	0.10329	0.781993	0.082884
PE 34:1	0.015495	0.004157	0.013837	0.001972	0.011597	0.003729
PE 36:6	0.304206	0.010731	0.195482	0.03913	0.127097	0.017627
PE 36:5	0.640076	0.018452	0.524021	0.040027	0.462128	0.053227
PE 36:4	0.406424	0.026873	0.419273	0.055793	0.412578	0.045328
PE 36:3	0.091696	0.004435	0.081148	0.008412	0.072878	0.01127
PE 36:2	0.036718	0.003224	0.045256	0.007649	0.057085	0.009286
PE 36:1	0.001211	0.001049	0.001109	0.001213	0	0
PE 38:6	0.005866	0.000991	0.005161	0.002386	0.003414	0.001045
PE 38:5	0.006655	0.001114	0.006916	0.001164	0.007152	0.000788
PE 38:4	0.003458	0.000883	0.006464	0.001635	0.009343	0.003339
PE 38:3	0.01194	0.001829	0.010452	0.00163	0.010006	0.003033
PE 38:2	0.013393	0.002643	0.014108	0.003487	0.012615	0.00345
PE 40:3	0.010027	0.002421	0.008263	0.001156	0.007118	0.001632
PE 40:2	0.019142	0.002388	0.020145	0.003614	0.023383	0.004189
PE 42:4	0.005015	0.001071	0.004429	0.001444	0.003492	0.001497
PE 42:3	0.021996	0.002657	0.019611	0.003794	0.018775	0.003282
PE 42:2	0.018672	0.002078	0.017134	0.002213	0.018638	0.003386
Total PE	3.450286	0.090569	2.85446	0.249754	2.528348	0.237849
Sample description		Col-o 12hr SA		101919 12hr SA		082589 12hr SA
PE 34:4	0.009588	0.001775	0.010832	0.00141	0.010399	0.00236
PE 34:3	1.115057	0.128758	0.82046	0.077148	0.55442	0.113538
PE 34:2	0.879624	0.101566	0.797801	0.047422	0.780053	0.035782
PE 34:1	0.021172	0.006887	0.01679	0.008074	0.015442	0.005405
PE 36:6	0.35787	0.03674	0.271406	0.028884	0.161115	0.046917
PE 36:5	0.709166	0.084471	0.611142	0.035274	0.486065	0.051352
PE 36:4	0.433036	0.064601	0.422399	0.02434	0.43334	0.036826
PE 36:3	0.104182	0.016881	0.089741	0.004015	0.078024	0.006082
PE 36:2	0.039407	0.004638	0.044455	0.005129	0.053683	0.006604
PE 36:1	0.002436	0.000675	0.00161	0.000965	0.001031	0.001537
PE 38:6	0.006867	0.0009	0.004518	0.001861	0.002985	0.000589
PE 38:5	0.008049	0.002	0.005619	0.001275	0.007628	0.000547
PE 38:4	0.004036	0.001121	0.004493	0.001903	0.007342	0.001624
PE 38:3	0.013163	0.001711	0.010269	0.001122	0.009383	0.002828
PE 38:2	0.015555	0.001246	0.011788	0.002973	0.012254	0.002454
PE 40:3	0.013039	0.001788	0.007778	0.001463	0.008816	0.003497
PE 40:2	0.020263	0.002109	0.018599	0.003458	0.022412	0.001788
PE 42:4	0.006557	0.001433	0.004549	0.000976	0.004511	0.000808
PE 42:3	0.023508	0.003038	0.021002	0.004215	0.019859	0.003762
PE 42:2	0.019154	0.003848	0.018408	0.002186	0.022428	0.001891
Total PE	3.801729	0.432368	3.193657	0.189268	2.691191	0.238106

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.011634	0.001395	0.009199	0.001015	0.008643	0.001493
PE 34:3	1.057521	0.132055	0.637107	0.045743	0.501659	0.084975
PE 34:2	0.778887	0.06857	0.757988	0.0441	0.697084	0.071273
PE 34:1	0.017926	0.006642	0.01866	0.005397	0.011589	0.002827
PE 36:6	0.346233	0.06504	0.199564	0.018192	0.143727	0.024003
PE 36:5	0.62437	0.05021	0.537242	0.031127	0.44514	0.045243
PE 36:4	0.3623	0.03662	0.422357	0.032717	0.382096	0.030659
PE 36:3	0.092194	0.00727	0.094196	0.01116	0.07693	0.00737
PE 36:2	0.032421	0.003917	0.049369	0.003105	0.04977	0.007007
PE 36:1	0.002043	0.001258	0.001837	0.000653	0.00061	0.000671
PE 38:6	0.005534	0.001307	0.004509	0.000745	0.003745	0.000906
PE 38:5	0.005787	0.000887	0.007743	0.001148	0.006826	0.000796
PE 38:4	0.003264	0.000629	0.007167	0.000637	0.00597	0.001044
PE 38:3	0.010875	0.002937	0.012176	0.001445	0.008504	0.002276
PE 38:2	0.012947	0.002607	0.017507	0.004073	0.013508	0.001964
PE 40:3	0.012278	0.00243	0.007716	0.001493	0.007833	0.00096
PE 40:2	0.018634	0.002829	0.020032	0.001552	0.021426	0.003211
PE 42:4	0.006476	0.001772	0.005314	0.000707	0.005474	0.001229
PE 42:3	0.023334	0.003938	0.021009	0.001426	0.021721	0.004377
PE 42:2	0.019725	0.002728	0.016787	0.002079	0.020288	0.004282
Total PE	3.444383	0.294251	2.847479	0.153072	2.432543	0.272288
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.008673	0.003267	0.006838	0.00229	0.007093	0.000778
PE 34:3	0.954682	0.109834	0.750085	0.10993	0.46519	0.035404
PE 34:2	0.801772	0.088951	0.769601	0.1081	0.75233	0.075186
PE 34:1	0.020845	0.00609	0.021783	0.010098	0.023519	0.010012
PE 36:6	0.301038	0.020681	0.218365	0.045351	0.126445	0.022324
PE 36:5	0.550424	0.054328	0.509324	0.09837	0.441298	0.022783
PE 36:4	0.348062	0.035396	0.414576	0.104151	0.387986	0.022098
PE 36:3	0.088725	0.023734	0.104195	0.012916	0.08794	0.010206
PE 36:2	0.030144	0.006351	0.049064	0.014848	0.059415	0.00909
PE 36:1	0.000906	0.000962	0.001316	0.000993	0.000798	0.001123
PE 38:6	0.004024	0.003146	0.002062	0.001295	0.00072	0.001012
PE 38:5	0.004989	0.002212	0.005781	0.003372	0.003138	0.003111
PE 38:4	0.001617	0.000949	0.003553	0.002426	0.005926	0.004573
PE 38:3	0.008338	0.001866	0.008559	0.001148	0.006133	0.000591
PE 38:2	0.008486	0.002456	0.013864	0.005422	0.014378	0.005159
PE 40:3	0.008758	0.002576	0.006444	0.003982	0.004894	0.003632
PE 40:2	0.013284	0.002691	0.017824	0.003441	0.017537	0.001859
PE 42:4	0.001466	0.000744	0.002287	0.001219	0.000634	0.000885
PE 42:3	0.017319	0.00308	0.013107	0.007063	0.018492	0.004194
PE 42:2	0.014307	0.003771	0.01828	0.004516	0.020292	0.003047
Total PE	3.187861	0.229556	2.936909	0.35387	2.444158	0.106721

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.000509	0.000711	0.002232	0.002271	0	0
PI 34:3	0.808404	0.07349	0.638957	0.073772	0.333052	0.028963
PI 34:2	0.456888	0.037071	0.466344	0.073597	0.312738	0.032475
PI 34:1	0.00717	0.005027	0.01193	0.00437	0.005259	0.003696
PI 36:6	0.025428	0.00971	0.023506	0.008509	0.009599	0.004778
PI 36:5	0.026685	0.002602	0.029377	0.007549	0.017068	0.009089
PI 36:4	0.007965	0.003837	0.018389	0.005267	0.012102	0.007072
PI 36:3	0.005556	0.004508	0.018437	0.010009	0.008971	0.006172
PI 36:2	0.003094	0.00216	0.014734	0.011087	0.008048	0.003445
PI 36:1	0.000413	0.000568	0.002339	0.001495	0.003633	0.002481
Total PI	1.342112	0.08784	1.226247	0.130552	0.71047	0.084258
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave		ave		ave	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.000603	0.001348	0.000824	0.001486	0.000803	0.000271
PI 34:3	0.883139	0.053231	0.565643	0.14394	0.411135	0.047609
PI 34:2	0.478798	0.027064	0.408688	0.056532	0.439023	0.074014
PI 34:1	0.010363	0.006676	0.008105	0.004348	0.003722	0.004078
PI 36:6	0.025137	0.007272	0.024612	0.003851	0.023703	0.00247
PI 36:5	0.026639	0.002847	0.023942	0.004957	0.030014	0.008762
PI 36:4	0.008124	0.00428	0.016387	0.007977	0.012548	0.009336
PI 36:3	0.007167	0.00187	0.013657	0.007186	0.010359	0.006967
PI 36:2	0.004365	0.003302	0.009811	0.007759	0.019082	0.0083
PI 36:1	0.004814	0.003248	0.003818	0.003806	0.004289	0.002887
Total PI	1.449149	0.074254	1.075486	0.171658	0.954678	0.121981
	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave		ave		ave	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.001108	0.001024	0.001068	0.000804	0.001962	0.002038
PI 34:3	0.975126	0.101428	0.744428	0.1043	0.490838	0.151623
PI 34:2	0.511793	0.066612	0.451626	0.053095	0.409454	0.090989
PI 34:1	0.01147	0.006852	0.005553	0.003632	0.003992	0.004822
PI 36:6	0.035976	0.012477	0.032038	0.005077	0.021571	0.007233
PI 36:5	0.036218	0.005396	0.034303	0.004331	0.036998	0.0138
PI 36:4	0.011939	0.001368	0.014791	0.008902	0.019563	0.003898
PI 36:3	0.012072	0.0033	0.007851	0.005329	0.007906	0.004394
PI 36:2	0.004571	0.001386	0.007505	0.003733	0.010759	0.005532
PI 36:1	0.005505	0.002524	0.00642	0.005389	0.002716	0.006073
Total PI	1.605779	0.164159	1.305582	0.163594	1.005758	0.248676

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.001064	0.00147	0.001719	0.000819	0.002787	0.002234
PI 34:3	0.781843	0.125719	0.51194	0.029323	0.421846	0.044913
PI 34:2	0.372951	0.077626	0.374085	0.02138	0.356448	0.023642
PI 34:1	0.00483	0.007001	0.002256	0.003225	0.002286	0.004797
PI 36:6	0.029444	0.010987	0.02479	0.005063	0.019677	0.00328
PI 36:5	0.026422	0.009631	0.026652	0.006122	0.027808	0.004852
PI 36:4	0.009098	0.002346	0.015515	0.008551	0.012205	0.003337
PI 36:3	0.007084	0.003066	0.017157	0.006125	0.01224	0.005049
PI 36:2	0.003574	0.001579	0.013224	0.002419	0.009774	0.004066
PI 36:1	0.001855	0.001735	0.005705	0.002494	0.002768	0.00251
Total PI	1.238165	0.197388	0.993044	0.0539	0.86784	0.070421
Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.000322	0.000719	0	0	0	0
PI 34:3	1.189768	0.378186	0.75351	0.377865	0.521107	0.042885
PI 34:2	0.58403	0.202052	0.424729	0.077244	0.466284	0.097493
PI 34:1	0.016128	0.0145	0.009321	0.010137	0.010186	0.014992
PI 36:6	0.02755	0.015485	0.022166	0.014579	0.01369	0.012177
PI 36:5	0.02514	0.006021	0.036409	0.018076	0.030209	0.010203
PI 36:4	0.009735	0.00615	0.003067	0.005931	0.006758	0.00383
PI 36:3	0.002354	0.003098	0.003494	0.003598	0.009557	0.008395
PI 36:2	0.001337	0.001841	0.00529	0.005299	0.007738	0.008944
PI 36:1	0.000662	0.001481	0.010681	0.00786	0	0
Total PI	1.857025	0.598644	1.268669	0.456039	1.065529	0.11808

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0	0	0	0	0	0
PS 34:3	0.021692	0.003205	0.01954	0.006906	0.006309	0.003261
PS 34:2	0.012799	0.002177	0.013743	0.003938	0.006353	0.003193
PS 34:1	0.000152	0.000287	0	0	0	0
PS 36:6	0.000127	0.00018	0	0	9E-05	0.000201
PS 36:5	0.000821	0.000849	0.000201	0.000275	0	0
PS 36:4	0.000764	0.000481	0	0	0.000264	0.000378
PS 36:3	0.002622	0.001031	0.005036	0.004208	0.001086	0.001804
PS 36:2	0.001966	0.001773	0.002467	0.00202	0.000796	0.000977
PS 36:1	0	0	0	0	0	0
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0.003329	0.002179	0.001471	0.002317	0.00113	0.001177
PS 38:2	0.004501	0.001834	0.00211	0.002452	0.001519	0.002295
PS 38:1	0	0	0	0	0.000203	0.000455
PS 40:4	4.29E-05	9.58E-05	0	0	0	0
PS 40:3	0.01294	0.003038	0.009278	0.007912	0.002386	0.002184
PS 40:2	0.014448	0.00195	0.009011	0.007421	0.003218	0.004682
PS 40:1	0.000321	0.0005	0	0	0	0
PS 42:4	0.011063	0.006495	0.002939	0.002869	0	0
PS 42:3	0.077254	0.013002	0.074986	0.015895	0.018585	0.010049
PS 42:2	0.055452	0.01546	0.047886	0.016538	0.009232	0.00727
PS 42:1	0.000364	0.000813	0	0	0	0
PS 44:3	0.000812	0.001816	0.002964	0.006628	0	0
PS 44:2	0.001429	0.003195	0	0	0	0
Total PS	0.2229	0.037095	0.191631	0.046115	0.051172	0.021612

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0	0	0	0	0	0
PS 34:3	0.021291	0.004367	0.015132	0.005866	0.009882	0.005978
PS 34:2	0.012073	0.003214	0.013402	0.005955	0.010958	0.004196
PS 34:1	0	0	0	0	0	0
PS 36:6	0	0	0	0	0.000142	0.000318
PS 36:5	0.000121	0.00027	0.000288	0.000279	0.000511	0.000483
PS 36:4	9.77E-05	0.000218	0.000246	0.000448	2.77E-05	6.19E-05
PS 36:3	0.000856	0.000562	0.003232	0.002862	0.003343	0.00358
PS 36:2	0.001419	0.001337	0.001387	0.000712	0.000961	0.001417
PS 36:1	0	0	0	0	8.03E-05	0.00018
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0.001871	0.002507	0.001991	0.001308	0.001867	0.003418
PS 38:2	0.002552	0.001158	0.0018	0.001952	0.00358	0.004483
PS 38:1	0	0	0	0	0	0
PS 40:4	0	0	0	0	0	0
PS 40:3	0.01188	0.006543	0.006059	0.003645	0.005354	0.003287
PS 40:2	0.009747	0.004138	0.008025	0.004358	0.006262	0.006959
PS 40:1	0.000321	0.00054	0.000889	0.001267	0.000656	0.000953
PS 42:4	0.005547	0.006482	0.001801	0.001363	0.001603	0.00151
PS 42:3	0.07458	0.010601	0.033252	0.015522	0.044831	0.013463
PS 42:2	0.040186	0.014341	0.029842	0.022521	0.0139	0.012515
PS 42:1	0	0	1.67E-05	3.74E-05	0.000129	0.000289
PS 44:3	0	0	0	0	0	0
PS 44:2	0	0	0.000932	0.002085	0	0
Total PS	0.182542	0.030411	0.118296	0.048758	0.104088	0.044536

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0	0	0	0	0	0
PS 34:3	0.027455	0.007878	0.01591	0.003956	0.014166	0.008307
PS 34:2	0.011989	0.005342	0.012766	0.003894	0.004736	0.003746
PS 34:1	0	0	0	0	0	0
PS 36:6	0.000423	0.000582	7.71E-05	0.000172	0	0
PS 36:5	0.000545	0.000831	0.0003	0.000469	0	0
PS 36:4	0.000349	0.000338	0.000307	0.000351	0.000319	0.000712
PS 36:3	0.001217	0.001442	0.001328	0.00097	0.000699	0.001
PS 36:2	0.000787	0.000728	0.000503	0.000927	0.000396	0.000689
PS 36:1	0	0	0	0	8.09E-05	0.000181
PS 38:6	0.000155	0.000347	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0.000941	0.001297	0.001057	0.000655	0	0
PS 38:2	0.003293	0.0032	0.001647	0.001359	0.000694	0.001552
PS 38:1	0	0	0	0	0	0
PS 40:4	0	0	0	0	0	0
PS 40:3	0.009629	0.007069	0.011532	0.006913	0.002396	0.003804
PS 40:2	0.011282	0.011183	0.009355	0.005523	0.002054	0.002248
PS 40:1	0	0	0.000306	0.000684	0.000431	0.000964
PS 42:4	0.00657	0.000551	0.004991	0.003464	0.000579	0.000818
PS 42:3	0.078685	0.028384	0.046871	0.017334	0.032868	0.020009
PS 42:2	0.048835	0.022566	0.028087	0.014305	0.011189	0.008342
PS 42:1	6.57E-05	0.000147	0.000278	0.000621	0.000191	0.000428
PS 44:3	0.000509	0.001138	0	0	0	0
PS 44:2	0	0	0	0	0.001364	0.00305
Total PS	0.202727	0.057393	0.135314	0.048313	0.072163	0.024472

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0	0	0	0	0	0
PS 34:3	0.016759	0.004766	0.011824	0.004226	0.012454	0.006747
PS 34:2	0.008105	0.001785	0.009722	0.005059	0.009657	0.002576
PS 34:1	0	0	3.41E-05	7.63E-05	0	0
PS 36:6	0.000197	0.000307	0.000217	0.000395	0.000118	0.000264
PS 36:5	0.000115	0.000158	0.000375	0.000528	0.000168	0.000375
PS 36:4	4.6E-05	0.000103	0.000245	0.000229	0	0
PS 36:3	0.000231	0.000266	0.001686	0.001366	0.001378	0.001249
PS 36:2	0.000239	0.000364	0.002423	0.002308	0.000914	0.001152
PS 36:1	0	0	6.89E-05	0.000154	0	0
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0.000745	0.000809	0.000952	0.000723	0.000197	0.00044
PS 38:2	0.001055	0.000828	0.003599	0.002649	0.00177	0.001527
PS 38:1	0	0	0.000187	0.000206	0	0
PS 40:4	4.16E-05	9.31E-05	0	0	0	0
PS 40:3	0.007771	0.001581	0.007469	0.004367	0.002271	0.002708
PS 40:2	0.005763	0.004063	0.007247	0.001493	0.004154	0.003768
PS 40:1	0	0	0	0	0.000655	0.000905
PS 42:4	0.003205	0.000526	0.002702	0.001723	0.001455	0.002663
PS 42:3	0.049738	0.010652	0.045914	0.017232	0.044338	0.030215
PS 42:2	0.033078	0.012343	0.033141	0.005648	0.016279	0.011123
PS 42:1	0	0	0	0	0.000445	0.000995
PS 44:3	0.0009	0.002013	0	0	0	0
PS 44:2	0.000991	0.002216	0	0	0	0
Total PS	0.128981	0.020948	0.127804	0.034052	0.096252	0.03343

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0	0	0	0	0	0
PS 34:3	0.028718	0.01209	0.013669	0.00695	0.018341	0.010702
PS 34:2	0.016309	0.005428	0.013808	0.005342	0.022154	0.00872
PS 34:1	0	0	0	0	0	0
PS 36:6	0	0	0	0	0	0
PS 36:5	0.000258	0.000456	0.000262	0.000585	0	0
PS 36:4	0.000252	0.000383	0	0	0	0
PS 36:3	0.001213	0.001754	0	0	0.003302	0.005886
PS 36:2	0	0	0	0	0.002905	0.001557
PS 36:1	0	0	0	0	0	0
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0.000968	0.001153	0.000782	0.001123	0.000661	0.000934
PS 38:2	0.001933	0.001876	0.001569	0.002586	0.003714	0.004259
PS 38:1	0	0	0	0	5.32E-05	0.000119
PS 40:4	0	0	0	0	0	0
PS 40:3	0.01154	0.007319	0.004605	0.004945	0.010428	0.004037
PS 40:2	0.013607	0.007407	0.004176	0.005375	0.014138	0.010027
PS 40:1	0	0	0	0	0	0
PS 42:4	0.004878	0.003326	0.001688	0.002474	0.004972	0.003574
PS 42:3	0.0589	0.015048	0.030649	0.013752	0.026625	0.014607
PS 42:2	0.037815	0.020181	0.019263	0.012843	0.030442	0.018934
PS 42:1	0.000521	0.000845	0	0	0	0
PS 44:3	0	0	0	0	0	0
PS 44:2	0	0	0.000566	0.001266	0	0
Total PS	0.176911	0.053358	0.091036	0.030671	0.137736	0.049858

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0	0	0	0	0	0
PA 34:4	0	0	0	0	0	0
PA 34:3	0.018287	0.017368	0	0	0	0
PA 34:2	0.004546	0.006249	0	0	0	0
PA 34:1	0.003883	0.008683	0.000388	0.000867	0.027571	0.036988
PA 36:6	0.005108	0.004768	0	0	0	0
PA 36:5	0.006686	0.006393	0	0	0	0
PA 36:4	0	0	0	0	0	0
PA 36:3	0.000601	0.001343	0	0	0	0
PA 36:2	0.002586	0.003554	0.001443	0.003227	0	0
Total PA	0.041697	0.039159	0.001831	0.004094	0.027571	0.036988
Total	100	7.11E-15	100	0	100	7.11E-15
Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0	0	0	0	0.000237	0.000531
PA 34:4	0	0	0	0	0	0
PA 34:3	0	0	0.003767	0.004643	0.002096	0.002511
PA 34:2	0	0	0.000292	0.000653	0.00028	0.000625
PA 34:1	0.012134	0.027133	0	0	0	0
PA 36:6	0	0	0.005431	0.004665	0.000388	0.000868
PA 36:5	0	0	0.00157	0.003511	0	0
PA 36:4	0	0	0.000941	0.001602	0	0
PA 36:3	0	0	0.001313	0.001308	0.000271	0.00041
PA 36:2	0	0	0.000477	0.000726	0.001129	0.001807
Total PA	0.012134	0.027133	0.013792	0.006307	0.004401	0.004223
Total	100	7.11E-15	100	7.11E-15	100	0
Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.002142	0.002167	0	0	0.000321	0.000718
PA 34:4	0	0	0	0	0	0
PA 34:3	0.007491	0.007062	0.001941	0.003048	0.00965	0.018668
PA 34:2	0.000522	0.000715	0	0	0.00844	0.014008
PA 34:1	0.022626	0.018261	0.050564	0.05092	0.003473	0.007765
PA 36:6	0.001322	0.001535	0.003091	0.004094	0.002889	0.003068
PA 36:5	0.003203	0.003684	0.006798	0.012909	0.009051	0.017496
PA 36:4	0	0	0.00084	0.001877	0.003051	0.003308
PA 36:3	0.000588	0.001315	0.00018	0.000403	0.002282	0.005103
PA 36:2	0.003126	0.006566	0.000845	0.00189	0.001877	0.004197
Total PA	0.04102	0.025687	0.064259	0.05548	0.041032	0.069016
Total	100	0	100	0	100	0

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.00034	0.00076	0.000526	0.00113	0.001874	0.002593
PA 34:4	0	0	0	0	0	0
PA 34:3	0.007737	0.005464	0.002197	0.003563	0.003872	0.002517
PA 34:2	0	0	0.00152	0.001592	5.42E-06	1.21E-05
PA 34:1	0	0	0	0	0	0
PA 36:6	0.001376	0.002118	0.000546	0.000943	0.00056	0.00091
PA 36:5	0.001053	0.000998	0.000586	0.001053	1.45E-05	3.25E-05
PA 36:4	0.000859	0.001842	0	0	0	0
PA 36:3	0.00216	0.004542	0.000233	0.000521	0	0
PA 36:2	0.000326	0.000729	0.002143	0.002959	0.003241	0.004604
Total PA	0.013852	0.010312	0.007752	0.004844	0.009567	0.007952
Total	100	7.11E-15	100	7.11E-15	100	0
Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0	0	0.001017	0.002275	0.004469	0.004546
PA 34:4	0	0	0	0	0	0
PA 34:3	0.068647	0.035568	0.033043	0.022073	0.036151	0.010712
PA 34:2	0.02175	0.015697	0.004316	0.00783	0.018203	0.013433
PA 34:1	0	0	0	0	0.007107	0.015891
PA 36:6	0.024276	0.017761	0.002182	0.002213	0.003858	0.004256
PA 36:5	0.025057	0.025752	0.008755	0.00559	0.006578	0.005212
PA 36:4	0.011171	0.009089	0.00823	0.004126	0.003849	0.0065
PA 36:3	0	0	0	0	0	0
PA 36:2	0.002687	0.003028	0.002413	0.00347	0.004841	0.00943
Total PA	0.153589	0.08956	0.059957	0.028528	0.085055	0.021053
Total	100	0	100	0	100	0

TABLE A – 5. TOTAL LIPID SPECIES – ESI MS/MS EXPERIMENT 2

Sample	Col-o 0hr		101919 0hr		082589 0hr	
description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	0.976155	0.045772	1.199231	0.11765	1.107258	0.098431
DGDG 34:5	0.150046	0.01717	0.156951	0.0127	0.160068	0.015769
DGDG 34:4	0.101744	0.014844	0.107791	0.006589	0.103107	0.007756
DGDG 34:2	2.463049	0.19697	3.177842	0.157545	2.68618	0.384431
DGDG 34:1	0.310719	0.032989	0.340527	0.033951	0.301028	0.040219
DGDG 36:6	0.135324	0.017833	0.099618	0.02113	0.109213	0.015967
DGDG 36:5	8.221806	0.422299	10.14732	0.686686	8.701111	1.219671
DGDG 36:4	0.237258	0.027268	0.363101	0.072051	0.247828	0.076908
DGDG 36:3	0.119964	0.007014	0.151755	0.004951	0.134358	0.020778
DGDG 36:2	0.08774	0.008988	0.09706	0.008444	0.081888	0.014968
DGDG 36:1	0.006195	0.001072	0.005854	0.00195	0.005376	0.001721
DGDG 38:6	0.001833	0.002666	0.000273	0.00061	0	0
DGDG 38:5	0.039989	0.007591	0.01662	0.008638	0.012586	0.007579
DGDG 38:4	0.004114	0.002176	0.00474	0.00167	0.00321	0.003426
DGDG 38:3	0.001024	0.000963	0.001651	0.001205	0.001105	0.00109
Total						
DGDG	0.000523	0.000481	0.000442	0.000497	0	0

Sample	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.138036	0.11828	1.179106	0.058987	1.084596	0.100206
DGDG 34:5	0.179504	0.030237	0.159925	0.008606	0.153547	0.009438
DGDG 34:4	0.095625	0.007553	0.102214	0.005064	0.095241	0.006405
DGDG 34:2	2.264899	0.088007	2.743864	0.357521	2.465168	0.164117
DGDG 34:1	0.292484	0.044858	0.32987	0.0482	0.287379	0.016192
DGDG 36:6	0.123174	0.007772	0.084944	0.017485	0.101522	0.010652
DGDG 36:5	7.806316	0.576673	9.491791	0.792943	8.434481	0.349573
DGDG 36:4	0.250777	0.066496	0.370828	0.059135	0.260425	0.032692
DGDG 36:3	0.122422	0.011434	0.14941	0.014441	0.132393	0.006384
DGDG 36:2	0.078606	0.012113	0.091261	0.011589	0.08405	0.008024
DGDG 36:1	0.0045	0.00332	0.003787	0.001268	0.006278	0.00222
DGDG 38:6	0.00211	0.002789	0.001909	0.00068	0.001299	0.000644
DGDG 38:5	0.036349	0.002641	0.040494	0.006398	0.019507	0.007196
DGDG 38:4	0.001985	0.001452	0.004461	0.001661	0.003507	0.001739
DGDG 38:3	0.001193	0.000968	0.002156	0.0016	0.00205	0.000992
Total						
DGDG	0.000519	0.000894	0.000861	0.000856	0.001004	0.000931

Sample	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.076327	0.040639	1.188843	0.161852	1.154261	0.116826
DGDG 34:5	0.16633	0.016909	0.169073	0.027179	0.165356	0.01561
DGDG 34:4	0.093218	0.005018	0.099489	0.008543	0.104441	0.011902
DGDG 34:2	2.227266	0.115787	2.59195	0.258159	2.525363	0.312792
DGDG 34:1	0.2782	0.021987	0.30016	0.036956	0.326421	0.047012
DGDG 36:6	0.114965	0.009743	0.108826	0.012494	0.118919	0.012719
DGDG 36:5	7.750262	0.586036	8.525782	0.905467	8.457798	0.759159
DGDG 36:4	0.203162	0.017061	0.264612	0.058136	0.271114	0.064498
DGDG 36:3	0.113189	0.012036	0.129104	0.007769	0.130743	0.013151
DGDG 36:2	0.076907	0.013414	0.078042	0.007303	0.082266	0.016213
DGDG 36:1	0.004083	0.002298	0.00798	0.001809	0.007853	0.002107
DGDG 38:6	0.001063	0.000493	0.000534	0.000749	0.002088	0.00241
DGDG 38:5	0.019646	0.008624	0.011595	0.006877	0.008237	0.007623
DGDG 38:4	0.00283	0.001231	0.003133	0.003247	0.002805	0.001593
DGDG 38:3	0.001627	0.001592	0.000531	0.000961	0.000406	0.00039
Total						
DGDG	0.000559	0.000681	0.000863	0.000807	0.001114	0.00159

Sample	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.160204	0.146054	1.227902	0.097925	1.132529	0.090902
DGDG 34:5	0.173712	0.016447	0.148221	0.014353	0.171179	0.00876
DGDG 34:4	0.106344	0.015504	0.107068	0.014866	0.11	0.006728
DGDG 34:2	2.379702	0.197654	2.836866	0.300781	2.23042	0.249133
DGDG 34:1	0.29129	0.036494	0.327405	0.051856	0.274761	0.024986
DGDG 36:6	0.119781	0.031228	0.113068	0.027189	0.119818	0.025887
DGDG 36:5	8.126683	0.86282	9.281253	1.319052	7.723359	0.671708
DGDG 36:4	0.235316	0.063034	0.314326	0.117168	0.213025	0.033124
DGDG 36:3	0.118586	0.009239	0.139434	0.026312	0.113521	0.01023
DGDG 36:2	0.073482	0.011995	0.087268	0.019272	0.070621	0.006311
DGDG 36:1	0.004408	0.003729	0.005094	0.004152	0.00227	0.002181
DGDG 38:6	0	0	0.000154	0.000343	0.001706	0.001663
DGDG 38:5	0	0	0.004234	0.007353	0.027097	0.013244
DGDG 38:4	0.002411	0.001979	0.001224	0.001425	0.00159	0.001645
DGDG 38:3	0.000799	0.001501	0.000184	0.000411	0.000622	0.00067
Total						
DGDG	0.000753	0.000785	0	0	0.000198	0.000443

Sample	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.291049	0.095521	1.181226	0.112217	1.228098	0.072988
DGDG 34:5	0.176845	0.016458	0.16427	0.011913	0.168141	0.006302
DGDG 34:4	0.09132	0.015396	0.094178	0.006124	0.101959	0.004273
DGDG 34:2	2.351822	0.196054	2.560398	0.220479	2.412143	0.146313
DGDG 34:1	0.298527	0.02762	0.310588	0.022383	0.304415	0.014618
DGDG 36:6	0.122992	0.010792	0.113804	0.013313	0.114775	0.007746
DGDG 36:5	8.279249	0.623775	8.894415	0.628781	9.050711	0.432125
DGDG 36:4	0.226436	0.043245	0.276158	0.030466	0.32784	0.066958
DGDG 36:3	0.120545	0.008475	0.133651	0.01292	0.131297	0.010148
DGDG 36:2	0.080227	0.007495	0.083231	0.008827	0.08177	0.010845
DGDG 36:1	0.003602	0.000886	0.006308	0.000885	0.004058	0.001865
DGDG 38:6	0.000733	0.000628	0.001084	0.001194	0.001035	0.001501
DGDG 38:5	0.036654	0.005516	0.031982	0.019465	0.018158	0.010666
DGDG 38:4	0.003397	0.002495	0.002636	0.002101	0.002863	0.001621
DGDG 38:3	0.000844	0.000976	0.002269	0.000627	0.001328	0.00094
Total						
DGDG	0	0	0.001112	0.00086	0.000619	0.000694

Sample	Col-o 0hr		101919 0hr		082589 0hr	
	description	ave	stdev	ave	stdev	ave
MGDG 34:6	12.85748	0.671984	15.87078	0.909038	13.65432	1.766996
MGDG 34:5	61.44616	2.290161	59.49828	1.261516	62.07145	2.105278
MGDG 34:4	2.64632	0.412002	1.641517	0.374548	2.625549	0.547046
MGDG 34:3	1.422279	0.318133	0.752714	0.195534	1.245374	0.275819
MGDG 34:2	0.587336	0.072888	0.462941	0.084136	0.574353	0.051893
MGDG 34:1	0.204411	0.048744	0.085671	0.026088	0.15095	0.061278
MGDG 36:6	0.073366	0.013521	0.038095	0.020723	0.04856	0.017225
MGDG 36:5	7.825327	0.373066	8.223436	0.423171	7.843256	0.71196
MGDG 36:4	0.456504	0.012791	0.547867	0.089167	0.45873	0.069411
MGDG 36:3	0.138098	0.033546	0.101193	0.019008	0.120598	0.007244
MGDG 36:2	0.020317	0.004675	0.015703	0.005637	0.016749	0.005791
MGDG 36:1	0.001869	0.000658	0	0	6.81E-05	0.000152
MGDG 38:6	0.001382	0.001927	0.000539	0.001205	0	0
MGDG 38:5	0.011387	0.002713	0.004282	0.001349	0.004892	0.001704
MGDG 38:4	0.002714	0.000512	0.001687	0.001205	0.001041	0.001741
MGDG 38:3	0.00088	0.001296	0.002176	0.000894	0.001033	0.000228
Total MGDG	0.000626	0.000447	2.78E-05	6.22E-05	0	0

Sample	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	12.3985	0.868764	14.75688	1.247919	13.13245	0.477791
MGDG 34:5	63.19291	0.811779	62.28263	1.770993	63.28114	2.286254
MGDG 34:4	2.70669	0.651094	1.66638	0.499561	2.34634	0.376189
MGDG 34:3	1.283788	0.177854	0.678492	0.191486	1.136759	0.225561
MGDG 34:2	0.600633	0.065705	0.445805	0.06896	0.504476	0.045122
MGDG 34:1	0.178774	0.019908	0.073967	0.0188	0.140744	0.036209
MGDG 36:6	0.120986	0.012624	0.068088	0.020302	0.055009	0.010372
MGDG 36:5	7.495969	0.271444	7.712989	0.72833	8.31132	0.56224
MGDG 36:4	0.546099	0.078691	0.635474	0.030146	0.495915	0.034671
MGDG 36:3	0.125433	0.020226	0.100401	0.014402	0.116691	0.014598
MGDG 36:2	0.018849	0.004173	0.014475	0.002881	0.018909	0.006976
MGDG 36:1	0.001893	0.001463	0.000315	0.000704	0.001526	0.00159
MGDG 38:6	0.002223	0.003153	0.001176	0.00161	0	0
MGDG 38:5	0.011483	0.002806	0.005616	0.002309	0.00731	0.00266
MGDG 38:4	0.003949	0.003362	0.004671	0.002182	0.002097	0.001528
MGDG 38:3	0.003164	0.000805	0.001229	0.001288	0.003465	0.001118
Total MGDG	0.001098	0.001976	0.000705	0.000522	0	0

Sample	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	12.12963	0.756209	13.48052	1.37778	13.35919	1.247982
MGDG 34:5	64.09575	0.950894	63.23799	2.470736	63.50305	2.778976
MGDG 34:4	2.796912	0.252815	2.477598	0.654972	2.729293	0.533484
MGDG 34:3	1.214093	0.09286	1.125937	0.224131	1.319718	0.205318
MGDG 34:2	0.500889	0.035013	0.549078	0.050533	0.579882	0.138031
MGDG 34:1	0.142371	0.009716	0.139922	0.035834	0.178	0.044675
MGDG 36:6	0.059978	0.011313	0.055071	0.008378	0.071421	0.036735
MGDG 36:5	7.435251	0.579398	7.868484	0.53574	7.644624	0.596799
MGDG 36:4	0.375602	0.060427	0.42775	0.056858	0.468822	0.035478
MGDG 36:3	0.104976	0.007395	0.109279	0.012794	0.107064	0.014323
MGDG 36:2	0.016283	0.003374	0.012736	0.004956	0.014941	0.00457
MGDG 36:1	0.001053	0.001228	0.002298	0.00227	0.001811	0.001686
MGDG 38:6	0.001811	0.001686	0.000472	0.000731	0.001266	0.001793
MGDG 38:5	0.006119	0.001899	0.005573	0.002571	0.005631	0.002842
MGDG 38:4	0.001913	0.001438	0.002265	0.001398	0.002418	0.001984
MGDG 38:3	0.003525	0.000679	0.00257	0.001023	0.001337	0.001034
Total MGDG	9.33E-05	0.000204	0	0	0	0

Sample	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	12.79347	1.208674	14.5937	1.820035	12.19272	0.900273
MGDG 34:5	60.23251	5.106202	59.9986	3.057011	63.60457	4.083591
MGDG 34:4	2.685047	0.368963	2.114489	0.759201	2.929625	0.594113
MGDG 34:3	1.403683	0.322991	0.920396	0.474521	1.440109	0.321247
MGDG 34:2	0.549238	0.060441	0.468937	0.113501	0.539249	0.090838
MGDG 34:1	0.161465	0.060144	0.097633	0.054733	0.186368	0.046428
MGDG 36:6	0.042879	0.016332	0.044786	0.020149	0.056772	0.024917
MGDG 36:5	7.371728	1.061515	7.045643	0.546509	7.040194	0.658479
MGDG 36:4	0.451177	0.064472	0.496919	0.091048	0.423239	0.030871
MGDG 36:3	0.125636	0.012721	0.102698	0.03244	0.11906	0.018703
MGDG 36:2	0.017166	0.001701	0.011926	0.005065	0.019116	0.006255
MGDG 36:1	0.000636	0.001422	0.000251	0.000433	0.003545	0.002044
MGDG 38:6	0	0	0.000436	0.000976	0.002119	0.002364
MGDG 38:5	0.00055	0.001231	0	0	0.011372	0.003658
MGDG 38:4	0.001193	0.001786	0.000708	0.00089	0.003131	0.002264
MGDG 38:3	0.00179	0.001328	0.001607	0.00068	0.001249	0.000968
Total MGDG	0.000825	0.001667	0.000131	0.000294	5.95E-05	0.000133

Sample	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	13.08424	0.948465	13.85731	0.915411	13.94921	0.613598
MGDG 34:5	64.0501	1.242022	62.966	2.230983	63.43312	0.66253
MGDG 34:4	2.389169	0.190147	2.06553	0.268828	2.093207	0.325108
MGDG 34:3	1.036827	0.127563	0.938932	0.149288	0.90735	0.133337
MGDG 34:2	0.480812	0.012956	0.498463	0.037714	0.473603	0.056343
MGDG 34:1	0.137275	0.016562	0.128762	0.023052	0.132195	0.013426
MGDG 36:6	0.090988	0.024009	0.07858	0.010185	0.082699	0.024663
MGDG 36:5	6.942679	0.354286	7.848466	0.511054	7.232076	0.399932
MGDG 36:4	0.494951	0.065154	0.514674	0.01393	0.548896	0.10012
MGDG 36:3	0.125256	0.015183	0.11723	0.008104	0.110381	0.01753
MGDG 36:2	0.021445	0.005988	0.018792	0.003859	0.016528	0.005404
MGDG 36:1	0.004418	0.00268	0.001255	0.000889	0.0013	0.002113
MGDG 38:6	0.001885	0.00198	0	0	0	0
MGDG 38:5	0.008598	0.001952	0.012958	0.01647	0.002355	0.00329
MGDG 38:4	0.002756	0.002242	0.000875	0.000827	0.001159	0.001764
MGDG 38:3	0.001242	0.000668	0.002599	0.001001	0.002004	0.001073
Total MGDG	0	0	0	0	0	0

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	74.83898	2.021565	71.37613	1.535992	75.1626	2.299017
PG 32:0	0.17669	0.014348	0.169046	0.027608	0.136593	0.043717
PG 34:4	0.057805	0.028185	0.048832	0.021529	0.035875	0.029496
PG 34:3	2.630855	0.303522	2.123216	0.134354	2.045666	0.290946
PG 34:2	0.63178	0.053778	0.65565	0.092771	0.659688	0.078243
PG 34:1	0.466446	0.068638	0.277813	0.068951	0.339888	0.076875
PG 34:0	0.253585	0.099666	0.155455	0.019805	0.251814	0.058737
Total PG	0	0	0.00065	0.001454	0.001477	0.003303

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	76.29394	1.588864	73.69241	2.264379	76.4217	1.675591
PG 32:0	0.147816	0.068148	0.148271	0.028348	0.133504	0.02332
PG 34:4	0.039922	0.030548	0.051556	0.033127	0.049262	0.03188
PG 34:3	2.267954	0.171996	2.017389	0.302297	2.008309	0.212027
PG 34:2	0.623733	0.15403	0.612069	0.158127	0.56046	0.03105
PG 34:1	0.387156	0.077179	0.281749	0.061977	0.410915	0.196913
PG 34:0	0.29856	0.089026	0.120175	0.03455	0.132415	0.059176
Total PG	0	0	0	0	0.00093	0.001739

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	76.75662	1.021194	76.01702	2.241427	76.62928	2.645071
PG 32:0	0.051369	0.049858	0.086998	0.050637	0.104203	0.047183
PG 34:4	0.018516	0.018926	0.01895	0.017013	0.027495	0.018403
PG 34:3	2.002167	0.1971	1.896305	0.2308	2.087167	0.204148
PG 34:2	0.52396	0.06563	0.553632	0.099579	0.599958	0.198419
PG 34:1	0.320526	0.051277	0.344065	0.059716	0.458539	0.065844
PG 34:0	0.142705	0.076654	0.219989	0.084138	0.20348	0.063463
Total PG	0	0	0	0	0	0

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	73.04553	5.610416	71.30516	4.37879	76.37978	4.740878
PG 32:0	0.169596	0.074138	0.188993	0.027058	0.173699	0.015481
PG 34:4	0.050243	0.039409	0.052673	0.021576	0.039229	0.017808
PG 34:3	2.614994	0.352099	2.220116	0.349792	2.483498	0.210752
PG 34:2	0.615865	0.096185	0.597108	0.085073	0.592506	0.170739

PG 34:1	0.445952	0.059442	0.343205	0.096239	0.309157	0.046667
PG 34:0	0.254676	0.083567	0.184268	0.044287	0.157464	0.074465
Total PG	0	0	0.000702	0.001042	0	0
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
PG 32:1	75.7884	1.536884	75.19312	2.393615	75.03687	1.108658
PG 32:0	0.118378	0.02781	0.140779	0.039975	0.138494	0.046049
PG 34:4	0.045902	0.018592	0.055809	0.01939	0.039411	0.037519
PG 34:3	1.996373	0.145003	1.870998	0.218798	1.774791	0.249932
PG 34:2	0.615346	0.091172	0.613755	0.098452	0.562185	0.162911
PG 34:1	0.289088	0.041834	0.309379	0.073393	0.254283	0.103743
PG 34:0	0.123169	0.045508	0.179668	0.041793	0.13949	0.079845
Total PG	0.006979	0.00912	0.002241	0.005012	0	0

	Col-o					
	0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	4.21716	0.3397	3.430662	0.167299	3.471001	0.392146
lysoPG 16:0	0.00286	0.002804	0.004945	0.003579	0.00787	0.00233
lysoPG 18:3	0.004056	0.003262	0.000905	0.001278	0.001486	0.001599
lysoPG 18:2	0.007767	0.004303	0.010306	0.004667	0.002851	0.004776
lysoPG 18:1	0.000815	0.001823	0.001366	0.00187	0	0
Total lysoPG	0.001111	0.002421	0	0	0	0

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	3.765141	0.425154	3.231211	0.442237	3.295796	0.383442
lysoPG 16:0	0.001537	0.003437	0	0	9.03E-05	0.000202
lysoPG 18:3	0.000932	0.002084	6.94E-05	0.000155	0.000514	0.001111
lysoPG 18:2	0.005364	0.004975	0.003874	0.002538	0.00517	0.001617
lysoPG 18:1	0.000326	0.000729	0	0	0	0
Total lysoPG	0.00099	0.002213	0.000291	0.000652	0	0

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	3.059243	0.283354	3.119939	0.442987	3.480841	0.405431
lysoPG 16:0	0.00034	0.000517	0.002145	0.003453	0.001081	0.002417
lysoPG 18:3	0	0	0.000875	0.001956	0.001075	0.001708
lysoPG 18:2	0.00242	0.004313	0.002438	0.003561	0.002662	0.002514
lysoPG 18:1	0	0	0.000208	0.000465	0	0
Total lysoPG	7.49E-05	0.000167	0	0	0.000294	0.000657

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	4.151325	0.559906	3.587065	0.444684	3.755553	0.309834
lysoPG 16:0	0	0	0.000794	0.001136	0	0
lysoPG 18:3	0.004323	0.007605	0.00275	0.004116	0	0
lysoPG 18:2	0.009297	0.005428	0.006816	0.003416	0.009072	0.004593
lysoPG 18:1	0	0	0.001171	0.001165	0.000228	0.00051
Total lysoPG	0	0	0	0	0.002221	0.00339

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	3.195235	0.156154	3.17263	0.294213	2.908654	0.344081
lysoPG 16:0	0.001485	0.002382	0.000694	0.001552	0.001849	0.001716
lysoPG 18:3	0	0	0.002029	0.00236	0.001769	0.002552
lysoPG 18:2	0.005563	0.003464	0.004227	0.003493	0.003177	0.002031
lysoPG 18:1	0	0	0.000885	0.00122	0	0
Total lysoPG	0.002405	0.004519	0	0	0	0

	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.01661	0.005186	0.017521	0.005251	0.012207	0.006134
LysoPC 16:0	0.002403	0.005374	0	0	0	0
LysoPC 18:3	0.024158	0.030515	0.009472	0.00152	0.002256	0.001729
LysoPC 18:2	0.002798	0.001783	0.003132	0.001289	0.001164	0.001771
LysoPC 18:1	0.002248	0.003175	0.001273	0.001543	0.001214	0.00134
LysoPC 18:0	0.002368	0.005294	0	0	0.001409	0.003151
Total LysoPC	0.005405	0.009902	0	0	0	0

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.009149	0.00915	0.004235	0.002892	0.005774	0.002342
LysoPC 16:0	0.000517	0.001157	0.000621	0.001039	0.00086	0.001922
LysoPC 18:3	0.014612	0.013167	0.006802	0.00502	0.001852	0.004142
LysoPC 18:2	0.00229	0.003827	0.003142	0.005004	0.0009	0.001233
LysoPC 18:1	0.004245	0.003163	0.005469	0.001508	0.001949	0.004061
LysoPC 18:0	0.001607	0.003594	0.000614	0.001372	0.001222	0.002214
Total LysoPC	0.009441	0.015629	0.003843	0.005405	0	0

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.002834	0.004575	0.005666	0.005064	0.005111	0.004906
LysoPC 16:0	0.000362	0.000811	0.003568	0.002819	0.001259	0.002182
LysoPC 18:3	0.007048	0.012038	0.012274	0.009073	0.00772	0.013235
LysoPC 18:2	0.000614	0.000842	0.002784	0.003051	0.003487	0.004242
LysoPC 18:1	0.002609	0.002148	0.00407	0.00485	0.003599	0.002386
LysoPC 18:0	0.00066	0.001219	0.000781	0.001322	0.001695	0.002037
Total LysoPC	0	0	0.003212	0.005831	0.004019	0.007324

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.01362	0.007745	0.01153	0.003474	0.011521	0.005634
LysoPC 16:0	0	0	0	0	0	0
LysoPC 18:3	0.01271	0.010867	0.003299	0.006408	0.020936	0.025951
LysoPC 18:2	0.0065	0.003679	0.0037	0.005092	0.004616	0.00612
LysoPC 18:1	0.004857	0.003256	0.004316	0.002269	0.002977	0.002016
LysoPC 18:0	0.002102	0.001921	0.000753	0.001599	0.003503	0.005029
Total LysoPC	0.002882	0.003236	0	0	0	0

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.009453	0.007551	0.007836	0.004146	0.006795	0.005055
LysoPC 16:0	0.002161	0.004831	0.000971	0.002171	0.001072	0.001239
LysoPC 18:3	0.01861	0.011223	0.012368	0.008415	0.010804	0.007984
LysoPC 18:2	0.007039	0.006168	0.007708	0.003259	0.008994	0.008119
LysoPC 18:1	0.007048	0.004887	0.00679	0.00597	0.006852	0.006845
LysoPC 18:0	0.01146	0.01868	0.003985	0.005858	0.001697	0.003511
Total LysoPC	0.000532	0.000838	0.013539	0.015027	0.005232	0.003691

	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.039381	0.044702	0.013877	0.002636	0.006044	0.004279
LysoPE 16:0	0.00034	0.00076	0	0	0	0
LysoPE 18:3	0.001105	0.001523	0.004925	0.001794	0.001836	0.001458
LysoPE 18:2	0.000768	0.001165	0.002032	0.001327	0.001617	0.001507
LysoPE 18:1	0.000522	0.001166	0.001428	0.001009	0.00424	0.001591
Total LysoPE	0	0	0.000111	0.000248	0	0

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.032714	0.039175	0.020491	0.013047	0.006784	0.0104
LysoPE 16:0	0	0	0	0	0	0
LysoPE 18:3	0.001294	0.002894	0.003321	0.001762	0.0034	0.000627
LysoPE 18:2	0	0	0.001205	0.001235	0.001125	0.00086
LysoPE 18:1	0.002294	0.002222	0.00273	0.00198	0.001395	0.00156
Total LysoPE	7.6E-05	0.00017	0.000158	0.000219	0.000157	0.000351

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.011293	0.012784	0.026689	0.017962	0.021778	0.021007
LysoPE 16:0	0	0	0	0	0	0
LysoPE 18:3	0.001099	0.001505	0.006133	0.004018	0.003037	0.002357
LysoPE 18:2	0.000353	0.000789	0.003139	0.001211	0.002319	0.001625
LysoPE 18:1	0.001874	0.001813	0.003361	0.000954	0.00251	0.001414
Total LysoPE	0	0	9.83E-05	0.000142	0.000649	0.001452

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.029051	0.015413	0.012068	0.011352	0.032032	0.028641
LysoPE 16:0	0.000228	0.000509	0	0	0	0
LysoPE 18:3	0.003252	0.001738	0.00525	0.003079	0.00384	0.002324
LysoPE 18:2	0.001627	0.001337	0.000143	0.00032	0.000643	0.001168
LysoPE 18:1	0.00172	0.001663	0.003724	0.002112	0.001622	0.001813
Total LysoPE	0	0	0.000184	0.000236	0	0

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.04685	0.031761	0.045361	0.012375	0.03465	0.029025
LysoPE 16:0	0.000165	0.000368	0	0	0	0
LysoPE 18:3	0.008133	0.006929	0.00358	0.00237	0.009192	0.006077
LysoPE 18:2	0.001847	0.001551	0.001692	0.001	0.003727	0.002222
LysoPE 18:1	0.005833	0.005562	0.003015	0.002079	0.003947	0.001767
Total LysoPE	0.000421	0.000766	0	0	0.00082	0.001321

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.003357	0.006579	0.004678	0.003473	0.0021	0.002761
PC 34:4	0.051834	0.028508	0.032689	0.0191	0.02228	0.004285
PC 34:3	0.88975	0.451017	1.613309	0.653884	0.971618	0.344006
PC 34:2	0.898247	0.641924	0.654564	0.495875	0.678628	0.372499
PC 34:1	0.090736	0.077693	0.048445	0.031585	0.045427	0.059129
PC 36:6	0.47531	0.111453	0.750401	0.176675	0.443844	0.078528
PC 36:5	0.977798	0.5762	1.057417	0.440995	1.318183	0.380002
PC 36:4	0.660809	0.509869	0.60586	0.486903	0.651459	0.242262
PC 36:3	0.428587	0.350273	0.247176	0.079189	0.183612	0.131448
PC 36:2	0.152947	0.109928	0.035936	0.028737	0.081362	0.071371
PC 36:1	0.028751	0.03695	0.006455	0.008577	0.008261	0.011337
PC 38:6	0.004844	0.003734	0.005965	0.003687	0.003241	0.002478
PC 38:5	0.010112	0.004363	0.009724	0.008106	0.0084	0.003146
PC 38:4	0.011617	0.012295	0.016321	0.010489	0.014792	0.011654
PC 38:3	0.044804	0.04156	0.013968	0.010852	0.015556	0.004131
PC 38:2	0.013282	0.00817	0.013771	0.011437	0.014432	0.008544
PC 40:5	0.000594	0.000831	0.000975	0.000757	0.000354	0.00059
PC 40:4	0.001272	0.00122	0.001549	0.00116	0.001612	0.001669
PC 40:3	0.002311	0.002218	0.000513	0.000703	0.000591	0.000843
PC 40:2	0.000843	0.001071	0.000562	0.000796	0.000498	0.000784
Total PC	4.747804	2.038642	5.12028	1.297294	4.466251	0.900141
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
	0.006428	0.010772	0.003946	0.006508	0	0
PC 32:0	0.030903	0.016646	0.032	0.011023	0.027219	0.007182
PC 34:3	0.819926	0.665552	0.973517	0.615726	0.931169	0.369665
PC 34:2	0.580718	0.239304	0.720271	0.419677	0.559678	0.178723
PC 34:1	0.074701	0.078049	0.052075	0.037914	0.08345	0.049853
PC 36:6	0.501812	0.312809	0.895375	0.28598	0.450811	0.188184
PC 36:5	1.30405	0.299208	1.374897	0.314159	0.798393	0.495889
PC 36:4	0.610571	0.417143	0.485881	0.256477	0.860024	0.467971
PC 36:3	0.122491	0.106548	0.181904	0.100008	0.153088	0.102151
PC 36:2	0.094082	0.098468	0.082224	0.03163	0.060636	0.052703
PC 36:1	0.0145	0.016746	0.017281	0.015057	0.007785	0.008127
PC 38:6	0.005747	0.002473	0.007438	0.005986	0.00561	0.005361
PC 38:5	0.01535	0.005748	0.00965	0.005224	0.007651	0.003803
PC 38:4	0.024351	0.011175	0.014591	0.011486	0.01809	0.007954
PC 38:3	0.029904	0.020287	0.020895	0.012101	0.017335	0.010234
PC 38:2	0.014632	0.01074	0.019769	0.004065	0.00538	0.007166
PC 40:5	0.002323	0.002359	0.001982	0.002843	0.000475	0.000587
PC 40:4	0.003178	0.005177	0.001497	0.001424	0.00138	0.001893
PC 40:3	0.002664	0.003096	0.003038	0.00247	0.001186	0.001116
PC 40:2	0.000795	0.001276	0.003083	0.003955	0.001767	0.001896
Total PC	4.259125	1.727267	4.901314	1.223201	3.991128	1.402829

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.001711	0.003826	0.005872	0.006325	0.00153	0.00216
PC 34:4	0.058516	0.03236	0.034971	0.037419	0.02992	0.012146
PC 34:3	0.884867	0.334273	0.622888	0.240257	0.803772	0.304817
PC 34:2	0.562164	0.378428	0.476964	0.304962	0.3342	0.066045
PC 34:1	0.225861	0.103886	0.158322	0.122158	0.102057	0.088552
PC 36:6	0.497191	0.271564	0.654285	0.49871	0.311705	0.139007
PC 36:5	0.813267	0.095474	1.072783	0.634881	0.633206	0.352896
PC 36:4	0.991928	0.433231	0.594757	0.858377	0.706959	0.39374
PC 36:3	0.216623	0.137891	0.419061	0.463105	0.1834	0.198813
PC 36:2	0.082927	0.124927	0.063148	0.069805	0.042434	0.019144
PC 36:1	0.014258	0.015943	0.011762	0.008214	0.014563	0.014551
PC 38:6	0.00343	0.001121	0.005111	0.004614	0.005155	0.003037
PC 38:5	0.011457	0.003497	0.010459	0.008605	0.007522	0.003098
PC 38:4	0.023531	0.018708	0.022784	0.022611	0.02115	0.012856
PC 38:3	0.037119	0.024151	0.035161	0.015064	0.01814	0.017757
PC 38:2	0.019363	0.012302	0.01187	0.019126	0.010121	0.006625
PC 40:5	0.001231	0.001242	0.000886	0.000999	0.002471	0.001903
PC 40:4	0.001241	0.00132	0.002692	0.002183	0.002411	0.002812
PC 40:3	0.001714	0.002004	0.00367	0.005126	0.00248	0.004339
PC 40:2	0.001838	0.002029	0.004713	0.004041	0.001634	0.001533
Total PC	4.450239	1.324436	4.212159	2.844026	3.23483	1.24118
Col-o 24hr mock		101919 24hr mock		082589 24hr mock		
Sample description	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002781	0.006219	0.000502	0.00069	0.005576	0.007719
PC 34:4	0.064277	0.068025	0.038435	0.025571	0.030669	0.029967
PC 34:3	1.324377	1.467316	2.791835	2.363042	0.882735	0.970878
PC 34:2	0.924261	0.962753	1.1636	1.382924	0.477111	0.334892
PC 34:1	0.158894	0.156241	0.127955	0.0597	0.064873	0.085305
PC 36:6	0.795341	0.892492	0.751475	0.467586	0.556904	0.963009
PC 36:5	1.420151	1.599298	1.219802	0.570563	1.265872	1.353393
PC 36:4	1.146826	0.915128	0.436767	0.400285	0.864179	0.476097
PC 36:3	0.502944	0.460937	0.203558	0.108786	0.365869	0.414729
PC 36:2	0.138686	0.181692	0.127237	0.088414	0.087802	0.117075
PC 36:1	0.026069	0.017022	0.004213	0.007811	0.012713	0.005332
PC 38:6	0.014182	0.014994	0.006186	0.004671	0.003269	0.003119
PC 38:5	0.014147	0.013329	0.010905	0.010073	0.008376	0.005459
PC 38:4	0.0407	0.035713	0.020978	0.011431	0.032029	0.02555
PC 38:3	0.022975	0.01989	0.021829	0.004331	0.030837	0.049021
PC 38:2	0.024602	0.019172	0.014745	0.006849	0.020118	0.02012
PC 40:5	0.00261	0.005364	0.003746	0.001644	0.000637	0.001424
PC 40:4	0.004211	0.004139	0.003999	0.001533	0.006415	0.004921
PC 40:3	0.01207	0.023743	0.002323	0.001792	0.003932	0.006025
PC 40:2	0.004963	0.002167	0.004075	0.007308	0.00328	0.004681
Total PC	6.64507	6.664304	6.954165	4.396466	4.723197	4.406876

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.005242	0.003856	0.005385	0.004883	0.005135	0.00134
PC 34:4	0.04538	0.024039	0.035568	0.011362	0.031933	0.015282
PC 34:3	0.887757	0.184037	0.753259	0.425711	0.983623	0.289328
PC 34:2	0.635003	0.346479	0.453241	0.266812	0.527792	0.263679
PC 34:1	0.103858	0.069932	0.066314	0.063503	0.165156	0.099701
PC 36:6	0.387129	0.28088	0.656693	0.397652	0.709927	0.189815
PC 36:5	0.977644	0.713204	0.959516	0.438743	0.859693	0.439589
PC 36:4	0.5686	0.479239	0.821068	0.711828	0.634862	0.175842
PC 36:3	0.418551	0.158721	0.345318	0.233283	0.28435	0.129969
PC 36:2	0.057817	0.053564	0.07663	0.030062	0.071037	0.042808
PC 36:1	0.016939	0.016445	0.008875	0.010905	0.007647	0.007727
PC 38:6	0.009578	0.004933	0.006578	0.001639	0.005569	0.002049
PC 38:5	0.012145	0.009668	0.014143	0.011063	0.009669	0.004392
PC 38:4	0.015835	0.010839	0.022762	0.024077	0.020736	0.014238
PC 38:3	0.028181	0.017063	0.023478	0.017393	0.01579	0.006179
PC 38:2	0.019493	0.007854	0.015785	0.011855	0.015076	0.00837
PC 40:5	0.001619	0.002171	0.002848	0.001362	0.001468	0.000845
PC 40:4	0.008569	0.010018	0.002131	0.001392	0.002087	0.002051
PC 40:3	0.002729	0.002394	0.00387	0.004172	0.001973	0.001469
PC 40:2	0.008093	0.012731	0.004585	0.003115	0.002869	0.002071
Total PC	4.21016	1.776265	4.278048	1.975751	4.356394	1.104677

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	4.747804	2.038642	5.12028	1.297294	4.466251	0.900141
PE 34:3	0.007351	0.004396	0.007134	0.002529	0.007586	0.00194
PE 34:2	0.408836	0.022871	0.753227	0.090076	0.48399	0.077455
PE 34:1	0.657767	0.083985	0.671654	0.058029	0.547815	0.051909
PE 36:6	0.016142	0.015765	0.010159	0.005798	0.020644	0.011716
PE 36:5	0.13123	0.014511	0.217144	0.031105	0.15884	0.0216
PE 36:4	0.418992	0.050603	0.4857	0.053754	0.383088	0.056854
PE 36:3	0.349596	0.041948	0.335646	0.055709	0.283289	0.044566
PE 36:2	0.093289	0.029223	0.079338	0.016794	0.079096	0.019924
PE 36:1	0.03953	0.007159	0.039728	0.002942	0.036335	0.008078
PE 38:6	0.001857	0.001989	0.001271	0.00123	0.001798	0.000808
PE 38:5	0.000381	0.000851	0.002949	0.002365	0.002394	0.001329
PE 38:4	0.003414	0.002838	0.004796	0.001055	0.004008	0.001898
PE 38:3	0.002706	0.002881	0.003683	0.001615	0.003491	0.001128
PE 38:2	0.005406	0.003925	0.010548	0.003632	0.007794	0.003226
PE 40:3	0.007245	0.00329	0.009976	0.005401	0.01164	0.001517
PE 40:2	0.002878	0.002768	0.005221	0.002539	0.005185	0.001759
PE 42:4	0.014052	0.008212	0.015871	0.004275	0.014168	0.003227
PE 42:3	0.00093	0.001343	0.002656	0.001405	0.003006	0.002253

PE 42:2	0.01069	0.004114	0.016139	0.00472	0.010363	0.003299
Total PE	0.006866	0.005337	0.009988	0.005804	0.010148	0.002178

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	4.259125	1.727267	4.901314	1.223201	3.991128	1.402829
PE 34:3	0.006544	0.00337	0.007039	0.000688	0.006515	0.001533
PE 34:2	0.469674	0.058536	0.580303	0.101217	0.49369	0.053559
PE 34:1	0.650039	0.117045	0.536011	0.029753	0.601821	0.094556
PE 36:6	0.016352	0.005497	0.01521	0.009907	0.01258	0.005328
PE 36:5	0.127228	0.022985	0.168135	0.045507	0.160602	0.01324
PE 36:4	0.39623	0.049524	0.388951	0.049499	0.404543	0.073639
PE 36:3	0.335913	0.045725	0.268617	0.02753	0.319575	0.035872
PE 36:2	0.07766	0.009586	0.067299	0.004912	0.076844	0.012756
PE 36:1	0.041879	0.010628	0.033234	0.006451	0.035391	0.008073
PE 38:6	0.001697	0.001002	0.001003	0.001337	0.000704	0.000645
PE 38:5	0.000977	0.000917	0.00377	0.000948	0.00209	0.001422
PE 38:4	0.003499	0.002513	0.002975	0.001854	0.003201	0.001804
PE 38:3	0.002171	0.003713	0.001867	0.001373	0.004559	0.001906
PE 38:2	0.00394	0.002959	0.006161	0.00221	0.007843	0.003488
PE 40:3	0.006103	0.003234	0.010755	0.004936	0.010782	0.002968
PE 40:2	0.002627	0.001362	0.00434	0.002075	0.004052	0.003071
PE 42:4	0.009097	0.005491	0.010235	0.004211	0.011467	0.002914
PE 42:3	0.000933	0.001513	0.001995	0.001519	0.002155	0.001061
PE 42:2	0.012414	0.005091	0.009156	0.003932	0.011821	0.003485
Total PE	0.006432	0.005457	0.006203	0.003902	0.008258	0.002247

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	4.450239	1.324436	4.212159	2.844026	3.23483	1.24118
PE 34:3	0.008411	0.001197	0.008152	0.001847	0.00823	0.001369
PE 34:2	0.473336	0.029554	0.447275	0.0713	0.47498	0.111591
PE 34:1	0.661236	0.044949	0.500383	0.085196	0.570149	0.074663
PE 36:6	0.029353	0.006721	0.020709	0.008796	0.018288	0.007936
PE 36:5	0.148931	0.017303	0.16327	0.021788	0.168917	0.044293
PE 36:4	0.431567	0.044308	0.35136	0.052523	0.394875	0.083957
PE 36:3	0.396372	0.024692	0.273396	0.040364	0.309628	0.044113
PE 36:2	0.097866	0.006054	0.074615	0.017337	0.080143	0.020612
PE 36:1	0.039762	0.006031	0.028315	0.002434	0.03112	0.00587
PE 38:6	0.001254	0.001775	0.00139	0.000932	0.001491	0.001559
PE 38:5	0.002013	0.001375	0.002591	0.001021	0.002783	0.000649
PE 38:4	0.004823	0.001251	0.003419	0.001637	0.004648	0.002349
PE 38:3	0.005454	0.001768	0.003515	0.0007	0.004926	0.001943
PE 38:2	0.007457	0.002092	0.007142	0.003222	0.006656	0.002839
PE 40:3	0.008265	0.003911	0.009943	0.001799	0.008941	0.004816
PE 40:2	0.003909	0.002287	0.00369	0.00152	0.004456	0.002318
PE 42:4	0.017036	0.006469	0.012505	0.001189	0.011229	0.005331

PE 42:3	0.001971	0.001347	0.001966	0.001184	0.00251	0.001316
PE 42:2	0.013393	0.002948	0.009268	0.001197	0.008744	0.003203
Total PE	0.01096	0.002762	0.00781	0.002032	0.008	0.001423
	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
PE 34:4	6.64507	6.664304	6.954165	4.396466	4.723197	4.406876
PE 34:3	0.008094	0.002397	0.004944	0.002132	0.004678	0.00191
PE 34:2	0.483934	0.089118	0.570891	0.08895	0.397006	0.0782
PE 34:1	0.620541	0.074159	0.661707	0.139722	0.53847	0.058934
PE 36:6	0.024109	0.012898	0.011217	0.002788	0.01991	0.008905
PE 36:5	0.164311	0.019694	0.163517	0.020659	0.148079	0.023463
PE 36:4	0.419384	0.051428	0.387716	0.046138	0.38722	0.066615
PE 36:3	0.339522	0.049249	0.282009	0.057238	0.316691	0.05593
PE 36:2	0.096224	0.025649	0.073226	0.015339	0.089498	0.01019
PE 36:1	0.045045	0.005494	0.037394	0.007018	0.036812	0.005882
PE 38:6	0.002535	0.00217	0.000798	0.000861	0.003012	0.001919
PE 38:5	0.001192	0.001389	0.002336	0.001002	0.000263	0.000587
PE 38:4	0.003247	0.001802	0.003053	0.002152	0.002969	0.001452
PE 38:3	0.004172	0.0021	0.002859	0.00134	0.005041	0.00154
PE 38:2	0.005617	0.00264	0.007794	0.003342	0.005831	0.002706
PE 40:3	0.008355	0.004578	0.011138	0.00317	0.006286	0.003938
PE 40:2	0.004591	0.002536	0.004472	0.001949	0.003299	0.002145
PE 42:4	0.013282	0.003099	0.015021	0.005186	0.017468	0.002853
PE 42:3	0.001052	0.000764	0.002235	0.001858	0.001593	0.001814
PE 42:2	0.012703	0.003175	0.011202	0.001555	0.008965	0.007031
Total PE	0.009735	0.003461	0.006719	0.002901	0.002499	0.003597
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
PE 34:4	4.21016	1.776265	4.278048	1.975751	4.356394	1.104677
PE 34:3	0.011431	0.002111	0.008939	0.001696	0.008851	0.003212
PE 34:2	0.492076	0.046461	0.567307	0.126014	0.560554	0.037347
PE 34:1	0.612962	0.071632	0.586089	0.108457	0.543695	0.05744
PE 36:6	0.037782	0.007884	0.023207	0.006263	0.029878	0.012426
PE 36:5	0.171976	0.008256	0.194722	0.04162	0.198562	0.024424
PE 36:4	0.444919	0.040403	0.423876	0.064697	0.448606	0.073903
PE 36:3	0.410437	0.037593	0.33016	0.048312	0.333436	0.05984
PE 36:2	0.127977	0.012011	0.096793	0.014726	0.10295	0.026119
PE 36:1	0.043788	0.008997	0.033409	0.00912	0.034768	0.010896
PE 38:6	0.001516	0.001987	0.001343	0.000929	0.002129	0.002523
PE 38:5	0.001987	0.001562	0.002411	0.000554	0.00304	0.001971
PE 38:4	0.004981	0.002616	0.004584	0.000642	0.006369	0.00128
PE 38:3	0.005652	0.002082	0.004395	0.001472	0.003741	0.00346
PE 38:2	0.008069	0.001132	0.009216	0.003291	0.008746	0.000863
PE 40:3	0.0089	0.001633	0.0102	0.001787	0.007589	0.003776
PE 40:2	0.008989	0.003907	0.006329	0.002279	0.00691	0.001787
PE 42:4	0.015511	0.00608	0.014116	0.001786	0.014561	0.00248
PE 42:3	0.002072	0.001457	0.002422	0.001629	0.002497	0.00227
PE 42:2	0.014009	0.002779	0.014105	0.003315	0.013792	0.003904

Total PE	0.013039	0.004042	0.014206	0.001911	0.01177	0.00208
Sample description	Col-o 0hr	101919 0hr		082589 0hr		
PI 34:4	ave 2.179159	stdev 0.163717	ave 2.682827	stdev 0.151834	ave 2.074679	stdev 0.107864
PI 34:3	0	0	0.000377	0.000567	0.001923	0.001914
PI 34:2	0.482868	0.138707	0.736143	0.222229	0.514469	0.120224
PI 34:1	0.421489	0.105299	0.401258	0.068168	0.339678	0.073144
PI 36:6	0.005265	0.009974	0.004647	0.006733	0.001524	0.003409
PI 36:5	0.009164	0.00704	0.020516	0.006283	0.021095	0.007758
PI 36:4	0.005751	0.008073	0.019217	0.006686	0.019739	0.009701
PI 36:3	0	0	0.003393	0.003153	0.011778	0.00552
PI 36:2	0.011057	0.016409	0.011461	0.007395	0.021029	0.005421
PI 36:1	0.005029	0.006521	0.008653	0.003558	0.009455	0.006882
Total PI	0	0	0.009922	0.002885	0.009956	0.007584
Sample description	Col-o 12hr mock	101919 12hr mock	082589 12hr mock			
PI 34:4	ave 2.171409	stdev 0.220618	ave 2.123259	stdev 0.196796	ave 2.178493	stdev 0.249474
PI 34:3	0	0	0	0	0	0
PI 34:2	0.441854	0.13204	0.638414	0.125735	0.467498	0.11218
PI 34:1	0.412948	0.082686	0.371552	0.056245	0.298255	0.122533
PI 36:6	0.012331	0.012045	0.004925	0.004845	0.000728	0.001628
PI 36:5	0.007061	0.005749	0.020209	0.006788	0.015358	0.006795
PI 36:4	0.019145	0.005387	0.015681	0.00359	0.015007	0.004795
PI 36:3	0.003779	0.00487	0.005578	0.005149	0.007779	0.00376
PI 36:2	0.005168	0.006285	0.003647	0.005171	0.013387	0.010563
PI 36:1	0.007781	0.006417	0.002937	0.004092	0.008136	0.003447
Total PI	0.005485	0.004189	0.007096	0.006773	0.001313	0.002676
Sample description	Col-o 12hr SA	101919 12hr SA	082589 12hr SA			
PI 34:4	ave 2.363369	stdev 0.133859	ave 1.930715	stdev 0.18661	ave 2.120715	stdev 0.317604
PI 34:3	0	0	0.003402	0.003509	0.002463	0.002736
PI 34:2	0.505572	0.054374	0.521375	0.130229	0.477134	0.087628
PI 34:1	0.382867	0.046811	0.328533	0.056259	0.294557	0.028842
PI 36:6	0.016483	0.020864	0.005261	0.007234	0.005678	0.007753
PI 36:5	0.036994	0.015765	0.036328	0.004545	0.034135	0.007114
PI 36:4	0.03988	0.012382	0.039496	0.008216	0.043194	0.008409
PI 36:3	0.027009	0.010276	0.024642	0.006986	0.020146	0.00665
PI 36:2	0.019547	0.011491	0.023128	0.006151	0.021075	0.003506
PI 36:1	0.01217	0.008149	0.009533	0.003551	0.014397	0.004928
Total PI	0.002983	0.002775	0.016624	0.007622	0.013899	0.004641

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	2.267646	0.236981	2.26025	0.200618	1.99559	0.175241
PI 34:3	0	0	0.000431	0.000964	0	0
PI 34:2	0.497348	0.17497	0.589143	0.201962	0.423656	0.215009
PI 34:1	0.35343	0.108718	0.422274	0.132059	0.282252	0.164582
PI 36:6	0.006871	0.015365	0.006941	0.009672	0.027965	0.006346
PI 36:5	0.01189	0.013096	0.016555	0.010084	0.013434	0.005242
PI 36:4	0.007822	0.011179	0.017594	0.0092	0.008018	0.006727
PI 36:3	0.006832	0.011242	0.007796	0.012135	0.004888	0.004995
PI 36:2	0.010156	0.010679	0.009774	0.004967	0.006059	0.008966
PI 36:1	0.005001	0.007005	0.006458	0.005971	0.003123	0.004813
Total PI	0.000422	0.000945	0.000379	0.000848	0.003972	0.005878
Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	2.438074	0.17003	2.34783	0.375951	2.342444	0.201093
PI 34:3	0.000855	0.001911	0.000143	0.00032	0	0
PI 34:2	0.47925	0.131254	0.482843	0.0683	0.653382	0.166411
PI 34:1	0.369165	0.127251	0.310431	0.058908	0.359497	0.09776
PI 36:6	0.013681	0.020232	0.018289	0.018535	0.010143	0.013889
PI 36:5	0.024447	0.012145	0.027406	0.00772	0.028507	0.012964
PI 36:4	0.033026	0.011009	0.022873	0.004842	0.014107	0.012919
PI 36:3	0.014758	0.00935	0.015216	0.011063	0.011215	0.006684
PI 36:2	0.02106	0.010908	0.01714	0.00432	0.00789	0.004695
PI 36:1	0.005191	0.00718	0.010458	0.004947	0.002725	0.004379
Total PI	0.006425	0.004941	0.004193	0.003984	0.008019	0.0079

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.940621	0.205646	1.215587	0.24552	0.950647	0.191577
PS 34:3	0	0	0	0	0	0
PS 34:2	0.030588	0.011221	0.031487	0.00501	0.023665	0.010029
PS 34:1	0.020246	0.014257	0.019532	0.001548	0.017002	0.006576
PS 36:6	0	0	7.6E-05	0.00017	0	0
PS 36:5	0	0	0.000166	0.00037	0.00029	0.000398
PS 36:4	0	0	0.000833	0.000635	0.00094	0.001027
PS 36:3	0	0	0.00095	0.000965	0.001431	0.001351
PS 36:2	0.002246	0.002343	0.006346	0.003179	0.005672	0.004632
PS 36:1	0.002912	0.003988	0.005206	0.002059	0.003964	0.002934
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	8.14E-05	0.000115	0	0
PS 38:3	0	0	0	0	0	0
PS 38:2	0	0	0.006272	0.002428	0.003496	0.002754
PS 38:1	0.000822	0.001837	0.009854	0.002417	0.008525	0.004786
PS 40:4	0	0	0.000195	0.000437	7.54E-05	0.000169
PS 40:3	0	0	0.000208	0.000302	0.000138	0.000308
PS 40:2	0.001717	0.001671	0.023268	0.003579	0.016865	0.009777
PS 40:1	0.009469	0.003203	0.017098	0.002929	0.019459	0.00873
PS 42:4	0	0	0.000404	0.000594	0.000592	0.001148
PS 42:3	0	0	0.016228	0.001967	0.006937	0.007228
PS 42:2	0.01439	0.012743	0.062648	0.013828	0.04669	0.022235
PS 42:1	0.005892	0.007555	0.036274	0.005674	0.025804	0.015976
PS 44:3	0	0	0	0	0.000283	0.000633
PS 44:2	0	0	0.000687	0.001221	0.000734	0.001233
Total PS	0	0	0.00103	0.001412	0.000375	0.000839

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.915551	0.155482	1.07004	0.126767	0.827461	0.227387
PS 34:3	0	0	0	0	0	0
PS 34:2	0.016575	0.00997	0.023158	0.003694	0.012576	0.007167
PS 34:1	0.012661	0.006472	0.013076	0.0059	0.009554	0.002018
PS 36:6	0	0	0	0	0	0
PS 36:5	0	0	0.000103	0.000231	0	0
PS 36:4	0	0	0.000286	0.00064	0.000468	0.000691
PS 36:3	0	0	0.000321	0.000718	0.000118	0.000263
PS 36:2	0.001978	0.002155	0.003523	0.0037	0.00382	0.003089
PS 36:1	0.002017	0.003187	0.003317	0.002307	0.002055	0.002825
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0	0	0	0	0	0
PS 38:2	0.000429	0.000958	0.00403	0.003895	0.002166	0.004257
PS 38:1	0.003427	0.002742	0.003933	0.003569	0.003045	0.003063
PS 40:4	0	0	0	0	0	0
PS 40:3	0	0	0	0	0	0
PS 40:2	0.009545	0.008049	0.0137	0.00467	0.010204	0.005253
PS 40:1	0.007297	0.004731	0.015006	0.004509	0.007112	0.004338
PS 42:4	0.000228	0.000509	0.000137	0.000306	0.00012	0.000268
PS 42:3	0.000753	0.001683	0.010015	0.004392	0.003631	0.004383
PS 42:2	0.022665	0.01328	0.045479	0.013448	0.024901	0.018579
PS 42:1	0.010816	0.009047	0.025631	0.015372	0.020239	0.007868
PS 44:3	0	0	0	0	0	0
PS 44:2	0	0	0.000534	0.000767	0	0
Total PS	0	0	0.000562	0.000839	0	0

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	1.043506	0.130697	1.008322	0.155957	0.926679	0.124394
PS 34:3	0	0	0	0	0	0
PS 34:2	0.016492	0.004107	0.021145	0.003714	0.020119	0.005271
PS 34:1	0.016411	0.003285	0.013366	0.003333	0.015138	0.002244
PS 36:6	0	0	0	0	0	0
PS 36:5	9.08E-05	0.000203	0	0	0.000148	0.00033
PS 36:4	0.000807	0.001111	0.001178	0.001108	0.000849	0.001038
PS 36:3	0.001589	0.001206	0.000881	0.00128	0.000388	0.000462
PS 36:2	0.003864	0.000792	0.005187	0.002166	0.005168	0.002834
PS 36:1	0.004523	0.001193	0.002687	0.002072	0.005119	0.003464
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0	0	0	0	0	0
PS 38:2	0.002434	0.002787	0.003962	0.002518	0.00538	0.004105
PS 38:1	0.00485	0.003033	0.005896	0.00227	0.006208	0.002216
PS 40:4	0	0	0.000574	0.001284	0.000134	0.000125
PS 40:3	0	0	9.6E-05	0.000215	0.000175	0.000392
PS 40:2	0.021958	0.007132	0.020801	0.003076	0.01794	0.004659
PS 40:1	0.017145	0.004897	0.01488	0.003089	0.020521	0.00812
PS 42:4	3.05E-05	6.83E-05	0.000753	0.000793	0.00018	0.000255
PS 42:3	0.002811	0.001932	0.004984	0.00267	0.005878	0.001878
PS 42:2	0.038881	0.009585	0.041826	0.008411	0.041981	0.008731
PS 42:1	0.027828	0.010015	0.020058	0.009598	0.028439	0.00763
PS 44:3	0	0	0.000435	0.000972	7.44E-05	0.000166
PS 44:2	0	0	0.00018	0.000403	9.71E-05	0.000217
Total PS	0.000178	0.000399	0.000688	0.000949	0.000277	0.000619

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.899773	0.275194	1.077346	0.314711	0.773367	0.385091
PS 34:3	0	0	0	0	0	0
PS 34:2	0.017694	0.009113	0.022126	0.007033	0.011341	0.00887
PS 34:1	0.017307	0.007845	0.015665	0.004723	0.012794	0.008952
PS 36:6	0	0	0	0	0	0
PS 36:5	0	0	0.00035	0.000494	0	0
PS 36:4	0.000525	0.001173	0.000518	0.000581	0	0
PS 36:3	0.000572	0.001279	0.000182	0.000406	0	0
PS 36:2	0.003113	0.0046	0.006616	0.001521	0.002332	0.003268
PS 36:1	0.001794	0.003292	0.004978	0.002014	0.001004	0.001628
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	1.6E-05	3.58E-05	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0	0	0	0	0	0
PS 38:2	0	0	0.005031	0.003231	0.001445	0.001409
PS 38:1	0.002926	0.002936	0.008718	0.003497	0.000942	0.00144
PS 40:4	0	0	0.00029	0.000648	0	0
PS 40:3	0	0	5.95E-05	0.000133	0	0
PS 40:2	0.002746	0.005106	0.015922	0.005421	0.006289	0.006142
PS 40:1	0.002483	0.002829	0.013801	0.006297	0.006802	0.006687
PS 42:4	0	0	0	0	0	0
PS 42:3	0.001306	0.002037	0.008769	0.007366	0.000612	0.001368
PS 42:2	0.033162	0.019604	0.04842	0.014457	0.018991	0.011711
PS 42:1	0.009812	0.009709	0.024156	0.012166	0.009339	0.010336
PS 44:3	0	0	0	0	0	0
PS 44:2	0	0	0.000221	0.000494	0	0
Total PS	0	0	0.000251	0.000562	0	0

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.967858	0.257038	0.908993	0.128703	1.095484	0.266765
PS 34:3	0	0	0	0	0	0
PS 34:2	0.008503	0.005374	0.016043	0.004577	0.010628	0.010047
PS 34:1	0.012302	0.005003	0.01036	0.002914	0.006488	0.005896
PS 36:6	0	0	0	0	0.000247	0.000553
PS 36:5	0.000123	0.000274	0	0	7.16E-05	0.00016
PS 36:4	0.000461	0.001031	7.21E-05	0.000161	8.06E-05	0.00018
PS 36:3	0.000235	0.000525	0.0006	0.000627	6.5E-05	0.000145
PS 36:2	0.00179	0.001835	0.001925	0.002371	0.000914	0.002045
PS 36:1	0.000911	0.001248	0.002199	0.001301	0.00057	0.001052
PS 38:6	0	0	0	0	0	0
PS 38:5	0	0	0	0	0	0
PS 38:4	0	0	0	0	0	0
PS 38:3	0	0	0	0	0	0
PS 38:2	0.001076	0.00196	0.001188	0.000547	0.001103	0.001206
PS 38:1	0.00351	0.004038	0.003092	0.001156	0.001633	0.001961
PS 40:4	0	0	0	0	0	0
PS 40:3	0	0	8.17E-05	0.000183	0	0
PS 40:2	0.012387	0.008027	0.012224	0.003513	0.008404	0.008772
PS 40:1	0.012366	0.005706	0.01278	0.00787	0.007505	0.00626
PS 42:4	0.000327	0.00073	0.000308	0.000422	4.61E-05	0.000103
PS 42:3	0.002729	0.00237	0.003036	0.003509	0.004394	0.004861
PS 42:2	0.038149	0.015161	0.038365	0.005087	0.037852	0.019039
PS 42:1	0.026093	0.01837	0.022397	0.010642	0.011708	0.013605
PS 44:3	0.000277	0.00062	0	0	0	0
PS 44:2	0.000554	0.001238	0	0	0	0
Total PS	0	0	0	0	0	0

	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.088284	0.042528	0.238842	0.028013	0.182936	0.093161
PA 34:4	0.003902	0.004722	0.000523	0.001169	0	0
PA 34:3	0	0	0	0	0	0
PA 34:2	0.022241	0.018614	0.013191	0.010128	0.008441	0.005077
PA 34:1	0.005495	0.003659	0.000587	0.001312	0	0
PA 36:6	0.024778	0.055405	0.003473	0.007765	0	0
PA 36:5	0.004857	0.006605	0.001905	0.002475	0	0
PA 36:4	0.002736	0.003823	0.003026	0.004976	0	0
PA 36:3	0.005879	0.005598	0.001524	0.003408	0	0
PA 36:2	0.001896	0.002648	0.000189	0.000422	0	0
Total PA	0	0	0.000582	0.001302	0.003185	0.007123
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.08839	0.02372	0.162811	0.058152	0.100009	0.047729
PA 34:4	0.008567	0.017231	0	0	0.000771	0.001724
PA 34:3	0	0	0	0	0	0
PA 34:2	0.023291	0.022898	0.01777	0.036599	0.009614	0.008281
PA 34:1	0.010955	0.024496	0.000285	0.000638	0	0
PA 36:6	0	0	0	0	0.015119	0.018927
PA 36:5	0.006335	0.010423	0.00292	0.004005	0.003454	0.00385
PA 36:4	0.00656	0.012178	0.000844	0.001887	0.002065	0.004617
PA 36:3	0.001517	0.003391	0.002295	0.005132	0.003309	0.007399
PA 36:2	0.002256	0.005045	0.002872	0.006421	0	0
Total PA	0.002937	0.006567	0.002951	0.006599	0	0
	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.159891	0.027662	0.159578	0.02287	0.174212	0.040481
PA 34:4	0	0	0	0	0	0
PA 34:3	0	0	0	0	0	0
PA 34:2	0.003398	0.007598	0.001523	0.003405	0.008725	0.016521
PA 34:1	0	0	0	0	0	0
PA 36:6	0.013489	0.019615	0.023228	0.023959	0.030132	0.045471
PA 36:5	0.001125	0.002515	0	0	0	0
PA 36:4	0	0	0.001569	0.003508	0	0
PA 36:3	0.002029	0.004536	0	0	0	0
PA 36:2	0	0	0	0	0	0
Total PA	0	0	0.000341	0.000762	0	0

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.09344	0.05134	0.176089	0.061734	0.071893	0.04557
PA 34:4	0	0	0	0	0.002271	0.005078
PA 34:3	0	0	0	0	0	0
PA 34:2	0.012053	0.014171	0.003948	0.007744	0.021098	0.013915
PA 34:1	0.008502	0.012348	0.000798	0.001784	0.008873	0.010622
PA 36:6	0	0	0.000506	0.001132	0	0
PA 36:5	0.01567	0.019296	0.004614	0.00767	0.015311	0.012535
PA 36:4	0.006162	0.009169	0.001724	0.003856	0.004835	0.007413
PA 36:3	0.005541	0.005934	0.000799	0.001787	0.00586	0.007541
PA 36:2	0.006322	0.007628	0	0	0	0
Total PA	0	0	0.00094	0.002102	0	0
Total	0.071785	0.075486	0.024999	0.011798	0.011626	0.010673
Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.12179	0.053768	0.12467	0.02931	0.09171	0.070991
PA 34:4	0.014512	0.026106	0.009534	0.011457	0.014586	0.021098
PA 34:3	0.003664	0.005324	0	0	0.009882	0.016881
PA 34:2	0.031745	0.025195	0.025067	0.013522	0.063227	0.064908
PA 34:1	0.016839	0.017435	0.00273	0.0038	0.019177	0.029306
PA 36:6	0	0	0	0	0	0
PA 36:5	0.014362	0.007458	0.012424	0.004239	0.024196	0.025627
PA 36:4	0.011143	0.015582	0.002325	0.00325	0.016436	0.022571
PA 36:3	0.01987	0.020378	0.002215	0.003187	0.00818	0.015149
PA 36:2	0.001906	0.004263	0.001002	0.002241	0.004418	0.009879
Total PA	0.007491	0.010534	0.000622	0.00139	0	0

TABLE A – 6. TOTAL LIPID SPECIES – ESI MS/MS EXPERIMENT 3

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.137783	0.085695	1.186871	0.031263	1.12896	0.052654
DGDG 34:5	0.124925	0.009229	0.135578	0.007215	0.138534	0.003497
DGDG 34:4	0.087521	0.00681	0.079112	0.004423	0.086521	0.004795
DGDG 34:3	2.160198	0.173329	2.008555	0.053123	1.951002	0.103884
DGDG 34:2	0.24929	0.024489	0.216096	0.00517	0.199986	0.010035
DGDG 34:1	0.10564	0.012738	0.08243	0.005071	0.081907	0.005207
DGDG 36:6	7.33952	0.383409	7.508176	0.317989	7.395837	0.14356
DGDG 36:5	0.200034	0.020713	0.190492	0.020768	0.213575	0.006807
DGDG 36:4	0.159979	0.010429	0.144984	0.019165	0.168794	0.016882
DGDG 36:3	0.074655	0.004118	0.069849	0.001811	0.069321	0.003571
DGDG 36:2	0.004578	0.000831	0.004248	0.000742	0.004433	0.000795
DGDG 36:1	0.000689	0.000214	0.000593	0.000254	0.000679	0.000425
DGDG 38:6	0.025457	0.002483	0.025299	0.001704	0.028688	0.002155
DGDG 38:5	0.003591	0.000695	0.004206	0.00039	0.004881	0.000682
DGDG 38:4	0.001522	0.000238	0.001728	0.000478	0.001661	0.000341
DGDG 38:3	0.001235	0.000177	0.00117	0.000212	0.001385	0.000169
Total DGDG	11.67662	0.5897	11.65939	0.41197	11.47616	0.286839
Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.15013	0.081699	1.125466	0.106765	1.142225	0.085294
DGDG 34:5	0.123815	0.002934	0.128098	0.010762	0.142899	0.010041
DGDG 34:4	0.081749	0.006172	0.084112	0.006505	0.085627	0.008572
DGDG 34:3	2.030059	0.040995	2.08971	0.10667	2.090977	0.076047
DGDG 34:2	0.216844	0.019416	0.225804	0.018705	0.208216	0.010668
DGDG 34:1	0.089914	0.010545	0.085813	0.006022	0.073511	0.006341
DGDG 36:6	7.098158	0.217538	7.475859	0.28851	7.890025	0.097437
DGDG 36:5	0.180562	0.012496	0.203864	0.012668	0.21864	0.020113
DGDG 36:4	0.153864	0.007824	0.158009	0.010094	0.155305	0.013102
DGDG 36:3	0.067524	0.005343	0.074641	0.003445	0.077125	0.011115
DGDG 36:2	0.004116	0.000957	0.004561	0.000478	0.004337	0.000652
DGDG 36:1	0.000729	0.000188	0.000668	0.000192	0.00055	0.000446
DGDG 38:6	0.023079	0.001175	0.025585	0.001181	0.029037	0.001958
DGDG 38:5	0.003252	0.000447	0.003633	0.000907	0.003998	0.000592
DGDG 38:4	0.001518	0.000364	0.001529	0.000739	0.001611	0.000285
DGDG 38:3	0.000935	0.000147	0.001208	0.000292	0.001416	0.000178
Total DGDG	11.22625	0.210745	11.68856	0.398932	12.1255	0.147887

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample	ave	stdev	ave	stdev	ave	stdev
description						
DGDG 34:6	1.246955	0.109148	1.188983	0.125008	1.094413	0.0675
DGDG 34:5	0.121599	0.005576	0.127412	0.013485	0.128233	0.006718
DGDG 34:4	0.079212	0.009428	0.087948	0.014709	0.084854	0.004628
DGDG 34:3	1.94705	0.149825	1.951501	0.132845	1.88492	0.127578
DGDG 34:2	0.216488	0.013779	0.20492	0.010081	0.199977	0.019939
DGDG 34:1	0.083123	0.011213	0.080372	0.013302	0.082914	0.005771
DGDG 36:6	7.246261	0.495359	7.331506	0.585645	6.991306	0.16556
DGDG 36:5	0.182684	0.026445	0.21596	0.045946	0.220287	0.022551
DGDG 36:4	0.149762	0.013181	0.160304	0.032695	0.160903	0.008012
DGDG 36:3	0.067154	0.008458	0.071136	0.00514	0.067888	0.004332
DGDG 36:2	0.002744	0.000966	0.003921	0.000866	0.004137	0.000531
DGDG 36:1	0.000588	0.000341	0.000573	0.000272	0.000353	0.000213
DGDG 38:6	0.020831	0.002297	0.022851	0.00207	0.025253	0.002731
DGDG 38:5	0.003109	0.000831	0.003745	0.000175	0.004106	0.000719
DGDG 38:4	0.001374	0.000642	0.001618	0.000292	0.001387	0.000114
DGDG 38:3	0.000914	0.000205	0.001289	0.000257	0.001273	0.000274
Total DGDG	11.36985	0.762229	11.45404	0.866621	10.9522	0.401492
	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample	ave	stdev	ave	stdev	ave	stdev
description						
DGDG 34:6	1.040055	0.133435	1.107973	0.207817	0.936766	0.077752
DGDG 34:5	0.118826	0.012775	0.119045	0.019541	0.114755	0.014433
DGDG 34:4	0.079171	0.009261	0.077985	0.009496	0.075936	0.003863
DGDG 34:3	1.938263	0.162796	1.964904	0.115582	1.772829	0.105232
DGDG 34:2	0.222094	0.021202	0.228958	0.022579	0.207011	0.021061
DGDG 34:1	0.097829	0.005511	0.092091	0.011967	0.091008	0.01179
DGDG 36:6	6.87098	0.595794	6.974359	0.202874	6.565165	0.463206
DGDG 36:5	0.204336	0.019082	0.181007	0.014643	0.20856	0.011382
DGDG 36:4	0.151522	0.020523	0.151624	0.012659	0.149269	0.011736
DGDG 36:3	0.07301	0.005349	0.066959	0.006866	0.073023	0.00215
DGDG 36:2	0.004327	0.000623	0.004455	0.000993	0.004215	0.001185
DGDG 36:1	0.000837	0.000432	0.000619	8.99E-05	0.000594	0.000301
DGDG 38:6	0.024558	0.002301	0.022034	0.001801	0.030187	0.007913
DGDG 38:5	0.003612	0.000665	0.003503	0.000551	0.004013	0.000642
DGDG 38:4	0.001164	0.000268	0.001546	0.000296	0.001379	0.000436
DGDG 38:3	0.00122	0.000403	0.001031	0.000125	0.001211	0.000201
Total DGDG	10.83181	0.927648	10.9981	0.506352	10.23592	0.583485

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
DGDG 34:6	1.258662	0.153588	1.221803	0.068191	1.094933	0.097516
DGDG 34:5	0.131375	0.012683	0.119557	0.006712	0.12689	0.007005
DGDG 34:4	0.086494	0.009296	0.082126	0.002349	0.081538	0.00915
DGDG 34:3	1.920618	0.206602	1.933237	0.044062	1.843542	0.1399
DGDG 34:2	0.209949	0.02755	0.203927	0.01516	0.204106	0.011835
DGDG 34:1	0.077968	0.011945	0.073261	0.007364	0.078829	0.005656
DGDG 36:6	7.47958	0.437508	7.437195	0.236104	6.656149	0.345131
DGDG 36:5	0.228407	0.02799	0.230142	0.026222	0.253812	0.011101
DGDG 36:4	0.159328	0.01777	0.14294	0.014016	0.144908	0.017602
DGDG 36:3	0.067148	0.006433	0.070794	0.007332	0.066821	0.001917
DGDG 36:2	0.003631	0.001101	0.003966	0.000723	0.003849	0.000627
DGDG 36:1	0.00044	0.000257	0.00064	0.000239	0.000847	0.000203
DGDG 38:6	0.023813	0.001261	0.023491	0.002167	0.024283	0.001244
DGDG 38:5	0.00375	0.00085	0.003939	0.000358	0.00394	0.000539
DGDG 38:4	0.00145	0.000296	0.00121	0.000155	0.001232	0.000236
DGDG 38:3	0.000986	0.000198	0.001096	0.000173	0.00103	0.000189
Total DGDG	11.6536	0.837544	11.54932	0.280632	10.58671	0.589566
	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	60.62406	0.842075	60.8548	2.019973	58.0746	0.8947
MGDG 34:5	4.40009	0.130033	4.038467	0.481784	4.227078	0.318163
MGDG 34:4	1.590788	0.067134	1.43049	0.140624	1.554903	0.093729
MGDG 34:3	0.669799	0.018842	0.594178	0.03783	0.646278	0.033759
MGDG 34:2	0.189353	0.012725	0.158133	0.017961	0.183791	0.026287
MGDG 34:1	0.082616	0.008392	0.057831	0.008669	0.057867	0.006023
MGDG 36:6	7.772777	0.257254	7.598651	0.387984	8.123252	0.247157
MGDG 36:5	0.512026	0.023865	0.491074	0.042475	0.57345	0.043095
MGDG 36:4	0.170378	0.008663	0.150692	0.017289	0.192186	0.016554
MGDG 36:3	0.022517	0.001053	0.020842	0.000818	0.023247	0.000973
MGDG 36:2	0.001623	0.000548	0.001305	0.000425	0.001934	0.000718
MGDG 36:1	0.000273	0.000172	0.000131	0.000179	0.000142	0.000181
MGDG 38:6	0.007393	0.000901	0.006655	0.000915	0.012173	0.006149
MGDG 38:5	0.002897	0.000188	0.002581	0.000369	0.003036	0.000202
MGDG 38:4	0.002519	0.000428	0.002502	0.000411	0.002745	0.000432
MGDG 38:3	0.000262	0.000117	0.000281	0.000151	0.000442	0.00031
Total MGDG	76.04937	0.632175	75.40861	1.560327	73.67713	0.575734

Sample	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	60.22801	0.482644	58.74053	0.89682	58.35383	0.728365
MGDG 34:5	3.575181	0.408697	3.472171	0.230167	3.441371	0.455701
MGDG 34:4	1.247086	0.110009	1.204155	0.063238	1.115422	0.127412
MGDG 34:3	0.589211	0.042487	0.594104	0.023664	0.614812	0.063111
MGDG 34:2	0.130584	0.014841	0.129191	0.014086	0.115492	0.022558
MGDG 34:1	0.052222	0.003593	0.061297	0.008268	0.047326	0.008806
MGDG 36:6	7.614428	0.173024	8.040946	0.215985	9.222218	0.446106
MGDG 36:5	0.54842	0.028518	0.583615	0.035502	0.6646	0.062184
MGDG 36:4	0.168432	0.006827	0.164316	0.003628	0.182341	0.011232
MGDG 36:3	0.022321	0.001898	0.023946	0.001445	0.025906	0.002645
MGDG 36:2	0.001609	0.000392	0.001616	0.000512	0.001643	0.000389
MGDG 36:1	1.24E-05	2.78E-05	0.000147	0.000145	6.27E-05	0.000108
MGDG 38:6	0.007442	0.001274	0.007506	0.001631	0.013503	0.003843
MGDG 38:5	0.002368	0.000296	0.002796	0.000323	0.003376	0.000264
MGDG 38:4	0.002294	0.000419	0.002296	0.000239	0.00267	0.000213
MGDG 38:3	0.000287	0.000108	0.000207	0.000171	0.000335	0.00027
Total MGDG	74.18991	0.694791	73.02884	0.694134	73.80491	0.360576

Sample	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	62.39451	1.861006	61.53175	1.204617	60.67722	0.811755
MGDG 34:5	3.309324	0.168074	3.514341	0.340628	3.688975	0.345513
MGDG 34:4	0.991138	0.087551	0.975094	0.181282	1.337376	0.035166
MGDG 34:3	0.414711	0.02939	0.450665	0.038858	0.56716	0.030898
MGDG 34:2	0.093564	0.012978	0.094401	0.024088	0.144547	0.017024
MGDG 34:1	0.037328	0.004159	0.043881	0.014175	0.048396	0.003528
MGDG 36:6	7.030781	0.60421	7.621404	0.361098	8.343197	0.36799
MGDG 36:5	0.399115	0.069279	0.491016	0.109857	0.564076	0.059708
MGDG 36:4	0.135605	0.015018	0.151323	0.020332	0.190524	0.010696
MGDG 36:3	0.015958	0.001954	0.016924	0.001988	0.020143	0.002247
MGDG 36:2	0.000968	0.000409	0.001265	0.000195	0.001536	0.000367
MGDG 36:1	8.46E-05	0.000102	1.2E-05	2.68E-05	3.47E-05	6.12E-05
MGDG 38:6	0.006159	0.002299	0.008072	0.001938	0.010438	0.001999
MGDG 38:5	0.002043	0.000559	0.002218	0.000348	0.002848	0.000296
MGDG 38:4	0.00205	0.000194	0.002424	0.000321	0.002415	0.000221
MGDG 38:3	0.000197	0.000183	0.000179	6.72E-05	0.000189	0.000207
Total MGDG	74.83354	1.247923	74.90496	1.078693	75.59907	0.643721

Sample	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	62.1305	1.222663	62.274	0.730607	61.86986	1.960684
MGDG 34:5	3.943845	0.537439	3.812411	0.411036	4.004427	0.433702
MGDG 34:4	1.588329	0.146966	1.457837	0.137591	1.695161	0.144607
MGDG 34:3	0.758643	0.068853	0.724776	0.087082	0.797968	0.050527
MGDG 34:2	0.20035	0.017658	0.187551	0.025149	0.225965	0.027614
MGDG 34:1	0.107641	0.013771	0.104932	0.0236	0.105035	0.023286
MGDG 36:6	7.924385	0.252208	7.594353	0.391283	8.074399	0.593179
MGDG 36:5	0.536263	0.020178	0.51143	0.046674	0.581073	0.04884
MGDG 36:4	0.179118	0.010371	0.167254	0.012533	0.200515	0.021758
MGDG 36:3	0.02307	0.001796	0.021647	0.002668	0.0269	0.003154
MGDG 36:2	0.002026	0.000296	0.00166	0.00052	0.001979	0.000243
MGDG 36:1	0.000297	0.000263	0.000129	0.000164	0.00024	0.000223
MGDG 38:6	0.007763	0.001387	0.005661	0.000636	0.015201	0.010301
MGDG 38:5	0.002649	0.000236	0.002595	0.000184	0.002862	0.00026
MGDG 38:4	0.002175	0.000362	0.002219	0.000258	0.002488	0.000516
MGDG 38:3	0.000248	0.000102	0.000194	7.35E-05	0.000398	0.000155
Total MGDG	77.4073	1.469889	76.86864	0.930207	77.60446	1.513916

Sample	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
description	ave	stdev	ave	stdev	ave	stdev
MGDG 34:6	62.81263	2.147573	62.75215	0.473789	61.53909	1.365952
MGDG 34:5	2.994495	0.42758	2.882514	0.255408	3.555914	0.247607
MGDG 34:4	0.897738	0.072357	0.856155	0.099963	1.234896	0.043505
MGDG 34:3	0.437699	0.044537	0.411311	0.040339	0.531112	0.015103
MGDG 34:2	0.091786	0.010934	0.086897	0.015513	0.14109	0.004182
MGDG 34:1	0.053063	0.004867	0.048006	0.009031	0.060166	0.006191
MGDG 36:6	7.143605	0.436212	7.122012	0.241507	7.916638	0.315789
MGDG 36:5	0.4972	0.06854	0.501936	0.011043	0.543084	0.021682
MGDG 36:4	0.138064	0.011676	0.127932	0.002417	0.16661	0.009928
MGDG 36:3	0.017144	0.002442	0.016503	0.002068	0.020254	0.000953
MGDG 36:2	0.000996	0.000445	0.000929	0.000551	0.001312	0.00026
MGDG 36:1	5.79E-05	9.25E-05	9.09E-05	0.000165	0.00016	0.000173
MGDG 38:6	0.006002	0.001542	0.005949	0.001181	0.007587	0.001665
MGDG 38:5	0.002169	0.000412	0.002019	0.000338	0.002345	0.000349
MGDG 38:4	0.002059	0.000313	0.002281	8.43E-05	0.002651	0.000198
MGDG 38:3	9.29E-05	6.21E-05	0.000265	0.000135	0.000201	0.000148
Total MGDG	75.0948	2.444528	74.81695	0.291296	75.72311	1.05407

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.138347	0.008447	0.114214	0.014676	0.137896	0.01058
PG 32:0	0.090133	0.018056	0.097134	0.009539	0.121228	0.014106
PG 34:4	2.176931	0.120619	2.036419	0.210076	2.30105	0.114831
PG 34:3	0.8164	0.08391	1.003519	0.13799	1.100412	0.066464
PG 34:2	0.403145	0.019459	0.390341	0.030787	0.482934	0.022038
PG 34:1	0.250688	0.020114	0.236663	0.021431	0.300551	0.033965
PG 34:0	0.003138	0.002122	0.005346	0.001458	0.001478	0.002831
Total PG	3.878783	0.228073	3.883637	0.390366	4.445549	0.193361
Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.167342	0.011074	0.158422	0.012552	0.143461	0.005008
PG 32:0	0.129827	0.014315	0.132635	0.021141	0.129319	0.006479
PG 34:4	2.405745	0.104052	2.349538	0.083753	2.059016	0.032585
PG 34:3	0.934243	0.074231	1.089298	0.127328	0.987192	0.0676
PG 34:2	0.446521	0.029401	0.457621	0.036513	0.407013	0.017144
PG 34:1	0.244525	0.021893	0.266386	0.030177	0.213954	0.022835
PG 34:0	0.007383	0.002331	0.008199	0.003752	0.005717	0.001467
Total PG	4.335585	0.173737	4.462099	0.252333	3.945672	0.088366
Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.146081	0.009617	0.14103	0.024916	0.131132	0.009809
PG 32:0	0.112924	0.014308	0.130889	0.015765	0.130406	0.017915
PG 34:4	2.013327	0.170481	1.939123	0.204201	1.916969	0.113982
PG 34:3	0.872218	0.059384	0.957634	0.086746	0.971137	0.064367
PG 34:2	0.390117	0.028559	0.396557	0.032251	0.430659	0.027223
PG 34:1	0.189092	0.013701	0.209832	0.029085	0.255759	0.022515
PG 34:0	0.008165	0.002159	0.007632	0.004213	0.002773	0.004465
Total PG	3.731924	0.232168	3.782696	0.271885	3.838835	0.232048
Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.128447	0.01006	0.124147	0.009611	0.129206	0.011095
PG 32:0	0.099457	0.008755	0.099736	0.009834	0.103245	0.006083
PG 34:4	1.858205	0.141256	1.8473	0.10907	1.814413	0.150647
PG 34:3	0.754891	0.065388	0.809268	0.052383	0.794464	0.0402
PG 34:2	0.403936	0.03604	0.38622	0.010695	0.425283	0.051292
PG 34:1	0.266823	0.031465	0.264855	0.016627	0.283907	0.018116
PG 34:0	0.004388	0.005419	0.00303	0.004241	0.001534	0.001799
Total PG	3.516147	0.247192	3.534557	0.127326	3.552051	0.245644

	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
PG 32:1	0.128845	0.023141	0.123332	0.013185	0.131146	0.016296
PG 32:0	0.13258	0.014413	0.139762	0.009299	0.134416	0.006993
PG 34:4	1.838216	0.23789	1.756094	0.098717	1.92079	0.19416
PG 34:3	0.969207	0.110015	1.040647	0.065969	1.045651	0.066536
PG 34:2	0.406578	0.054342	0.376588	0.039905	0.460423	0.044565
PG 34:1	0.220974	0.024378	0.207853	0.020878	0.26368	0.017766
PG 34:0	0.004827	0.003567	0.005831	0.00279	0.003808	0.00153
Total PG	3.701227	0.443108	3.650107	0.165617	3.959913	0.318701
	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000195	0.000146	6.3E-05	4.19E-05	0.000348	0.000191
lysoPG 16:0	0.001087	0.000254	0.000937	0.000145	0.001218	8.21E-05
lysoPG 18:3	0.00825	0.000544	0.00853	0.000493	0.00892	0.000386
lysoPG 18:2	0.000497	0.000117	0.000378	0.000146	0.00056	0.000205
lysoPG 18:1	0.000346	0.000109	0.000418	0.000111	0.000547	0.00018
Total lysoPG	0.010375	0.000584	0.010326	0.000631	0.011592	0.000516
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000506	0.000119	0.000431	0.000284	0.000344	0.000322
lysoPG 16:0	0.001425	0.000213	0.001357	0.000145	0.001442	0.000279
lysoPG 18:3	0.009436	0.00047	0.010331	0.0009	0.008718	0.000361
lysoPG 18:2	0.00067	6.12E-05	0.000733	0.000162	0.000553	0.000191
lysoPG 18:1	0.000577	0.000149	0.000603	0.000291	0.000456	0.000262
Total lysoPG	0.012614	0.000677	0.013456	0.00063	0.011513	0.001105
	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000693	0.000178	0.000642	0.000258	0.000171	0.00014
lysoPG 16:0	0.001293	0.000203	0.001412	0.000176	0.001067	0.000141
lysoPG 18:3	0.009556	0.000668	0.00852	0.000658	0.007751	0.000856
lysoPG 18:2	0.000542	0.000263	0.000685	0.000234	0.000513	0.00013
lysoPG 18:1	0.000375	0.000178	0.000516	0.000217	0.000642	0.000152
Total lysoPG	0.01246	0.000561	0.011775	0.001208	0.010144	0.000899

	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000387	0.000128	0.000682	0.000173	0.000696	0.000215
lysoPG 16:0	0.001185	0.000178	0.00123	0.000132	0.001396	0.000226
lysoPG 18:3	0.007814	0.001027	0.008329	0.000795	0.007761	0.000517
lysoPG 18:2	0.000613	0.000123	0.000711	0.000175	0.000802	0.000118
lysoPG 18:1	0.000706	0.000202	0.000626	9.17E-05	0.000665	8.04E-05
Total lysoPG	0.010706	0.00149	0.011577	0.001015	0.011319	0.000358
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
lysoPG 16:1	0.000897	0.000263	0.000699	0.000249	0.001206	0.000469
lysoPG 16:0	0.001461	0.000334	0.001544	0.000144	0.00172	0.000189
lysoPG 18:3	0.009086	0.002119	0.00893	0.000275	0.009577	0.001138
lysoPG 18:2	0.000687	0.000146	0.000627	0.000223	0.001176	0.000167
lysoPG 18:1	0.000488	9.46E-05	0.000453	9.67E-05	0.000627	0.000154
Total lysoPG	0.012619	0.00257	0.012253	0.000266	0.014306	0.001727
	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	8.56E-05	3.68E-05	6.84E-05	4.05E-05	0.000125	5.47E-05
LysoPC 16:0	0.001997	0.000157	0.001949	0.000281	0.002985	0.000381
LysoPC 18:3	0.003353	0.000362	0.003514	0.000395	0.004852	0.000285
LysoPC 18:2	0.003824	0.000331	0.004283	0.000322	0.005673	0.000681
LysoPC 18:1	0.000952	9.49E-05	0.000971	6.83E-05	0.00137	0.000182
LysoPC 18:0	0.00018	2.58E-05	0.000196	6.3E-05	0.000377	6.52E-05
Total LysoPC	0.010392	0.00092	0.010981	0.001032	0.015382	0.001504
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.000115	5.94E-05	0.000104	4.55E-05	9.45E-05	5.1E-05
LysoPC 16:0	0.002438	0.000283	0.002723	0.000251	0.002836	0.000365
LysoPC 18:3	0.004296	0.000378	0.005232	0.000545	0.004758	0.000206
LysoPC 18:2	0.004373	0.000237	0.005637	0.000314	0.004928	0.00041
LysoPC 18:1	0.001045	0.000149	0.00117	0.000168	0.000987	0.000134
LysoPC 18:0	0.000225	3.86E-05	0.000296	5.54E-05	0.000251	8.51E-05
Total LysoPC	0.012492	0.000893	0.015162	0.000921	0.013855	0.000789

	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.000115	6.5E-05	0.000152	3.91E-05	0.000134	7.15E-05
LysoPC 16:0	0.00229	0.000182	0.002573	0.000213	0.002444	0.000253
LysoPC 18:3	0.004511	0.000313	0.004948	0.000257	0.004638	0.000646
LysoPC 18:2	0.004257	0.000201	0.004546	0.000478	0.004814	0.000516
LysoPC 18:1	0.001307	0.000185	0.001125	0.000229	0.001021	0.000117
LysoPC 18:0	0.000192	5.14E-05	0.000172	3.79E-05	0.000188	3.75E-05
Total LysoPC	0.012673	0.000658	0.013515	0.000714	0.013238	0.001329
	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	6.32E-05	2.34E-05	0.000114	2.85E-05	0.000122	2.41E-05
LysoPC 16:0	0.002113	0.000263	0.002339	0.000425	0.002334	0.000125
LysoPC 18:3	0.003516	0.000474	0.004332	0.00036	0.004176	0.000229
LysoPC 18:2	0.004039	0.00051	0.004451	0.000378	0.004862	0.000378
LysoPC 18:1	0.00094	0.000118	0.001227	0.000251	0.001182	0.000145
LysoPC 18:0	0.000212	5.99E-05	0.000267	5.72E-05	0.000292	7.37E-05
Total LysoPC	0.010883	0.001156	0.012729	0.001351	0.012967	0.00072
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPC 16:1	0.000143	9.35E-05	0.000178	2.42E-05	0.000152	3.12E-05
LysoPC 16:0	0.002659	0.000382	0.002927	0.000355	0.00293	0.000144
LysoPC 18:3	0.006029	0.000906	0.0065	0.000536	0.006339	0.000729
LysoPC 18:2	0.005008	0.000669	0.005587	0.000403	0.006229	0.000606
LysoPC 18:1	0.001421	0.000208	0.00166	0.000123	0.001562	0.00023
LysoPC 18:0	0.000184	7.48E-05	0.000203	7.1E-05	0.000241	3.81E-05
Total LysoPC	0.015444	0.002052	0.017055	0.00133	0.017454	0.001622
	Col-o 0hr		101919 0hr		082589 0hr	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.000187	0.00011	0.000124	0.000109	0.000212	4.99E-05
LysoPE 16:0	0.005467	0.000669	0.005428	0.000696	0.007021	0.000148
LysoPE 18:3	0.003892	0.000497	0.004398	0.000676	0.005662	0.000232
LysoPE 18:2	0.004992	0.000543	0.005263	0.000467	0.007102	0.000444
LysoPE 18:1	0.000764	0.000127	0.000626	7.39E-05	0.000982	9.39E-05
Total LysoPE	0.015301	0.001703	0.015839	0.00173	0.020979	0.000694

	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.000114	9.85E-05	5.73E-05	7.99E-05	0.000132	0.000141
LysoPE 16:0	0.007264	0.000594	0.007706	0.0012	0.007571	0.000873
LysoPE 18:3	0.004997	0.000762	0.005797	0.000618	0.005436	0.000505
LysoPE 18:2	0.006207	0.000474	0.006541	0.000593	0.006456	0.000424
LysoPE 18:1	0.000765	0.000168	0.000947	0.000146	0.000576	0.000173
Total LysoPE	0.019347	0.001237	0.021048	0.002482	0.020171	0.001217
	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	6.33E-05	7.65E-05	0.000213	0.00019	7.77E-05	4.86E-05
LysoPE 16:0	0.006804	0.000731	0.007225	0.001441	0.006761	0.000391
LysoPE 18:3	0.004955	0.000607	0.005197	0.000393	0.004909	0.000398
LysoPE 18:2	0.006372	0.000349	0.006476	0.000423	0.006317	0.000282
LysoPE 18:1	0.000872	9E-05	0.000919	0.00023	0.000792	0.000163
Total LysoPE	0.019067	0.001489	0.020029	0.001902	0.018856	0.000686
	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.000103	6.62E-05	0.000167	2.92E-05	7.87E-05	6.47E-05
LysoPE 16:0	0.005319	0.00096	0.006157	0.000397	0.005951	0.000234
LysoPE 18:3	0.004225	0.000593	0.004835	0.000426	0.004617	0.000461
LysoPE 18:2	0.005136	0.00067	0.005933	0.00063	0.006291	0.000964
LysoPE 18:1	0.000554	0.000129	0.000841	0.000191	0.000752	0.000219
Total LysoPE	0.015336	0.002039	0.017934	0.001245	0.017689	0.001524
	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
Sample description	ave	stdev	ave	stdev	ave	stdev
LysoPE 16:1	0.000106	0.000121	0.000107	8.6E-05	0.000138	0.000111
LysoPE 16:0	0.00784	0.001435	0.008123	0.00093	0.008002	0.000291
LysoPE 18:3	0.006095	0.000753	0.006458	0.00078	0.006398	0.000344
LysoPE 18:2	0.007357	0.001058	0.007852	0.000525	0.00836	0.000392
LysoPE 18:1	0.00109	0.000408	0.00098	0.00022	0.001062	8.4E-05
Total LysoPE	0.022488	0.003412	0.02352	0.001514	0.02396	0.000775

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.00256	0.000292	0.002338	0.000118	0.002753	0.000245
PC 34:4	0.035384	0.002527	0.036568	0.004326	0.046503	0.001146
PC 34:3	0.741931	0.057303	0.802694	0.080127	0.890247	0.031805
PC 34:2	0.575837	0.038121	0.630995	0.02838	0.693648	0.016328
PC 34:1	0.131965	0.008439	0.128092	0.007365	0.14989	0.009566
PC 36:6	0.491338	0.038145	0.529241	0.059215	0.57496	0.035733
PC 36:5	0.816264	0.043308	0.915425	0.066092	0.962585	0.037554
PC 36:4	0.683174	0.039271	0.749843	0.042703	0.820874	0.024065
PC 36:3	0.309549	0.024781	0.326487	0.01895	0.384244	0.023468
PC 36:2	0.083808	0.008934	0.089272	0.007671	0.100303	0.005473
PC 36:1	0	0	0	0	0	0
PC 38:6	0.002442	0.000139	0.002476	0.000448	0.00282	0.000213
PC 38:5	0.007414	0.000734	0.007984	0.000893	0.009681	0.000668
PC 38:4	0.013335	0.002931	0.014137	0.002184	0.014686	0.001012
PC 38:3	0.012808	0.002208	0.013702	0.001124	0.015859	0.002014
PC 38:2	0.006488	0.001447	0.006511	0.000305	0.008058	0.001
PC 40:5	0.000562	8.25E-05	0.00062	0.000209	0.000646	0.000143
PC 40:4	0.000846	0.000109	0.000853	0.000202	0.000982	0.00016
PC 40:3	0.000725	7.12E-05	0.000912	0.000219	0.000839	7.05E-05
PC 40:2	0.000535	7.5E-05	0.000695	8.44E-05	0.000776	0.000171
Total PC	3.916966	0.255449	4.258844	0.280343	4.680353	0.157843
	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
Sample description	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.003587	0.000275	0.003474	0.000122	0.003266	0.000313
PC 34:4	0.046699	0.003111	0.045678	0.003429	0.04585	0.00166
PC 34:3	0.953182	0.039128	0.993041	0.04462	0.940662	0.044493
PC 34:2	0.655458	0.02684	0.685408	0.037487	0.644608	0.021482
PC 34:1	0.139509	0.006854	0.141862	0.014481	0.115717	0.010278
PC 36:6	0.639522	0.039614	0.663327	0.043771	0.59569	0.035156
PC 36:5	0.991396	0.048527	1.052151	0.053061	0.94584	0.032557
PC 36:4	0.766162	0.04577	0.803806	0.048567	0.715837	0.027029
PC 36:3	0.343371	0.02574	0.349639	0.019016	0.308407	0.013495
PC 36:2	0.086702	0.004378	0.094677	0.005213	0.087548	0.01048
PC 36:1	0	0	0	0	0	0
PC 38:6	0.002826	0.000356	0.003477	0.000323	0.003375	0.00039
PC 38:5	0.008024	0.000497	0.008686	0.000675	0.009185	0.000789
PC 38:4	0.013411	0.000623	0.014488	0.001111	0.012957	0.000722
PC 38:3	0.013301	0.001142	0.014627	0.001327	0.012436	0.001232
PC 38:2	0.006536	0.000874	0.007016	0.000957	0.00532	0.000426
PC 40:5	0.000815	0.000119	0.000817	0.000153	0.000776	0.000114
PC 40:4	0.001048	8.92E-05	0.001126	0.000159	0.001114	0.000147
PC 40:3	0.000921	0.000143	0.000919	7.72E-05	0.000808	8.07E-05
PC 40:2	0.000623	9.48E-05	0.000647	9.18E-05	0.000708	7.48E-05

Total PC	4.673092	0.212687	4.884868	0.171415	4.450104	0.112992
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Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002856	0.000233	0.002798	0.00019	0.002976	0.000172
PC 34:4	0.044708	0.002898	0.043142	0.003298	0.046774	0.003214
PC 34:3	0.896426	0.070457	0.917856	0.07027	0.88308	0.059052
PC 34:2	0.54303	0.03013	0.545539	0.044174	0.61259	0.043148
PC 34:1	0.160823	0.011561	0.136879	0.019556	0.11945	0.007591
PC 36:6	0.691399	0.049222	0.673443	0.063118	0.588315	0.047455
PC 36:5	0.990755	0.047982	0.944294	0.063395	0.934037	0.096625
PC 36:4	0.780682	0.024555	0.701063	0.08544	0.709985	0.05952
PC 36:3	0.377151	0.017126	0.326809	0.055538	0.31792	0.025333
PC 36:2	0.075718	0.00461	0.065507	0.012735	0.072144	0.005809
PC 36:1	0	0	2.19E-05	4.89E-05	0	0
PC 38:6	0.004106	0.000509	0.004214	0.000831	0.0034	0.000225
PC 38:5	0.008811	0.000855	0.008723	0.000968	0.007668	0.000173
PC 38:4	0.01351	0.000692	0.01317	0.001098	0.011818	0.000747
PC 38:3	0.012273	0.000925	0.012115	0.00101	0.012396	0.00127
PC 38:2	0.00604	0.000311	0.005456	0.000231	0.005518	0.000718
PC 40:5	0.000793	0.000114	0.000866	0.000324	0.000833	0.000111
PC 40:4	0.001138	0.00014	0.000913	0.000142	0.001026	0.000164
PC 40:3	0.001002	9.82E-05	0.000779	0.000125	0.000772	0.000161
PC 40:2	0.000697	6.98E-05	0.000623	2.46E-05	0.00062	6.67E-05
Total PC	4.611918	0.209589	4.404209	0.230541	4.331323	0.334218
Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002266	0.000248	0.002265	0.000224	0.002157	0.000142
PC 34:4	0.035399	0.004056	0.035525	0.003471	0.035153	0.003986
PC 34:3	0.763384	0.079316	0.773903	0.060275	0.752037	0.078815
PC 34:2	0.577519	0.033199	0.575717	0.031086	0.580556	0.054395
PC 34:1	0.127635	0.01121	0.131012	0.013907	0.13018	0.010822
PC 36:6	0.442776	0.045052	0.463142	0.049687	0.426262	0.045638
PC 36:5	0.795574	0.059924	0.804458	0.040022	0.776262	0.073886
PC 36:4	0.672081	0.038547	0.672401	0.034396	0.680214	0.07695
PC 36:3	0.320367	0.010305	0.324132	0.026061	0.335035	0.043314
PC 36:2	0.092557	0.008119	0.084407	0.004769	0.097972	0.011382
PC 36:1	0	0	0	0	0	0
PC 38:6	0.002778	0.000626	0.002874	0.000488	0.00231	0.000479
PC 38:5	0.007354	0.000219	0.007693	0.001061	0.007414	0.000976
PC 38:4	0.012639	0.001605	0.012287	0.001405	0.012285	0.001777
PC 38:3	0.012759	0.001756	0.012585	0.001826	0.013078	0.001988
PC 38:2	0.006573	0.001084	0.006258	0.000915	0.006276	0.000886
PC 40:5	0.000635	8.35E-05	0.000605	0.00013	0.000679	0.000109
PC 40:4	0.000824	0.000134	0.000845	0.000173	0.000854	0.000118
PC 40:3	0.000848	0.000152	0.00075	0.00011	0.000715	8.99E-05

PC 40:2	0.000663	0.000122	0.000587	7.93E-05	0.000597	0.000154
Total PC	3.87463	0.255767	3.911444	0.237326	3.860035	0.389736

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PC 32:0	0.002455	0.000424	0.002404	0.000165	0.002737	0.00027
PC 34:4	0.044207	0.005697	0.043393	0.001672	0.043976	0.003047
PC 34:3	0.892792	0.135704	0.915407	0.037484	0.87206	0.036745
PC 34:2	0.528137	0.076489	0.547805	0.018752	0.614101	0.044318
PC 34:1	0.147627	0.03195	0.156924	0.01709	0.149497	0.009941
PC 36:6	0.639173	0.121674	0.666544	0.038746	0.561618	0.020512
PC 36:5	0.881056	0.157391	0.912916	0.021579	0.872034	0.049207
PC 36:4	0.681944	0.123626	0.715473	0.029565	0.737028	0.050055
PC 36:3	0.348803	0.071499	0.358352	0.027035	0.367691	0.027357
PC 36:2	0.078554	0.017157	0.079736	0.007903	0.088264	0.008574
PC 36:1	3.86E-06	8.62E-06	0	0	0	0
PC 38:6	0.004065	0.000711	0.004046	0.000435	0.003254	0.000368
PC 38:5	0.008167	0.001142	0.008475	0.00035	0.008386	0.000357
PC 38:4	0.013105	0.001959	0.014017	0.000984	0.013642	0.001161
PC 38:3	0.014494	0.00236	0.014703	0.001384	0.015355	0.001684
PC 38:2	0.00691	0.001497	0.007302	0.000734	0.008254	0.000671
PC 40:5	0.00089	0.000212	0.000833	0.000144	0.000868	0.000131
PC 40:4	0.000887	0.00014	0.000914	0.000114	0.000992	0.000179
PC 40:3	0.001038	0.000185	0.001043	7.95E-05	0.001019	0.000117
PC 40:2	0.000768	0.000199	0.000733	9.34E-05	0.000772	8.44E-05
Total PC	4.295078	0.712235	4.451022	0.075066	4.361548	0.233646

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.011334	0.001671	0.012489	0.001759	0.014146	0.001117
PE 34:3	0.629952	0.049247	0.694235	0.102285	0.758007	0.056089
PE 34:2	0.824277	0.043711	0.902819	0.102601	0.988063	0.035778
PE 34:1	0.043288	0.001523	0.043451	0.005536	0.045692	0.004104
PE 36:6	0.218038	0.02157	0.247966	0.04176	0.26473	0.031154
PE 36:5	0.594333	0.051294	0.652134	0.085568	0.706594	0.04545
PE 36:4	0.493535	0.032121	0.513867	0.048205	0.588541	0.026191
PE 36:3	0.148921	0.013849	0.156472	0.012025	0.19058	0.010881
PE 36:2	0.050502	0.003444	0.05649	0.006216	0.061224	0.003935
PE 36:1	0.001774	0.000561	0.001358	0.00062	0.00168	0.000848
PE 38:6	0.004014	0.000332	0.004526	0.00047	0.004337	0.000463
PE 38:5	0.005532	0.000569	0.005988	0.001107	0.006445	0.000493
PE 38:4	0.005839	0.000505	0.006876	0.000746	0.007542	0.000398
PE 38:3	0.012424	0.001515	0.014444	0.001299	0.017338	0.001922
PE 38:2	0.017454	0.002508	0.019916	0.001295	0.024124	0.001868
PE 40:3	0.007316	0.001017	0.008767	0.000989	0.010461	0.000671
PE 40:2	0.01503	0.001194	0.01808	0.002124	0.020791	0.000986

PE 42:4	0.006205	0.000797	0.007065	0.000672	0.009492	0.000628
PE 42:3	0.022381	0.002259	0.027019	0.002711	0.036008	0.000797
PE 42:2	0.016387	0.001648	0.019134	0.001894	0.026543	0.000837
Total PE	3.128538	0.218793	3.413098	0.405544	3.782338	0.196431

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.014685	0.000815	0.015307	0.001907	0.01498	0.000643
PE 34:3	0.843614	0.06557	0.879894	0.11799	0.86024	0.041422
PE 34:2	0.997216	0.069517	1.05139	0.08368	1.052195	0.056261
PE 34:1	0.053202	0.004659	0.050311	0.004465	0.043948	0.00182
PE 36:6	0.274287	0.018652	0.292198	0.046583	0.266292	0.013876
PE 36:5	0.732374	0.041836	0.768403	0.079838	0.726379	0.038455
PE 36:4	0.58074	0.033666	0.595247	0.053759	0.573911	0.018077
PE 36:3	0.175861	0.014876	0.177516	0.021257	0.162206	0.008987
PE 36:2	0.060179	0.003841	0.066535	0.004399	0.067384	0.007201
PE 36:1	0.00136	0.000893	0.001046	0.000738	0.00082	0.000704
PE 38:6	0.004145	0.000533	0.004643	0.000789	0.003977	0.000347
PE 38:5	0.006616	0.000574	0.007028	0.000554	0.006692	0.000332
PE 38:4	0.006608	0.000371	0.007298	0.000428	0.007482	0.000475
PE 38:3	0.013652	0.000806	0.015679	0.001277	0.013666	0.000744
PE 38:2	0.019442	0.001604	0.020883	0.001942	0.01909	0.001649
PE 40:3	0.00967	0.000954	0.01046	0.001137	0.010401	0.000506
PE 40:2	0.018574	0.001103	0.019956	0.001617	0.021374	0.000916
PE 42:4	0.007748	0.001198	0.008022	0.00053	0.009055	0.000639
PE 42:3	0.027407	0.002255	0.029596	0.002663	0.032353	0.000639
PE 42:2	0.020993	0.002118	0.022789	0.002749	0.026277	0.000723
Total PE	3.868371	0.252496	4.044201	0.40004	3.918723	0.164303

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.015164	0.000646	0.014835	0.00066	0.013834	0.000508
PE 34:3	0.771394	0.064867	0.770601	0.036097	0.739804	0.022045
PE 34:2	0.915441	0.053757	0.904146	0.094885	0.931718	0.02508
PE 34:1	0.055879	0.003915	0.044395	0.005431	0.040903	0.003633
PE 36:6	0.281212	0.03274	0.268903	0.01908	0.240235	0.014582
PE 36:5	0.761375	0.053577	0.724217	0.036143	0.670871	0.030612
PE 36:4	0.621211	0.027105	0.594791	0.053551	0.55831	0.020859
PE 36:3	0.180749	0.008147	0.166073	0.019675	0.164175	0.004441
PE 36:2	0.049139	0.003386	0.047501	0.007375	0.053851	0.002156
PE 36:1	0.001505	0.000507	0.001387	0.000502	0.001634	0.001076
PE 38:6	0.004991	0.000521	0.005045	0.000482	0.004433	0.000494
PE 38:5	0.006358	0.000838	0.006489	0.000376	0.006206	0.000499
PE 38:4	0.007086	0.000654	0.006598	0.001234	0.006321	0.000533
PE 38:3	0.013394	0.000887	0.011442	0.00048	0.012935	0.001344
PE 38:2	0.017965	0.001172	0.015121	0.001485	0.017737	0.001796
PE 40:3	0.008048	0.000646	0.008545	0.000686	0.009646	0.000805
PE 40:2	0.016413	0.001362	0.016894	0.001922	0.020275	0.001492
PE 42:4	0.00725	0.001159	0.007562	0.001163	0.008412	0.000567
PE 42:3	0.025903	0.00218	0.027821	0.004085	0.031892	0.002403

PE 42:2	0.021774	0.002	0.02306	0.002944	0.025712	0.001527
Total PE	3.782251	0.247262	3.665426	0.212503	3.558905	0.115736

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.011175	0.001287	0.011574	0.001498	0.012059	0.001765
PE 34:3	0.605059	0.057015	0.654754	0.073633	0.636861	0.086355
PE 34:2	0.767075	0.067491	0.791301	0.066701	0.813875	0.107393
PE 34:1	0.037141	0.003165	0.03853	0.0072	0.035109	0.004183
PE 36:6	0.200521	0.020938	0.22424	0.030758	0.202007	0.029229
PE 36:5	0.569994	0.057358	0.603554	0.058822	0.583239	0.077549
PE 36:4	0.479034	0.054116	0.499772	0.041795	0.502475	0.072942
PE 36:3	0.146943	0.0138	0.151774	0.018099	0.154608	0.025777
PE 36:2	0.0516	0.003127	0.050526	0.003092	0.056176	0.009245
PE 36:1	0.001262	0.000838	0.001149	0.000587	0.00096	0.000551
PE 38:6	0.003551	0.000545	0.003311	0.000344	0.003375	0.000622
PE 38:5	0.005647	0.000767	0.005679	0.000395	0.005618	0.000717
PE 38:4	0.005559	0.000533	0.00611	0.000588	0.006342	0.000908
PE 38:3	0.012486	0.002127	0.012239	0.001284	0.013023	0.002015
PE 38:2	0.017269	0.003955	0.016197	0.001234	0.017839	0.002164
PE 40:3	0.008337	0.001057	0.008271	0.000821	0.009143	0.001222
PE 40:2	0.01693	0.001799	0.015736	0.001661	0.018806	0.002404
PE 42:4	0.007364	0.001075	0.00785	0.000939	0.008514	0.001347
PE 42:3	0.02507	0.002738	0.025357	0.003281	0.028218	0.00376
PE 42:2	0.019292	0.002588	0.018547	0.002348	0.022461	0.003321
Total PE	2.991309	0.286174	3.14647	0.297794	3.13071	0.427543
Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PE 34:4	0.014017	0.00229	0.014183	0.000925	0.014403	0.000699
PE 34:3	0.718836	0.09282	0.751363	0.05229	0.712216	0.027101
PE 34:2	0.805503	0.111344	0.839592	0.047287	0.88615	0.053122
PE 34:1	0.043098	0.009989	0.04327	0.003811	0.04249	0.003743
PE 36:6	0.275384	0.042369	0.295525	0.021622	0.245784	0.00983
PE 36:5	0.709696	0.10359	0.743475	0.044093	0.676103	0.039335
PE 36:4	0.571622	0.084773	0.584982	0.031201	0.568693	0.037819
PE 36:3	0.157906	0.024827	0.161281	0.008271	0.165236	0.008285
PE 36:2	0.04644	0.009826	0.047988	0.003807	0.055478	0.004947
PE 36:1	0.001674	0.00064	0.002059	0.000375	0.000884	0.000469
PE 38:6	0.004088	0.000643	0.004233	0.000385	0.004081	0.000389
PE 38:5	0.006276	0.000397	0.006798	0.000492	0.006238	0.000395
PE 38:4	0.006153	0.001054	0.006602	0.000262	0.006618	0.000397
PE 38:3	0.013578	0.00225	0.013357	0.000869	0.014181	0.00125
PE 38:2	0.018262	0.003066	0.018117	0.001426	0.020749	0.001902
PE 40:3	0.008606	0.001911	0.00868	0.000468	0.01011	0.000684
PE 40:2	0.017073	0.003143	0.016864	0.001243	0.020924	0.001493
PE 42:4	0.007866	0.001582	0.007688	0.000788	0.008455	0.000848

PE 42:3	0.028783	0.005806	0.028282	0.00184	0.031664	0.002244
PE 42:2	0.02469	0.00468	0.025265	0.001303	0.02596	0.001591
Total PE	3.479552	0.480308	3.619601	0.197494	3.516414	0.17772

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.003408	0.00052	0.003528	0.000618	0.006255	0.001081
PI 34:3	0.448288	0.039277	0.454993	0.092175	0.639944	0.070158
PI 34:2	0.402679	0.034734	0.410227	0.065289	0.578185	0.080556
PI 34:1	0.015109	0.002623	0.013436	0.003955	0.024773	0.003666
PI 36:6	0.017709	0.001988	0.018831	0.00402	0.024869	0.001729
PI 36:5	0.032795	0.002335	0.034099	0.006219	0.046704	0.005325
PI 36:4	0.025034	0.003727	0.027637	0.004237	0.038203	0.003322
PI 36:3	0.035512	0.006256	0.042807	0.009977	0.058689	0.009858
PI 36:2	0.020863	0.003508	0.024043	0.003438	0.032756	0.00441
PI 36:1	0	0	0	0	0	0
Total PI	1.001397	0.08411	1.0296	0.177365	1.450379	0.165275

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.004187	0.000688	0.005305	0.001163	0.005012	0.000852
PI 34:3	0.577332	0.034201	0.637645	0.051729	0.572522	0.061789
PI 34:2	0.480431	0.024748	0.528574	0.056193	0.477056	0.034878
PI 34:1	0.019263	0.002068	0.022222	0.002181	0.016434	0.002595
PI 36:6	0.022396	0.002528	0.027938	0.004291	0.025052	0.004046
PI 36:5	0.037765	0.005239	0.038819	0.003543	0.036933	0.002414
PI 36:4	0.026595	0.002549	0.031992	0.001763	0.026297	0.001012
PI 36:3	0.044892	0.007594	0.050641	0.007702	0.049561	0.001906
PI 36:2	0.025415	0.002025	0.029873	0.001708	0.028782	0.00248
PI 36:1	0	0	0	0	0	0
Total PI	1.238276	0.061307	1.373008	0.109546	1.237648	0.097362

Sample descriptio n	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.005687	0.001017	0.006916	0.001722	0.005641	0.000827
PI 34:3	0.605946	0.081363	0.626326	0.108271	0.559621	0.030665
PI 34:2	0.461759	0.050768	0.484705	0.042863	0.479989	0.033305
PI 34:1	0.032362	0.005162	0.023288	0.006379	0.01842	0.004078
PI 36:6	0.029196	0.003893	0.031834	0.008695	0.024246	0.000393
PI 36:5	0.04287	0.004036	0.045098	0.006981	0.039654	0.00489
PI 36:4	0.031208	0.002094	0.030229	0.003171	0.028253	0.002992
PI 36:3	0.031666	0.003692	0.035015	0.010962	0.041714	0.006508

PI 36:2	0.01935	0.003052	0.019318	0.005315	0.025322	0.003869
PI 36:1	4.37E-05	9.77E-05	0	0	0	0
Total PI	1.260088	0.14135	1.302729	0.136869	1.222862	0.071259

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.003941	0.001125	0.003977	0.000762	0.004963	0.000773
PI 34:3	0.459505	0.033493	0.531359	0.050152	0.526246	0.055379
PI 34:2	0.396332	0.029494	0.45171	0.040133	0.45806	0.034351
PI 34:1	0.015194	0.003024	0.017517	0.004631	0.018745	0.002683
PI 36:6	0.018023	0.001428	0.017246	0.000824	0.018525	0.002369
PI 36:5	0.028708	0.002842	0.029916	0.004727	0.036267	0.002499
PI 36:4	0.025372	0.002659	0.026501	0.002699	0.029749	0.003833
PI 36:3	0.039776	0.005435	0.045357	0.00212	0.050977	0.009328
PI 36:2	0.02486	0.001851	0.023816	0.001716	0.02947	0.003434
PI 36:1	0	0	0.000275	0.000614	0	0
Total PI	1.011709	0.068703	1.147673	0.087764	1.173002	0.092968

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PI 34:4	0.006307	0.001327	0.005884	0.001219	0.006323	0.000963
PI 34:3	0.657453	0.101196	0.724188	0.064817	0.615254	0.035824
PI 34:2	0.488669	0.092409	0.530619	0.031199	0.510828	0.036729
PI 34:1	0.027841	0.012098	0.027838	0.002343	0.021614	0.00134
PI 36:6	0.024954	0.00344	0.025517	0.002329	0.023508	0.002692
PI 36:5	0.034671	0.00655	0.036557	0.004121	0.035016	0.002254
PI 36:4	0.025883	0.005237	0.027541	0.001572	0.030238	0.002421
PI 36:3	0.03484	0.005479	0.038027	0.006207	0.041015	0.004491
PI 36:2	0.018871	0.005089	0.020917	0.002315	0.026712	0.003158
PI 36:1	6.92E-05	0.000155	0	0	0	0
Total PI	1.319558	0.218488	1.437088	0.10399	1.310508	0.071879

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.000216	4.06E-05	0.000232	0.000124	0.000366	7.32E-06
PS 34:3	0.025391	0.004687	0.024851	0.005842	0.0357	0.002779
PS 34:2	0.021685	0.004198	0.021917	0.005216	0.030588	0.00401
PS 34:1	0.000794	0.000238	0.000846	0.000213	0.001076	0.000244
PS 36:6	0.001087	0.00022	0.00112	0.000219	0.001363	7.39E-05
PS 36:5	0.002991	0.000587	0.002879	0.000476	0.003831	0.000602
PS 36:4	0.004191	0.000868	0.004144	0.000613	0.005802	0.000766
PS 36:3	0.010896	0.001937	0.011866	0.002278	0.016191	0.001903
PS 36:2	0.00737	0.00122	0.007924	0.001324	0.010863	0.001708
PS 36:1	0.000241	0.000132	0.000153	0.000134	0.000277	0.000118
PS 38:6	0.000148	2.28E-05	0.000105	8.23E-05	0.000162	4.05E-05
PS 38:5	0.00037	0.000147	0.00032	6.27E-05	0.000544	8.11E-05
PS 38:4	0.001082	0.000323	0.001012	0.000302	0.001762	0.000185
PS 38:3	0.009303	0.002086	0.010687	0.002123	0.015008	0.001706
PS 38:2	0.007169	0.00172	0.007331	0.001275	0.011065	0.001514
PS 38:1	0.000236	0.000151	0.000266	0.000156	0.000535	0.000111
PS 40:4	0.000634	0.000155	0.000772	0.000251	0.001149	0.000182
PS 40:3	0.018598	0.004037	0.020705	0.004663	0.027742	0.00262
PS 40:2	0.013327	0.002869	0.013153	0.003256	0.018711	0.002615
PS 40:1	0.000331	8.33E-05	0.000318	0.000219	0.000498	0.000161
PS 42:4	0.014768	0.00402	0.015848	0.004111	0.023604	0.003274
PS 42:3	0.046104	0.009782	0.047409	0.013496	0.068772	0.009776
PS 42:2	0.021648	0.005058	0.021455	0.006801	0.032372	0.004545
PS 42:1	0.000751	0.00034	0.00106	0.000502	0.001207	0.00044
PS 44:3	0.00173	0.000264	0.00202	0.000553	0.002117	0.000487
PS 44:2	0.001572	0.000401	0.001473	0.000646	0.001818	0.000187
Total PS	0.212631	0.043567	0.219868	0.053271	0.313122	0.037573

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.000267	0.000142	0.000309	9.12E-05	0.000351	9.79E-05
PS 34:3	0.036566	0.004333	0.040433	0.005095	0.038815	0.003681
PS 34:2	0.029315	0.003384	0.032394	0.001814	0.031471	0.00266
PS 34:1	0.001214	0.000285	0.001095	0.000297	0.001001	0.000124
PS 36:6	0.001424	0.0003	0.001553	0.000168	0.001376	8.05E-05
PS 36:5	0.003828	0.000365	0.004215	0.00051	0.003951	0.000224
PS 36:4	0.00521	0.000547	0.005384	0.000657	0.005235	0.00026
PS 36:3	0.01458	0.001706	0.017132	0.001544	0.016426	0.001374
PS 36:2	0.009891	0.000972	0.011671	0.001102	0.011146	0.001375
PS 36:1	0.000261	6.3E-05	0.000306	0.000141	0.00024	8.18E-05
PS 38:6	0.000153	5.04E-05	0.000157	5.86E-05	0.000102	2.52E-05
PS 38:5	0.000401	7.77E-05	0.000597	0.000149	0.000482	0.000182
PS 38:4	0.001478	0.00019	0.001601	0.000432	0.001666	0.000268
PS 38:3	0.013619	0.001926	0.015509	0.002176	0.016245	0.001831
PS 38:2	0.010103	0.000995	0.011893	0.00139	0.011975	0.00113
PS 38:1	0.000482	0.000114	0.000465	0.00029	0.000456	6.57E-05
PS 40:4	0.000992	0.000301	0.000874	0.000185	0.001051	0.000199
PS 40:3	0.027351	0.004219	0.028922	0.00514	0.028718	0.002819
PS 40:2	0.018725	0.002553	0.018599	0.003709	0.018516	0.00114
PS 40:1	0.000447	0.000335	0.000641	0.000126	0.00061	0.000237
PS 42:4	0.021003	0.002718	0.019707	0.002937	0.021561	0.002097
PS 42:3	0.063697	0.009567	0.064802	0.01122	0.068934	0.004919
PS 42:2	0.030211	0.00355	0.03159	0.006038	0.033687	0.001065
PS 42:1	0.001476	0.000723	0.001285	0.000454	0.000838	0.000456
PS 44:3	0.002264	0.000423	0.002228	0.00025	0.00208	0.000258
PS 44:2	0.001906	0.000551	0.001745	0.000586	0.001879	0.000312
Total PS	0.296864	0.037877	0.315109	0.041252	0.318813	0.020136

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.000224	8.14E-05	0.000333	9.37E-05	0.000308	3.6E-05
PS 34:3	0.029341	0.005602	0.035851	0.006032	0.034182	0.004082
PS 34:2	0.023905	0.005352	0.028976	0.002438	0.029198	0.003818
PS 34:1	0.001222	0.000244	0.00128	0.000288	0.001004	0.000125
PS 36:6	0.00112	0.000182	0.001256	0.000184	0.001206	0.000201
PS 36:5	0.002953	0.000856	0.003442	0.000713	0.003495	0.000367
PS 36:4	0.004302	0.000769	0.004856	0.000416	0.005037	0.000545
PS 36:3	0.010456	0.002172	0.012559	0.001681	0.013711	0.002014
PS 36:2	0.007327	0.001486	0.008833	0.00164	0.010042	0.001542
PS 36:1	0.00025	0.000111	0.000106	0.000119	0.00013	6.04E-05
PS 38:6	0.000136	9.15E-05	0.000137	9.76E-05	0.000134	3.23E-05
PS 38:5	0.000274	0.000153	0.000399	7.88E-05	0.000407	7.65E-05
PS 38:4	0.001093	0.00028	0.001264	0.000309	0.001504	0.000236
PS 38:3	0.009406	0.002435	0.012027	0.002567	0.01334	0.002537
PS 38:2	0.007416	0.002157	0.009234	0.000759	0.009955	0.001206
PS 38:1	0.000265	0.000194	0.000335	0.000126	0.000352	0.000162
PS 40:4	0.000826	0.000264	0.000966	0.000239	0.000937	0.000218
PS 40:3	0.025499	0.007012	0.02907	0.003358	0.026505	0.004078
PS 40:2	0.017713	0.004051	0.021794	0.003447	0.019713	0.003343
PS 40:1	0.000368	0.00021	0.000625	0.000241	0.000602	0.000193
PS 42:4	0.017662	0.005467	0.020996	0.001918	0.020353	0.003256
PS 42:3	0.059669	0.016277	0.07285	0.009543	0.067706	0.010623
PS 42:2	0.030316	0.007985	0.037924	0.007819	0.033726	0.005223
PS 42:1	0.001672	0.000518	0.001444	0.000351	0.001274	0.000167
PS 44:3	0.002141	0.000602	0.002157	0.0002	0.002031	0.000172
PS 44:2	0.001849	0.000431	0.001863	0.000389	0.001713	0.000337
Total PS	0.257404	0.06254	0.310578	0.031951	0.298565	0.04239

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.000246	5.12E-05	0.000257	5.49E-05	0.00025	5.19E-05
PS 34:3	0.028979	0.003607	0.028587	0.00154	0.031238	0.002959
PS 34:2	0.023885	0.003401	0.023287	0.001601	0.02679	0.002488
PS 34:1	0.000956	9.79E-05	0.000883	0.000102	0.000938	0.000177
PS 36:6	0.001013	0.000157	0.000962	0.000138	0.001025	0.000219
PS 36:5	0.002929	0.000342	0.002791	0.00013	0.003065	0.000355
PS 36:4	0.00443	0.00076	0.004351	0.000242	0.004884	0.000613
PS 36:3	0.013265	0.001635	0.013504	0.000958	0.014649	0.00216
PS 36:2	0.009339	0.00132	0.009362	0.001349	0.0105	0.001311
PS 36:1	0.000224	9.21E-05	0.000204	8.4E-05	0.000282	0.000114
PS 38:6	0.000153	4.89E-05	0.000104	3.76E-05	9.95E-05	3.82E-05
PS 38:5	0.000262	2.36E-05	0.000328	7.27E-05	0.000384	0.000111
PS 38:4	0.001154	0.000258	0.00107	4.28E-05	0.001434	0.000176
PS 38:3	0.012253	0.001598	0.01275	0.000812	0.014338	0.001268
PS 38:2	0.00917	0.00125	0.009488	0.001109	0.01053	0.000988
PS 38:1	0.000348	9.57E-05	0.000423	0.000138	0.000407	0.000167
PS 40:4	0.000708	0.000102	0.000715	2.61E-05	0.00099	8.48E-05
PS 40:3	0.022552	0.001483	0.022236	0.001321	0.025497	0.003162
PS 40:2	0.015246	0.001489	0.015428	0.000728	0.017816	0.001999
PS 40:1	0.000622	0.000168	0.000484	0.000126	0.000568	7.5E-05
PS 42:4	0.018245	0.003069	0.018418	0.001855	0.02005	0.002592
PS 42:3	0.056902	0.007352	0.056149	0.004236	0.0627	0.00919
PS 42:2	0.026653	0.003519	0.026296	0.002162	0.030537	0.004735
PS 42:1	0.000771	0.000214	0.000965	0.000442	0.001019	0.000225
PS 44:3	0.001906	0.000303	0.001773	0.000114	0.001939	0.00031
PS 44:2	0.001563	0.000204	0.00167	0.000272	0.001529	0.000312
Total PS	0.253775	0.029521	0.252486	0.008992	0.283457	0.031897

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PS 34:4	0.000264	3.52E-05	0.000258	4.23E-05	0.000325	7.69E-05
PS 34:3	0.029702	0.003121	0.030427	0.002298	0.03551	0.002621
PS 34:2	0.024328	0.00291	0.024353	0.001999	0.031311	0.002785
PS 34:1	0.001215	0.000265	0.001195	0.000204	0.001232	0.000131
PS 36:6	0.001178	0.000229	0.001124	8.76E-05	0.001229	0.000102
PS 36:5	0.002867	0.000411	0.002967	0.000129	0.003563	0.000371
PS 36:4	0.00406	0.000602	0.004002	0.00014	0.005217	0.000583
PS 36:3	0.011042	0.002266	0.011258	0.00053	0.014792	0.001377
PS 36:2	0.007797	0.001757	0.007874	0.00041	0.011418	0.000975
PS 36:1	0.00024	0.000161	0.00024	0.000126	0.000251	9.5E-05
PS 38:6	0.000148	7.71E-05	0.000155	3.18E-05	0.000154	4.69E-05
PS 38:5	0.000279	7.96E-05	0.000231	5.6E-05	0.000385	5.8E-05
PS 38:4	0.001002	0.000299	0.00106	9.58E-05	0.001514	0.000284
PS 38:3	0.010611	0.002464	0.011456	0.000758	0.014765	0.001725
PS 38:2	0.009085	0.002075	0.009329	0.000934	0.012058	0.001154
PS 38:1	0.000416	0.000154	0.000305	0.000109	0.000481	0.000119
PS 40:4	0.001035	0.000223	0.000865	0.000151	0.000928	0.000207
PS 40:3	0.027151	0.003707	0.027769	0.002171	0.030851	0.000988
PS 40:2	0.019409	0.003029	0.019838	0.002061	0.022277	0.001351
PS 40:1	0.000653	0.00012	0.000611	0.000121	0.000519	0.000204
PS 42:4	0.022298	0.003847	0.022954	0.000822	0.024485	0.001475
PS 42:3	0.075317	0.011218	0.079184	0.005513	0.077107	0.003713
PS 42:2	0.040487	0.005762	0.041158	0.004157	0.039317	0.00269
PS 42:1	0.001471	0.000549	0.001597	0.000161	0.001251	0.000357
PS 44:3	0.002463	0.000339	0.002979	0.000389	0.002456	0.000314
PS 44:2	0.002143	0.000566	0.002425	0.000345	0.002107	0.000212
Total PS	0.296663	0.040628	0.305615	0.019514	0.335501	0.020905

Sample description	Col-o 0hr		101919 0hr		082589 0hr	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.000175	0.000244	0.000854	0.000575	0.000746	0.000503
PA 34:4	0.003003	0.001364	0.0039	0.000932	0.004221	0.001484
PA 34:3	0.034623	0.004071	0.030795	0.006805	0.042054	0.004438
PA 34:2	0.02411	0.004099	0.020623	0.006135	0.02973	0.004532
PA 34:1	0	0	0	0	0	0
PA 36:6	0.008065	0.000917	0.006303	0.00123	0.009614	0.001834
PA 36:5	0.013477	0.002969	0.0113	0.001241	0.019068	0.001379

PA 36:4	0.010475	0.001875	0.009693	0.00203	0.015238	0.001485
PA 36:3	0.004767	0.000799	0.004957	0.001907	0.005586	0.000556
PA 36:2	0.000933	0.000551	0.001384	0.001022	0.000761	0.000176
Total PA	0.099627	0.010287	0.089808	0.01828	0.127017	0.009358

Sample description	Col-o 12hr mock		101919 12hr mock		082589 12hr mock	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.001646	0.001006	0.001766	0.002245	0.001171	0.000392
PA 34:4	0.002777	0.00133	0.002405	0.00049	0.002508	0.001724
PA 34:3	0.044259	0.005958	0.051982	0.010675	0.052578	0.006581
PA 34:2	0.02792	0.00227	0.034159	0.005879	0.037277	0.004414
PA 34:1	0	0	0	0	0	0
PA 36:6	0.010354	0.001652	0.012886	0.001877	0.011146	0.001353
PA 36:5	0.018684	0.002271	0.024612	0.003828	0.023338	0.002952
PA 36:4	0.013851	0.001947	0.0178	0.003226	0.016473	0.002201
PA 36:3	0.005868	0.001808	0.006582	0.003228	0.006913	0.001039
PA 36:2	0.001845	0.00051	0.001458	0.000469	0.001694	0.001141
Total PA	0.127204	0.014145	0.153648	0.023541	0.153096	0.018989

Sample description	Col-o 12hr SA		101919 12hr SA		082589 12hr SA	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.001007	0.001456	0.000973	0.00131	0.000664	0.000539
PA 34:4	0.001376	0.001285	0.00262	0.001278	0.001681	0.000732
PA 34:3	0.037294	0.009214	0.044005	0.005773	0.052179	0.010009
PA 34:2	0.022968	0.005771	0.028375	0.003363	0.03623	0.008662
PA 34:1	0	0	0	0	0	0
PA 36:6	0.009536	0.002178	0.012627	0.002969	0.013398	0.002568
PA 36:5	0.016353	0.003569	0.019294	0.002134	0.025697	0.006192
PA 36:4	0.013105	0.002137	0.015296	0.001267	0.018493	0.004569
PA 36:3	0.005536	0.000827	0.005746	0.000568	0.006534	0.001986
PA 36:2	0.001656	0.001089	0.001102	0.000219	0.00112	0.000411
Total PA	0.108831	0.023665	0.130038	0.009949	0.155995	0.033801

Sample description	Col-o 24hr mock		101919 24hr mock		082589 24hr mock	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.000651	0.0004	0.001	0.000436	0.000909	0.000489
PA 34:4	0.002323	0.001162	0.003412	0.001601	0.002906	0.001358
PA 34:3	0.026342	0.004741	0.033166	0.006522	0.039201	0.013875
PA 34:2	0.01733	0.003618	0.023296	0.006005	0.027634	0.010635
PA 34:1	0	0	0	0	0	0
PA 36:6	0.00578	0.00173	0.00721	0.001221	0.008653	0.003503
PA 36:5	0.011139	0.001807	0.014009	0.004584	0.017796	0.008717
PA 36:4	0.007934	0.00162	0.010623	0.003828	0.014651	0.006083

PA 36:3	0.004266	0.000951	0.004693	0.001116	0.005422	0.001462
PA 36:2	0.000628	0.000376	0.000981	0.000303	0.001212	0.001102
Total PA	0.076393	0.011878	0.098391	0.022456	0.118384	0.045481

Sample description	Col-o 24hr SA		101919 24hr SA		082589 24hr SA	
	ave	stdev	ave	stdev	ave	stdev
PA 34:6	0.001738	0.002282	0.001381	0.00084	0.001004	0.000351
PA 34:4	0.002476	0.001539	0.00415	0.003733	0.003332	0.001207
PA 34:3	0.036844	0.008292	0.042616	0.008629	0.049166	0.018313
PA 34:2	0.022285	0.005254	0.022909	0.003694	0.035102	0.012417
PA 34:1	0	0	0	0	0	0
PA 36:6	0.012163	0.004783	0.011281	0.002982	0.012106	0.004407
PA 36:5	0.015572	0.003374	0.016539	0.003305	0.022244	0.007924
PA 36:4	0.011456	0.001858	0.013111	0.002641	0.019433	0.008769
PA 36:3	0.005638	0.002069	0.004667	0.000786	0.006955	0.002001
PA 36:2	0.000802	0.000325	0.000808	0.000309	0.001229	0.000683
Total PA	0.108973	0.025288	0.117461	0.025309	0.150572	0.055117

TABLE A – 7. PCR PRIMERS

Gene ID	Forward Primer Sequence 5' → 3'	Reverse Primer Sequence 5' → 3'
Salk_101919	GGTAAATTAGATAATGGTTGCCCA	GGCTATATGCCTTAAAGCGGG
Salk_082589	GGTAAATTAGATAATGGTTGCCCA	GGCTATATGCCTTAAAGCGGG
T-DNA LB	GCGTGGACCGCTTGCTGCAAC	GCGTGGACCGCTTGCTGCAAC
<i>ssi2</i>	GGCCATGGATATGGTCAAAC	ATCCAGCGGATCAAAATCTG
At5g14180	TTGTTTG GTGGGGGACATGATCAC ACAGAAGGTGCA	TCGATCTGCCTCATGTCAACAGG
PR1	ATGAATTTACTGGCTATT	ATGAATTTACTGGCTATT
ACTIN	ATGAAGATTAAGGTCGTGGCA	TCCGAGTTGAAGAGGCTAC