Genetic Control of Starch Granule Architecture

Undergraduate Researcher(s): Carla Harper

College of Agriculture/Agronomy

Faculty Advisor(s): Dr. Cliff Weil

ABSTRACT:

Carbohydrate research increasingly is focused on changing the biochemical nature of starch to create more efficient substrates for biofuel production, healthier foods for human consumption and more efficient livestock feed. A key factor in these processes is the rate at which starch is digested by amylases. Starch digestibility is influenced heavily by genetically controlled factors including starch granular and molecular structure and composition. We have begun to characterize genes involved in controlling formation of protein-lined channels connecting the starch granule surface to the central cavity of the granule where digestion is initiated. In addition to starch biosynthetic proteins, these channels contain actin and tubulin, suggesting they are formed by invaginations of the amyloplast created by cytoskeletal elements as the amyloplast expands during starch granule formation. Characterizing the differences between B73 and Mo17 in relative degree of channelization using the IBM population suggests two major loci, on Chromosomes 2 and 3, and a number of minor loci that contribute to channelization. We have defined more dramatic differences among other inbred lines and will be using the NAM lines to map the loci involved and isolate the relevant genes.

Haplotyping Wheat Lines for Fusarium Head Blight Resistance

Undergraduate Researcher(s): Charles T. Zila; Jill R. Recker

College of Agriculture/Agronomy

Faculty Advisor(s): Dr. Herbert W. Ohm; Dr. Xiaorong Shen

ABSTRACT:

Fusarium head blight (FHB) is a devastating disease of wheat (Triticum aestivum L.) that causes reduced grain yield and causes infected grain to be unfit for consumption due to mycotoxins produced by the fungus. The objective of this study was to determine the potential novelty of FHB resistance of two new wheat lines, X117 and 4153 by comparing their haplotypes to the haplotypes at marker loci associated with FHB resistance of other resistant wheat lines. X117 and 4153, and the seven FHB resistant wheat lines with previously mapped resistance - Ning 7840, Frontana, Wangshuibai, Arina, Renan, F201R, and Chokwang, along with the FHB susceptible wheat lines 9762 and 9774 (crossed to X117), and P92226 and P981312 (crossed to 4153), were haplotyped. X117 and 4153 each have been crossed to two FHB susceptible lines to produce recombinant inbred populations; currently, a phenotypic test is concluding on the F5 generation and another test is to be conducted on the F6 generation in the field this summer, with the goal of identifying and mapping their FHB resistance.

Evaluating In-Vitro Maturation Competance with Fatty Acid Supplements in Porcine Oocytes

Undergraduate Researcher(s): Alyssa Diane Auer

College of Agriculture/Animal Sciences

Faculty Advisor(s): Dr. Rebecca Krisher

ABSTRACT:

A large source of potential energy is needed for maturing oocytes and early embryos to metabolize during development. Energy from fatty acid metabolism has consistently been verified as a source of much of the ATP in bovine and porcine oocytes and embryos. Fatty acid metabolism does occur and affects normal oocyte maturation, fertility, and embryo development. In order to metabolize these fatty acids the molecules must first be transported across the Inner Mitochondrial Membrane by an amino acid derivative L-carnitine. The objective of this experiment was to enhance fatty acid metabolism with L-carnitine and the lipid supplement, LipiMate, to increase the development of porcine in-vitro embryos. Due to their large amount of fatty acids, porcine oocytes were used to characterize the effects of enhancing fatty acid metabolism. Oocytes were collected from abattoir-derived ovaries and separated into three groups: Control, L-carnitine supplemented, and LipiMate plus L-carnitine supplemented. The oocytes in each group were matured in TCM199 media and incubated for 40 hours, fertilized in mTBM media and incubated for 6 hours, and cultured in NCSU23 media for 144 hours of incubation. Respective supplements were added to each media. After completing the final incubation, zygotes were assessed by number cleaved and number of blastocysts. The rate of cleavage on day 6 for Control zygotes was 73.0% compared to 29.8% for the L-carnitine supplemented, and 0.0% in LipiMate plus Lcarnitine supplemented. Blastocyst formation rates resulting from number cleaved were 2.6% for the Control, 10.5% for the L-carnitine, and 0.0% for the LipiMate plus L-carnitine supplemented. The results demonstrate that the effects of adding L-carnitine with or without LipiMate to media do not improve the cleavage rate. However, blastocysts were more likely to occur once the cell had cleaved in the Lcarnitine supplemented group.

Histone Methyltransferase GLP Shows Reduced Amount of Transcript from the GV-Stage Oocyte to the 8-Cell Stage in Porcine Parthenogenetic Embryos

Undergraduate Researcher(s): Christine M. Johnson

College of Agriculture/Animal Sciences

Faculty Advisor(s): Dr. Ryan A. Cabot

ABSTRACT:

Mammalian embryos undergo a significant amount of epigenetic modifications during cleavage development. These modifications, including histone methylation and acetylation, have a profound impact on transcriptional regulation and chromatin structure remodeling. Understanding the mechanisms that regulate specific epigenetic changes during this developmental period will provide insight into the pathways that contribute to transcriptional control. The histone methyltransferase GLP (G9a-Like Protein, Eu-HMTase1) dimethylates the lysine 9 residue of histone protein H3 (H3/K9). Dimethylation of H3/K9 is an epigenetic mark that plays a central role in transcriptional regulation, which ultimately results in gene silencing and the formation of inactive heterochromatin. It is also known that H3/K9 dimethylation displays a unique pattern of localization in porcine cleavage development. The aim of this study was to quantify the amount of GLP transcript in porcine oocytes and parthenogenetic embryos. Messenger RNA was isolated from pools of GV and MII oocytes and 4-cell, 8-cell, and blastocyst stage embryos and reverse transcribed. Resultant cDNA was amplified by quantitative, real-time PCR. Reactions were performed such that from each cDNA, transcripts for GLP and YWHAG (control) were amplified in triplicate across 3-4 replicates. Transcripts were then quantified using the comparative CT method. Data were analyzed using one-way ANOVA with Tukey's test for multiple comparisons. Our results show that GLP transcripts were reduced 17-fold from the GV-stage oocyte to the 8-cell stage parthenogenetic embryo (GV vs. 8-cell, p<0.05). No significant changes in transcript abundance were detected between GV and MIIstage oocytes or 4-cell and blastocyst stage embryos.

Evaluating Growth and Carcass Changes in Cull Gilts fed Distiller's Dried Grains with Solubles

Undergraduate Researcher(s): Elizabeth Legan

College of Agriculture/Animal Sciences

Faculty Advisor(s): Dr. Mickey Latour

ABSTRACT:

The purpose of this study was to examine growth and carcass changes in unbred terminal gilts. In this study, gilts (n=12) were fed either whole-corn or Distiller's Dried Grains with Solubles (DDGS) for 14 days. There were significant changes between the two treatments in terms of feed consumption and growth. Over the 14 day period, the whole corn group consumed 755 lbs (342.46 kg), while the DDGS group consumed 206 lbs (93.44 kg). At Day 14, the average body weight of the whole-corn fed pigs was significantly (P<0.05) greater compared to DDGS fed gilts. Primal cuts were compared between treatments and only relative picnic (picnic/hot carcass weight) was significantly (P<0.005) greater compared to whole corn treated animals. There was also a variation in meat color between the animals. The gilts fed DDGS consistently had a darker loin color score compared to whole corn fed gilts. In summary, feeding straight DDGS will significantly alter feed consumption, final body weights, primal cut, and loin color.

Cloning of leishmania major RNA binding protein Lm25.0490 (UPB1)

Undergraduate Researcher(s): Siqi Liu

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. H. L. Weith

ABSTRACT:

Leishmania(Lm) is an intracellular parasite of the immune system targeting macrophages and dendrite cells. The disease Leishmaniasis attacks the populations worldwide. Lm transcripts the entire genome and the regulation of transcription is not quite clear yet which makes it quite necessary to research on the transcription The research was designed to clone RNAmechanism for disease controlling. binding protein UPB1 by PCR, plasmid transformation and His tag affinity chromatography. The gene was amplified by vent polymerase in PCR cycles. The products were ligated to pGEM-T plasmids and the plasmids were transformed to JM 109 cells. The cells proliferated in Luria Broth. We chose the plasmid with right sequence and amplified it by PCR using newly-designed primers with restriction sites (NdeI and NheI). Then we digested the pET 27 plasmids and the PCR products separately with the same restriction enzyme, purified with Wizard PCR preps and made the ligation. The pET 27 plasmid were transformed to BL cells and the proteins were expressed in Terrific Broth. The target protein UPB1 was purified by His-tag and initially confirmed by SDS-electrophoresis. We also extracted the proteins from inclusion body but found no target protein contained. With the purified protein we have gotten, a protein-RNA interaction study is on the way. We are going to study the binding characteristics of UPB-1 to Lm RNA library using a column binding assay.

Extraction of Starch Branching Enzymes (SBE), Amylase and Starch Debranching Enzyme (DBE) from maize endosperm

Undergraduate Researcher(s): Siqi Liu

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. Yuan Yao

ABSTRACT:

A group of enzymes are involved in the biosynthesis of maize starch. The enzymes function differently and will be examined for their effections on starch structure and digestibility associated with starch functional properties and nutrition value. More branched starch has a slower digestibility which is a promising diet for diabetes and people on diet. In term of this, the isolated enzymes will not only benefit fundamental research but also technology development of food industry. This study is designed using preparative-electrophoresis to isolate SBE, Amylase and DBE using protein extraction from 20 days after pollination corn. Also, we will characterize the activity of SBE, Amylase and DBE by microplate iodine staining method. Native-electrophoresis and zymogram on both waxy starch and potato starch basis were further used to characterize SBEI, SBEII and DBE activities.

In Vivo and In Vitro Analysis of Selaginella Moellendorffii Cinnamate-4-Hydroxylase

Undergraduate Researcher(s): Kevin Donohue

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. Clint Chapple

Graduate Advisor(s): Jing-Ke Weng

ABSTRACT:

Phenylpropanoid metabolism is a key biochemical pathway, which gives rise to many important secondary metabolites in plants. Although our current understanding of phenylpropanoid metabolism is confined almost exclusively to higher plants, understanding secondary metabolic pathways in ancient plant species would provide important information with regard to how plants have evolved to cope with life in a terrestrial environment. Spike moss Selaginella moellendorffii represents a plant lineage that diverged from flowering plants 400 million years ago. The genome of Selaginella moellendorffii has recently been sequenced, making this species an ideal platform for comparative biochemical studies. Having already identified the gene encoding a key phenylpropanoid enzyme, cinnamate-4hydroxylase (C4H) in Arabidopsis thaliana, a candidate for a C4H homolog in Selaginella (SmC4H) was identified. The cDNA sequence was isolated and cloned. An Arabidopsis C4H mutant ref3-3 was transformed with the SmC4H candidate and tested for functional complementation. The SmC4H candidate gene was found to restore near wild-type levels of downstream metabolites in this particular mutant. SmC4H was also expressed in yeast, and assayed against various potential substrates in vitro. Its encoded protein displays kinetic constants Km and Vmax towards the substrate cinnamate close to the reported values for C4H in Arabidopsis. These results suggest that Selaginella genome encodes a C4H that is orthologous to angiosperm C4Hs and the early steps of the phenylpropanoid pathway may be widely conserved throughout the evolution of land plants.

Leishmania major RNA-Binding Proteins Purification and Binding Analysis

Undergraduate Researcher(s): Bo Hu

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. H. Lee Weith; Dr. James D. Forney

ABSTRACT:

Leishmania parasites are responsible for Leishmanisis, which is a tropic disease infects 88 countries and 12 millions people, with Leishmania major the most prevalent one. Unfortunately, there are no suitable treatments for this disease: current chemotherapeutic agents are very toxic and no approved vaccines are available. Leishmania major cycles between extra cellular (Promastigote form in sandfly, with flagella) and intracellular (Amastigote form in human, no flagella) habitats and they are highly dependent on regulation of stage-specific genes to survive extreme environmental changes. Though several protein-coding genes and small noncoding RNAs are identified to have different level of expression, the mechanism and reason of the process is unclear yet. Currently, posttranscriptional gene regulation is found to mediate changes when Leishmania move from sandflies into humans. While RNA binding proteins play an important role in the processing, transportation, and degradation of RNAs, it is hypothesized that RNA binding proteins bind and stabilize certain mRNAs during the stage transition of Leishmania major. The study of the mechanism of regulation and the role of RNAbinding proteins in posttranscriptional regulation may represent therapeutic targets for Leishmanisis. Analysis of Leishmania major open reading frames with homologous DNA sequences predicts there are 43 RNA-binding protein domains encoded in the genome. In this experiment, putative RNA-binding protein LmjF09.0060 is cloned and purified, further experiments are conducted to analyze the structure and binding specificity of this protein.

RNAi analysis of DNA polymerases in Paramecium tetraurelia

Undergraduate Researcher(s): Michael Kalwat

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. James Forney

ABSTRACT:

Ciliates possess two kinds of nuclei within a single cell: a polygenomic, transcriptionally active macronucleus and a diploid, transcriptionally silent, germline micronucleus. During sexual reproduction the old macronucleus is destroyed and a new macronuclear genome is formed from the micronuclear DNA. Extensive genome reorganization occurs during macronuclear development including DNA amplification, chromosome fragmentation and DNA elimination through site-specific splicing reactions. Our long-term goal is to identify specific DNA polymerases involved in macronuclear development. In this study, homologues of known DNA polymerases were identified in the Paramecium tetraurelia genome using bioinformatics. Gene fragments from delta and beta DNA polymerase homologues were amplified using PCR and cloned into plasmid vectors for use in RNA interference (RNAi) assays. RNAi induced gene silencing is accomplished by feeding Paramecium bacteria that produce double-strand RNA from the corresponding Paramecium gene. This method provides a simple technique for the knock-down of expression from specific Paramecium genes. RNAi against the delta polymerase catalytic subunit significantly reduced the frequency of cell division during vegetative growth. Similar experiments with the beta DNA polymerase genes had no effect on cell division during vegetative growth. Other beta DNA polymerases are known to function in DNA repair pathways. Paramecium beta polymerases may be required for repair of DNA splicing events during macronuclear development and assays to examine DNA polymerase function during formation of the macronuclear genome are under development. The results demonstrate that RNAi gene silencing can be used as an initial screen for DNA polymerase function in Paramecium.

Role of GSTs in Herbicide Tolerance

Undergraduate Researcher(s): Kyle Mohler

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Faculty Advisor(s): Dr. Peter Goldsbrough

Graduate Advisor(s): Metha Meetam

ABSTRACT:

Glutathione-S-transferases (GSTs) have a role in detoxification of xenobiotics such as the agronomically important herbicide, alachlor. Herbicide safeners are chemicals used to increase tolerance of some crop plants to application of herbicides. In Arabidopsis thaliana, treatment with herbicide safeners increases RNA expression of a GST gene, AtGSTU19 but did not make plants tolerant to alachlor. One hypothesis to explain these results is that AtGSTU19 is not expressed in the appropriate tissues or at a sufficient level to provide alachlor tolerance. Alternatively, AtGSTU19 may be unable to detoxify alachlor because of its biochemical properties. To test these hypotheses I analyzed transgenic plants that expressed three different GSTs (AtGSTU19 from Arabidopsis, TtGSTU1 from Triticum tauschii, and ZmGST4 from Zea mays). Expression of the GST genes was driven by the 35S promoter. Plants were tested for resistance to alachlor when the herbicide was supplied in an agar medium, applied to leaves of soil-grown plants, or applied to soil where seeds were germinating. Level of tolerance was measured by root elongation and shoot dry weight of transgenic plants grown on plates. Shoot dry weight was also measured from plants grown in soil. Results show that expression of ZmGST4 from Z. mays confers greater resistance to alachlor than either TtGSTU1 or overexpressed AtGSTU19. To test the efficiency of these three GSTs in alachlor conjugation, they were expressed in E. coli. The purified proteins will be analyzed for herbicide conjugation properties. To test the hypothesis that expression of the maize GST in epidermal cells was sufficient to confer resistance, ZmGST4 was expressed in different tissues using shoot- and epidermis-specific promoters. The herbicide tolerance of these plants will be evaluated in experiments similar to those described above.

Characterization of a Novel APC/C Inhibitor

Undergraduate Researcher(s): Julie Chaney

College of Agriculture/Biochemistry

Faculty Advisor(s): Dr. Mark Hall

ABSTRACT:

The anaphase promoting complex/cyclosome(APC/C) is a protein complex which functions to regulate progression through the cell cycle by tagging certain cell cycle related proteins with ubiquitin, leading to their destruction by proteolysis. In vivo studies suggest that Acm1 acts as an inhibitor of the APC/C, apparently by binding the APC/C coactivator Cdh1. Cdh1 is thought to promote APC/C activity by binding to specific substrates and recruiting them to the APC/C. We hypothesize that Acm1 acts as a competitive inhibitor of this process, blocking substrate binding to Cdh1 by occupying the substrate binding sites in the Cdh1 WD40 domain. To test this hypothesis, I plan to perform an in vitro binding assay, using purified, recombinant proteins. A binding assay using purified Cdh1 WD40 domain will test whether Acm1 can bind to this region. A second assay using purified Cdh1, Acm1, and the substrate Clb2 will test whether Acm1 can block Clb2 binding by a competitive mechanism.

Glyphosate Dose-Response of Selected Indiana Horseweed Biotypes

Undergraduate Researcher(s): Janelle Donahue

College of Agriculture/Botany and Plant Pathology

Faculty Advisor(s): Dr. William G. Johnson

Graduate Advisor(s): Vince Davis; Greg Kruger

ABSTRACT:

Glyphosate-resistant horseweed (Conyza canadensis) biotypes have been reported in 14 states. Populations from several states have demonstrated glyphosate tolerance in dose-response experiments. However, there is little information about the inheritance of variable levels of glyphosate tolerance in horseweed. The objective of this experiment is to determine if the rank in levels of glyphosate tolerance among first generation progeny corresponds to the rank in tolerance from respective maternal parents. Initial glyphosate screens were conducted on horseweed populations comprised of 40 composite mother plants. Resistant survivors that demonstrated varying levels of glyphosate tolerance were identified and allowed to self-pollinate. Seeds from individual plants were collected and grown in the greenhouse. Three experimental runs with plants 2 to 4 centimeters in diameter were sprayed with 0, 0.11, 0.21, 0.42, 0.84, 1.68, 3.36, 6.72, and 13.44 kg ae/ha of glyphosate and replica ted 4 times. At 28 days after treatment (DAT), horseweed rosette widths were measured, individual plants were rated for visual control on a scale of 0 to 100, and plants were harvested for fresh and dry weight biomass production. The correlation between rankings of glyphosate tolerance levels from the mother plant to respective progeny was poor for most growth parameters. However, the ranking of glyphosate tolerance in the mother plants corresponded well with progeny survival at the 1.68 kg ae/ha rate. Mother plants with a "high" level of resistance had progeny survival of 92%, while a population with a "low" level of resistance had progeny survival of 25% at the 1.68 kg ae/ha rate.

Determining the Physiological Response of Plants to Rooftop Soil Media

Undergraduate Researcher(s): Patricia Quackenbush

College of Agriculture/Botany and Plant Pathology

Faculty Advisor(s): Dr. Mary Alice Webb; Dr. Kevin Gibson; Dr. Brad Joern; Dr. James Camberato

ABSTRACT:

As the rate of urbanization continues to increase in major metropolitan cities and small suburban areas, public horticulturists, urban planners and landscape architects have been using green roof technology to help combat the urban heat sink effect and to assist in lowering heating and cooling costs in buildings. Using green roofs can also aid in preserving the greenspace in an already heavily populated area. With designing green roofs come specially engineered soils that not only are light enough to hold plant material and not collapse the roof, but to provide proper plant nutrition as well. In the case of the Indianapolis Museum of Art, which has a green roof installed over its underground parking garage, plant nutrition is poor and treatments to fix this problem must be looked at. Preliminary soil tests and water test indicate that this structured soil has a calcareous profile with high pH and very high calcium content. Low organic matter and low cation exchange capacity values (CEC) indicate a low buffering capacity in the soil. Irrigation water is high in calcium and bicarbonate content, indicating calcium loading into the soil. Continued work with Botany and Agronomy departments in designing fertilizer management plans, soil amendments, irrigation amendments and possible plant species to place on the soil will potentially help remediate the soil into a better media for plant growth and development.

Characterization of mur5 and mur6, two low cell-wall arabinose mutants in Arabidopsis thaliana

Undergraduate Researcher(s): Rachel A. Mertz

College of Agriculture/Botany and Plant Pathology

Faculty Advisor(s): Dr. Nicholas C. Carpita

ABSTRACT:

The plant cell wall is a rigid but dynamic skeleton that controls cell expansion and development into final form. It protects cells from several biotic and abiotic environmental stresses. Knowledge of the locations of the genes responsible for cell wall biosynthesis and polysaccharide distribution and function is critical to our understanding of plant growth, development, and biomass accumulation. Understanding of gene function is a valuable tool in the characterization and propagation of plants with novel phenotypes that are uniquely suited to human agricultural, pharmaceutical, and energy needs. My goal is to identify the defective genes responsible for mur5 and mur6, two cell wall arabinose-deficient Arabidopsis thaliana mutants. These mutants are thought to be non-allelic, and have chemical and structural cell properties distinct from each other and wild type. A mapping population was made by cross-pollination of parent populations of a wild type WS ecotype, mur5, and mur6 in the parental Colombia ecotype, and the resulting F1 population then self-pollinated to produce a segregating recombinant F2 population. Low arabinose-phenotypes are being mapped using the unique molecular markers from the parent genotypes, narrowing down the range in which the underlying genes responsible for the mur5 and mur6 phenotypes may be located. An allelism test was also performed by cross-pollination of mur5 and mur6. Additionally, isolation and chemical analysis of cell walls of the parent and F2 populations will be conducted, and an alditol acetate assay will be performed to ascertain and compare the levels of the seven major sugars that comprise the cell wall. A linkage (methylation) analysis will provide more detailed information about the nature of the low-arabinose phenotype and the specific polysaccharides affected.

mtDNA Barcoding for Taxonomic Identification Within the Genus Agrilus

Undergraduate Researcher(s): John Shukle

College of Agriculture/Entomology

Faculty Advisor(s): Dr. Jeff Holland

ABSTRACT:

Species identification in the genus Agrilus is difficult, and often requires dissection and examination of the male adeagus. This method, however, excludes both females and larvae from positive identification. Ecological studies that rely on accurate species identification may be complicated and time consuming due to these taxonomic difficulties. Furthermore, increases in the number of invasive species highlight the need for faster identification of potentially harmful exotics. Recent studies have shown the efficacy of mtDNA barcoding to differentiate morphologically similar species. I used a 658bp fragment internal in the cytochrome c oxidase I (COI) gene to evaluate the effectiveness of this technique within the genus Agrilus. Results indicate that mtDNA barcoding using the COI gene will provide a viable method to distinguish between species in the genus.

Air Permeability Across a Chicken Egg Membrane

Undergraduate Researcher(s): Allison Clemons; Anne Spantzel

College of Agriculture/Food Science

Faculty Advisor(s): Dr. Kevin Keener

ABSTRACT:

Chicken egg membranes serve an important function in eggs. In fertilized eggs, they allow gas exchange to occur during embryo development. In unfertilized eggs (table eggs), reducing gas exchange would preserve egg quality and extend shelf-life. During gas exchange, carbon dioxide is released and air enters via the membrane. In this study, air permeability of fresh chicken eggs was measured. The membrane was physically separated from the shell after egg contents were removed. Membrane pieces of approximately one square centimeter were attached to a gas flow apparatus and air permeability was measured. These measurements were performed on acid treated and untreated membranes over a range of moisture contents (54% - 85%). To treat eggs with acid, whole eggs were submerged in vinegar (5% acidity) for approximately two days at room temperature. The vinegar removed the shell while leaving the membrane intact. Whole membranes of vinegar treated eggs were evaluated. The membrane pieces were weighed before and after testing to determine moisture content. Multiple permeability measurements were performed on each membrane at different moisture contents. After testing, the membrane pieces were oven dried. Data suggests that the lower the moisture content of an egg membrane, the higher the air permeability. Acid treatment greatly reduces permeability of the membrane. Comparison of gas permeability between two membranes of similar moisture content, one with acid treatment and one without, shows a tenfold decrease in gas permeability. This difference is likely due to protein denaturation resulting from acid treatment. The knowledge gained from this study will allow development of a process to treat chicken eggs and reduce gas permeability of their membranes, potentially extending egg shelf-life.

Preparation and Characterization of Pregelatinized Starch

Undergraduate Researcher(s): Irma Amelia

College of Agriculture/Food Science

Faculty Advisor(s): Dr. James N. BeMiller

ABSTRACT:

Two main types of pregelatinized starch are produced: those made using a hot roll and those made using an extruder. Both of these methods produce starch polymer fragmentation, which is evident from Size Exclusion Chromatography. Ungelatinized birefringent granules are also seen in some commercial preparations. In pregelatinized starch made using a hot roll, the starch polymer fragmentation probably occurs during grinding of the flakes. Meanwhile, in pregelatinized starch made in an extruder, the fragmentation probably results from the high shear in the extruder. The objective of this research is to prepare pregelatinized starches that are completely amorphous (no crystalline structure) and unfragmented. The resulting products were then fragmented with acid and the effect of this fragmentation on the degree of stickiness / tackiness determined. Starch slurries were heated to $100 \hat{A}^{\circ} C$ with rapid stirring to cook the starch. Solubilized starch molecules were precipitated by addition of a water-soluble organic solvent before they had had a chance to recrystallize. Another approach was to freeze dry a cooked starch paste (applicable to waxy corn starch). The degree of depolymerization/fragmentation was determined by size-exclusion HPLC and a Rapid Visco Analyzer (RVA). The degree of crystallinity was observed from x-ray powder diffraction results. The adhesive/sticky nature of the non-crystalline starch was determined using a TA-XTPlus Texture Analyzer. It has been found that pregelatinized starch can be prepared by precipitation of a paste with ethanol; however, the resulting products would complex with ethanol and cause crystallinity. This product is then cooked, and the hot paste is precipitated with acetone to produce amorphous pregelatinized normal corn starch. The amorphous property is evident form x-ray powder diffraction. The precipitation method with an organic solvent was not applicable for the preparation of pregelatinized waxy corn starch because it produced a sticky, cohesive and adhesive mass. Therefore, freeze drying was used to prepare amorphous pregelatinized waxy corn starch. The acid hydrolysis reduced molecular weight of amylopectin more extensively in normal

corn starch than in waxy corn starch. The texture analyzer data showed that a higher degree of hydrolysis in pregelatnized normal corn starch increased stickiness up to a maximum, after which the degree of adhesiveness decreased. In all aspects of this research, it was clears that the behavior of waxy corn starch was rather different than that of normal corn starch. This research contributes to a basic understanding of the sticky nature of food products. It also helps to determine the effect of milling on producing stickiness in foods.

Stable isotope signatures and preliminary contaminant analyses of stratified Great Blue Heron colonies across the state of Indiana

Undergraduate Researcher(s): Zachary Bagley

College of Agriculture/Forestry and Natural Resources

Faculty Advisor(s): Dr. Marisol Sepulveda

Graduate Advisor(s): Stephanie Baker

ABSTRACT:

Over a two year period data was collected from six Great Blue Heron colonies stratified into three distinct regions across the state of Indiana. Through a process involving egg albumen, nitrogen and carbon stable isotope signatures along with PCB, PAH, and OCP contaminant levels were assimilated from the original samples. This data has now been analyzed using the Statistical Package for Social Sciences to reveal relationships between stable isotopes and particular contaminants and also between stable isotope data by region. What we desire to make evident with this research is the feeding level of the Great Blue Heron within the state of Indiana in the food chain and how this affects contaminant levels, and ultimately breeding success of these birds.

Age and Growth of Longnose and Shortnose Gar on the Wabash River

Undergraduate Researcher(s): Jessica L. Hoffmeister

College of Agriculture/Forestry and Naturual Resources

Faculty Advisor(s): Dr. Trent M. Sutton

Graduate Advisor(s): Rebecca A. Zeiber

ABSTRACT:

Longnose gar Lepisosteus osseus and shortnose gar L. platostomus are one of North Americaâ€[™]s oldest fish groups and are abundant in the Wabash River. Little is known about these two fish species because they are considered undesirable, although they fulfill an important ecological niche by maintaining stable fish populations. We collected 77 longnose gar and 118 shortnose gar from the middle and lower Wabash River using boat electrofishing from June through August 2006 to assess the population characteristics of these two species. Mean total length of longnose gar was 749 mm (range, 492 to 1,375 mm), and the mean wet weight was 1.10 kg (range, 0.25 to 6.6 kg). The mean total length of shortnose gar was 584 mm (range, 498 to 776 mm) and mean wet weight was 0.65 kg (range, 0.35 to 1.9 kg). Longnose gar ranged in age from 1 through 16, while shortnose gar ranged from 1 to 12 years of age. The von Bertalanffy growth relationship indicated that the theoretical maximum length (Lâ^ž) of longnose gar in the Wabash River was 1,004.8 mm, the Brody growth coefficient (K) was 0.058, and the time period when fish body length equals 0 mm (t0) was -10, while shortnose gar had Lâ^ž of 898.8, K of 0.182, and t0 of -4.569. Annual total mortality was 27.54% and 40.22% for longnose and shortnose gar, respectively. The results of this research will increase our understanding of the stock structure and population dynamics of this species in the Wabash River.

Physiological Analysis of Arabidopsis thaliana Mutants

Undergraduate Researcher(s): Amanda Mitchell

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Burkhard Schulz

Graduate Advisor(s): Renate Weizbauer

ABSTRACT:

Currently, research is being completed to analyze the differences that may be observed between various Arabidopsis thaliana mutants. The experiment has been designed so that each different mutant is exposed to the same environment and not any one plant has an advantage over another. At the conclusion of the experiment, data will be collected which compares differences in germination time, flowering time (days after germination until bolt is 1cm long), overall plant height, fresh weight versus dry weight(water content of the plant), the number of seeds produced, the size of the seeds produced, number of rosette leaves, total leaf number, length of vegetative and reproductive phase, number of 'branches', leaf length, silique length and rosette circumference. The seeds of eight different Arabidopsis mutants were planted out in petri dishes and then placed in a seed stratification cooler. Following this cold treatment, the seeds were placed in a culture room and when germination had occurred, they were planted out into soil. The plants may also be subjected to different environmental treatments, which include being grown during long days and short days. The control for this part of the experiment will be grown in the greenhouse under normal conditions.

Arabidopsis TWISTED DWARF1 (TWD1) as a model system for â€efunctional phylogeny†studies

Undergraduate Researcher(s): Steve Holladay

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Burkhard Shultz

ABSTRACT:

Immunophilins are receptors for immunosupressants which block T-cell receptor mediated signal transduction in mammals. These proteins are ubiquitous in all organisms from bacteria to animals and plants. Plants have a great number of immunophilins but no immune system, what is their role in plants? Plant immunophilins are found in multiple developmental processes. The membranebound FK506-binding (FKBP) immunophilin, TWISTED DWARF1 (TWD1) regulates the polar export of auxin by ABC transporters PGP1 and PGP19. Knockouts of TWD1 in Arabidopsis result in dwarfism and disorientated growth. Is regulation of polar auxin transport a common function for TWD1 in plants? A phylogenetic approach of cross-species complementation of knock-out mutants will be used to determine the function of TWD1 orthologs. The goal is to elucidate the role of TWD1 in plant development. Arabidopsis twd1 mutants are a test system for functional complementation by transformation with TWD1 orthologs from other species. Depending on amino acid (aa) alterations acquired during evolution, a more or less complete functional complementation of twd1 phenotype should be visible. Sequence comparisons of the different proteins will reveal which as positions are altered and which alterations are essential for complete complementation. Likewise, aa alterations without effects will also be identified. Together with protein structure information this approach should give us insight into the function of an immunophilin in its cellular context at very detailed level. TWD1 homologs from dicots tomato and C. rubella have been successfully used as proof of concept. Here we report on cloning and complementation/transformation of TWD1 homologs from the monocots rice and corn in twd1 of Arabidopsis.

Sweet Research on Bitter Rot

Undergraduate Researcher(s): Karen Mitchell

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Janna Beckerman

ABSTRACT:

Bitter rot is a common disease of apples and pears, especially in warm, wet environments, which appears to be increasing in Indiana. Bitter rot, caused by Colletotrichum gloeosporioides or Colletotrichum acutatum, produces symptoms of light to dark brown, sunken lesions on the fruit. Although bitter rot can be controlled with fungicide applications consistently throughout the growing season, many of the more effective fungicides now have restrictions limiting their use to 77 days before harvest, which is a vulnerable period of time for the fruit. There are two hypotheses to explain the increase in bitter rot infections in Indiana orchards, either the restrictions on fungicide applications have left fruit more susceptible to infection or the Colletotrichum species have evolved fungicide resistance. To test these hypotheses, I have collected cultures of Colletotrichum from Indiana orchards. I am identifying the species through the use of PCR, and I am testing isolates for fungicide resistance by poison plate assay. Initial assays suggest a resistance to low levels of fungicide.

Interactions of proteins with naturally occurring clay minerals

Undergraduate Researcher(s): Joyce Lok

College of Agriculture/Agronomy

Faculty Advisor(s): Dr. Cliff Johnston

ABSTRACT:

Research on the interaction of biological molecules, such as proteins, with clay minerals has not been studied in depth. This project will focus on understanding the interactions of three model proteins sorbed onto smectite, an expandable 2:1 phyllosilicate. Smectites are common clay minerals found in soils throughout the world. To better understand the interactions of proteins with clay minerals, three proteins, lysozyme, bovine serum albumin (BSA), and fibrinogen, were sorbed onto Na-saponite in an aqueous solution. Important protein characteristics for the selected proteins were their size and shape, isoelectric point, and binding properties. The interactions of sorbed proteins onto clay minerals were analyzed using sorption, structural and spectroscopic methods. Protein sorption isotherms revealed that all three proteins showed a high affinity for Na-exchange saponite. X-ray diffraction analysis on powdered samples indicated an interlayer sorption for lysozyme and BSA having d-spacings approaching 4.5 nm. There was no indication of interlayer sorption for fibrinogen which was attributed to its large size and inability to \hat{a} €fit \hat{a} €M in the clay interlayer. Fourier transform infrared (FTIR) spectroscopic analysis provided information about the conformation of the protein on the clay surface and the role of water.

A Preliminary Evaluation of the Reproductive Physiology of the Bluntnose Minnow, Pimpehales notatus.

Undergraduate Researcher(s): Nathan T. Barton

College of Agriculture/Forestry and Natural Resources

Faculty Advisor(s): Dr. Maria S. Sepulveda

Graduate Advisor(s): Sonia Mae Johns

ABSTRACT:

The bluntnose minnow (Pimephales notatus) is endemic to the state of Indiana, and is commonly found in many streams and rivers. Despite the relative widespread distribution and abundance of bluntnose minnows in the state, there are no studies describing the reproductive physiology and normal reproductive parameters of this species. The objective of this study was to obtain some preliminary information on the reproductive physiology of this species. For this purpose, we evaluated several reproductive parameters from minnows collected from Tippecanoe River, including body weights and lengths, secondary sex characteristics, gonadosomatic indices and gonad histology. In addition, we established a captive colony of bluntnose minnows, and are currently testing different spawning protocols.

Poster# 56 / Humanities/Social Science

Export Recovery Following Livestock Disease Outbreaks

Undergraduate Researcher(s): Jeanna Pitstick

College of Agriculture/Agricultural Economics

Faculty Advisor(s): Dr. Phillip Paarlberg

ABSTRACT:

Livestock disease outbreaks are greatly affecting international trade. The countries that are experiencing these outbreaks are having a decrease in their exports. The longer it takes for trade to recover the more severe the economic losses are. The ability to anticipate how large these export losses will be from livestock disease outbreaks depends on how fast exports recover and the factors that influence this recovery. The diseases of interest are Foot and Mouth Disease (FMD), Highly Pathogen Avian Influenza (HPAI), and Classical Swine Fever (CSF) because each has unique characteristics. Foot and Mouth Disease affects multiple species of cloven hoofed animals. However, the disease is not transmissible to humans. Highly Pathogen Avian Influenza affects mainly the avian species, but can be transmissible to humans causing a very high fatality rate. Classical Swine Fever is a single species disease affecting only swine. The general objective is to research the extent of the economic losses, the speed at which trade recovers, and the factors that affect recovery after an outbreak.

Poster# 57 / Humanities/Social Science

Hedonic Pricing of Bulls

Undergraduate Researcher(s): Jenna Smith

College of Agriculture/Agricultural Economics

Faculty Advisor(s): Dr. Kenneth Foster

ABSTRACT:

Bulls account for half of the genetic input when making improvements in cattle herds. Changing bulls is less costly than changing cows; therefore it is often the case that bulls account for more rapid improvements in heritable traits. One of the problems that breeders who supply bulls face is that the attributes of bulls come bundled together so that it is difficult to determine what the value of improvements in a bull might be worth. This research estimates what values beef producers implicitly place on particular characteristics when deciding which bull will best fit the needs of their farm. A hedonic pricing model was estimated using ordinary least squares on actual transaction data and reveals the value buyers of bulls implicitly place on specific traits. For example, a ribeye area of 12.8 in 2 at the mean sale price reveals a buyer would be willing to pay an additional \$80.39 for a bull with an additional square inch. Likewise, a bull with a 1242 lb. 365-day weight at the mean sale price reveals a buyer would be willing to pay an additional \$1.83 for an additional pound. Therefore, this research reveals an incentive for producers of bulls to focus on improving the genetic make-up of the bulls they offer for sale.

Pilsen: A New Beginning - C.R.A.I.L. research summary

Undergraduate Researcher(s): Matthew Strange

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Kim Wilson

ABSTRACT:

SUMMARY My research project began as a study of whether or not physical design can create more viable communities. Once adequate research was compiled and analyzed, I determined that design can in fact impact a community's viability, but none of the existing design strategies addressed the sustainability of a community in a wholistic way to prioritize development. Thus, my research project has evolved into the creation of an original sustainable community design strategy called C.R.A.I.L. that synthesizes many existing design strategies with the goal of creating more viable communities. The method will be evaluated over the course of this semester on an actual community in the Chicago area in which the principles will be used to analyze and develop a framework plan for the community's development. A more complete explanation of the process is listed below: RESEARCH OBJECTIVE It is obvious that community design can create more visually beautiful neighborhoods, but my interest was whether these strategies can actually create more viable communities. I began by researching both what defines a community and viability and ultimately where they overlap. A community can be defined in two ways. The first, and more crude, definition is a rigidly defined (by streets or geographic features) area such as a city or any neighborhood within. The second definition is more fluid in nature as it looks at a group with a set of commonalities such as an ethnic group, religious body, a ecological community. Both definitions mutually reinforce one another; a network of roadways without the human dimension lacks a sense of place and likewise, groups of people sharing commonalities but physical definition lack cohesiveness. Thus, the ellusive "sense of community" is a defined locallity that fosters a climate of mutual respect, appreciation, and cohesiveness between all of its sub-communities. Viability, or sustainability as it is more often named, is the quality and balance between the environmental, social, and economic factors of a community and the ability for these factors to be sustained into the indefinite long term. Similarly to the definition of

community, the segments of sustainability also have a symbiotic relationship; a change to any one of the three factors usually has implications for the others. Vital communities have a capacity to grow and develop, respect the environmental, social, and economic frameworks of the community as a whole, are easily accessible physically, culturally, and economically, have a positive sense of identity, and have a solid strategy to extend the ifecycles of each of the three factors of viability. **EVALUATION** Once I had a stronger understanding of what defines a sustainable community, I began evaluating a variety of community types and strategies from New Urbanism to suburban sprawl based on data indicating how positively they impacted the environmental, social, and economic conditions of a community. The research determined conclusively that physical design can impact the long-term viability of a community. However, all of the strategies had negative elements and none was a comprehensive, easy to implement strategy to improve sustainability. That inspired the C.R.A.I.L. method for community design which synthesizes the positives from many existing strategies while focussing on prioritizing development efforts to improve overall sustainability. C.R.A.I.L. Method The C.R.A.I.L. method is founded upon the premise that design can in fact influence the viability of a community when these physical design factors have a direct relationship to the factors that influence a community's social, environmental, and economic sustainability. It was also one of our objectives to create a method that was not only effective when building a new community, but a series of strategies that could be retrofit onto any community. Our method is a synthesis of many different community design approaches, where we have integrated the positive attributes of each while minimizing the negatives. The C.R.A.I.L. method distills the concept of environmental, social, and economic sustainability into measurable components evaluated on a spectrum based on C.R.A.I.L. evaluation criteria. The goal of C.R.A.I.L. is to use these components to identify priorities in the design and development of a community - when strategies are implemented to individually improve each letter of the acronym, the overall sustainability of the community can be improved. The acronym breaks community sustainability down into the following: Capacity The potential for the environmental/physical, social/cultural and economic components of a community to grow positively. A community with high capacity not only has a strong foundation and opportunities for positive growth, but a population that is proactive and willing to accept change. Respect The level to which the design of a community respects the environmental resources of its land, the cultural history and lifestyle patterns of its people, and its local and governmental economy. It also refers to how well the residents of the community respect their community's resources. A community with high respect positively represents the lifestyles of its residents and is well-maintained by residents. Accessibility The level to which the community can be accessed physically. culturally/socially, and economically by both its residents and visitors. A community with high accessibility has clear circulation and spatial organization which educates and culturally elevates its residents and visitors. Funding for community improvement projects is easily accessible and is allocated appropriately. Identity The collective of a community's environmental, social, and economic conditions that gives the community a unique and positive representation to residents and visitors.

A community with a strong identity has a genus loci (sense of place) that positively represents the culture of its people, the integrity of its environmental resources, and the financial realities of the community. Lifecycle The lifespan and ability for physical, social, and economic resources to be adaptively reused to evolve in a positive fashion. A community with a high lifecycle score has effective strategies in place to allow their environmental resources to be sustained and recycled, the social climate to positively change and maintain a sense of harmony, and the economic conditions of a community to constantly improve. Using the C.R.A.I.L. Chart Once the designer evaluates the community using the aforementioned criteria, these values are plugged into their corresponding place on the chart along with a reasonable explanation of why the community received the score. Add the values vertically to determine the value for each letter of the acronym, and horizontally to determine the overall environmental, social, and economic sustainability. These totals are intended to provide insight about where to prioritize development efforts. To determine the C.R.A.I.L. index which evaluates the overall sustainability of the community, add the totals of each letter of the acronym to the sustainability totals. The C.R.A.I.L. index for the community can be evaluated as follows: 0-1500: Unsustainable 1501-3000: Moderately Sustainable 3001-4500: Sustainable

Poster# 63 / Humanities/Social Science

Social Responsibility in Landscape Architecture

Undergraduate Researcher(s): David Witte

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Faculty Advisor(s): Dr. Kim Wilson

ABSTRACT:

The growing trend of Environmental Sustainability and the environmentally responsible approaches to Landscape Architecture have been positive steps in the development of our profession. From its inception Landscape Architecture has been a profession focused on the environment and its relationship to the people who are part of it. However the role of the individual and the community seems to take a backseat in many of the decisions that are being made in regards to planning and design. Addressing the importance of Social Sustainability and the Social responsibility companies need to adhere to in order to better serve the communities they are designing for will be the main focus of this project. By means of this I intend to address the role Landscape Architects should play in social issues such as homelessness, poverty and Social inequality.

Poster# 64 / Humanities/Social Science

Design Solutions for Stormwater

Undergraduate Researcher(s): Cameron Hull

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Kim Wilson

ABSTRACT:

Everyday the future of the world becomes less and less certain. With the list of maladies, most caused be the wanton destructive nature of man, growing daily, we must take action to curtail the avalanche before all is lost in the name progress. As Landscape Architects the burden falls to us to find ways, both large and small, to bind man and earth together. Stormwater problems, quantity and quality, is one of the facets of environmental degradation that must be managed and solved if we, as a people, intend to survive and grow. The solutions can be found in the study and extraction of natural processes to influence and replace the failing design strategies of old. Guided by the knowledge of nature, new ways of design and planning have been developed that not only control and treat stormwater, but enhance the aesthetic beauty of the built world.

Envisioning Gateway

Undergraduate Researcher(s): Reuben K. Verkamp

College of Agriculture/Horticulture and Landscape Architecture

Faculty Advisor(s): Dr. Kim L. Wilson

ABSTRACT:

Gateway National Recreation Area occupies 26,600 acres in the New York-New Jersey Harbor. The land has been a part of the US National Park System since 1972. Van Alen Institute, in partnership with Columbia University Graduate School of Architecture, Planning + Preservation and National Park Conservation Association, is sponsoring an international design competition, "proposing innovative, critical and compelling designs that celebrate the unique potential of this park as both a regional asset and a national treasure." The park has been divided into three units, Jamaica Bay, Staten Island, and Sandy Hook. Existing natural resources include a wildlife refuge, ocean dunes, coastal uplands, and a holly forest. Gateway National Recreation Area also offers many historical resources including lighthouses, centuries old military forts and relics from the early days of aviation. Visitors can partake in a wide range of activities such as swimming, boating, fishing, team sports, bicycling, nature study and educational services. The intent of this design exercise is to explore the influence of the Gateway National Recreation Area on the surrounding region and vice versa, with the hopes of increasing its potential as a regional feature and national treasure. The first phase of the project is research based and analytical in nature. The second phase explores design solutions. The emphasis of this study is to create a sustainable design solution, not only environmentally but also socially and economically.