

DISSERTATION SUMMARIES

Biogas production from chicken manure

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Biomass based technologies have several benefits: the necessary substrates are available in large quantities, the generated energy carriers are environmentally friendly, easy to carry and store. Biogas is a renewable energy carrier which is similar to natural gas and can be used in practically all applications to replace natural gas. It is produced by anaerobic fermentation of the organic materials. The problem of poultry waste management has been an increasing concern in many parts of the world due to the huge amount of this waste stream. Pollutants from improperly managed poultry waste can cause serious environmental problems in terms of water, air, and health quality.

Chicken manure (CM) contains two main forms of nitrogen: uric acid and undigested proteins, which represent 70% and 30% of the total nitrogen in CM, respectively. Anaerobic decomposition of uric acid and undigested proteins in CM results in high amounts of unionized ammonia and ammonium ions. Excess ammonia inhibits the anaerobic microbial community and thus methane production. Because of the regular antibiotic treatment in the large-scale chicken plants, first we compared two types of CM containing straw bedding: samples were taken from broiler chickens receiving antibiotic treatment and from untreated chickens. The wide-spectrum antibiotics employed at the chicken farms negatively affected the biogas producing microbial community leading to rapid process failure. The antibiotic-free substrate was more suitable for microbial decomposition. Increasing the organic loading in the batch reactors led to elevated biomethane yields. Without antibiotics the system was more stable and it produced significant amounts of biogas for a longer period. The antibiotic-free substrate was used in the subsequent experiments.

The possibility of washing CM with water was tested next with the aim of decreasing the inhibitory nitrogen content of the substrate. After two days soaking CM in tap water, the liquid and solid phases were separated and significant nitrogen content was detected in the water phase. Removal of excess nitrogen-containing compounds improved the suitability of CM as sole substrate for anaerobic digestion. Biogas fermentation experiments were carried out with washed CM at mesophilic temperature (37 °C) in batch and continuous operational modes. The results demonstrated that anaerobic fermentation became sustainable when the reactors were fed with washed CM, which confirmed that ammonium concentration was indeed the limiting parameter in the anaerobic digestion of CM.

The simple and inexpensive method of removing nitrogen-rich water soluble components from CM, however, resulted in a large volume of water with high nitrogen and other dissolved organics content as a residual waste. In order to improve the economic value of the process, the separated liquid phase (CM-water) was used as a nutrient solution for algae because algae need a significant amount of nitrogen source for growth. A *Chlorella sp.* strain was cultivated in these experiments. The extreme dark color of the CM-water fraction did not permit its direct use for algae cultivation. In order to avoid light limitation CM-water was further diluted with water. At the optimal dilution ratio (CM-water : distilled water) the *Chlorella* culture grew vigorously and reached higher optical density than in its default growth medium. The harvested algal biomass could be recycled into the biogas generating process.

CM has low C/N ratio which is not favorable for evolving biogas with high methane content. To solve this problem we developed a co-fermentation scheme using the solid fraction of washed CM and corn stover, to increase this ratio. These experiments yielded promising results.

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Genes of the LATERAL ORGAN BOUNDARIES domain transcription factor family in *Brachypodium distachyon*

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The fascinating morphological plasticity of higher plants and formation of efficient shoot and root architecture for adaptation to continuously changing environment is ensured by their ability to establish new organs during their entire lifespan. This unique feature of plants is maintained by the reactivation of cell cycle in certain cells of differentiated tissues and by dynamic regulation of meristematic cell activity as well (Birnbaum et al. 2008; Bhojwani et al. 2013).

The precise coordination of division and differentiation of indetermined cells is essential in respect of proper and proportional organogenesis. Researches on plant development showed that special cells at boundaries between parental meristems and developing primordia play a dual role in this process by separating and maintaining meristem and organ domains. In accordance with their special function these cells express unique set of genes that reduce cell division and auxin efflux carrier activity but activate meristematic gene expression at the same time (Rast et al. 2008).

LOB-domain (LATERAL ORGAN BOUNDARIES DOMAIN) genes are among typical genes expressed prominently by these boundary-forming cells. They encode a family of plant specific transcription factors characterized by the 100 amino acid long conserved LOB-domain structure being responsible for DNA-binding and protein-protein interactions (Matsumura et al. 2009). Although their exact function is mostly unknown, observations up to now suggest that LOB-domain proteins are involved in almost all aspects of plant development from germination to seed production (Majer and Hochholdinger 2011). Their importance on organogenesis has been studied so far mostly in *Arabidopsis thaliana*, notwithstanding that revealing their role in monocots might be at least so relevant and interesting both for scientific and agronomical considerations, too. Therefore, we aimed to get to know in detail the processes controlled by LOB-domain protein coding genes (LBDs) in *Brachypodium distachyon*, a recently accepted and widely used model plant for cereals with high agronomical importance (Draper et al. 2001).

In current *Brachypodium* genome database we identified 28 LOB-domain protein coding genes. The encoded proteins are clustered into two major classes and some minor subclasses can be distinguished on the basis of their amino acid sequence homology. At first we characterized the relative expression levels of *Brachypodium*-LBDs by quantitative-real-time PCR in dozens of plant parts from root tip to shoot apex, both in vegetative and generative organs. According to our uniquely detailed analysis, LBD genes show variety of tissue specific expression: some of them are definitely active in flowers, in developing seeds and different parts of the floret, some of them have high activity in green plant parts while some others can be described as root specific genes, supporting the extremely diverse function of LBD gene family. Moreover, we got several expression patterns which have a good correlation to the transcriptional activity of their homologues from other species (e.g. *Arabidopsis thaliana* or *Oryza sativa*) that strongly suggests evolutionary conserved function of LBDs.

Aside from few exceptions, the closely related genes clustering into same subgroup showed overlapping expression pattern suggesting potential functional redundancy among them. However, one of the most interesting examples for exceptions are the Bd2g3450 and Bd2g53690 genes which have significantly divergent transcript profile from each other despite of their very close phylogenetic relationship (expression of Bd2g34520 is restricted to root tips while activity of Bd2g53690 is especially high in generative organs). For further exploration of possible functions we have selected these two LBD genes, with special regard on their presumable direct connection with the cell cycle regulating machinery.

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Birds of a feather - comparative gene expression analysis of Ada3 - a meta-analytic review with experimental flavors

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In eukaryotes, alterations of the chromatin structure have an important role in silencing and activation of particular genomic regions. The epigenetic processes are involved in changing the genes expression patterns, therefore the cell is capable of responding to different environmental impacts. Posttranslational modifications of histone proteins are crucial in structural changes of the chromatin. One of the most frequent covalent histone modifications is the acetylation of lysine side chains. The GNAT (GCN5-related N- acetyltransferase) complexes (which contain GCN5 protein as a catalytic component) have a common subunit called ADA3 (alteration/deficiency in activation 3). The homologues of this protein can be found all over in eukaryotes, from yeast to humans.

Lots of experimental data bits can be found which show that ADA3 protein is a lot more than a simple adaptor subunit: although it doesn't have any catalytic activity, it may have a sophisticated role in the function of the multiprotein machineries it is involved in. ADA3