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Review

Water hyacinth a potential source for value addition: An overview



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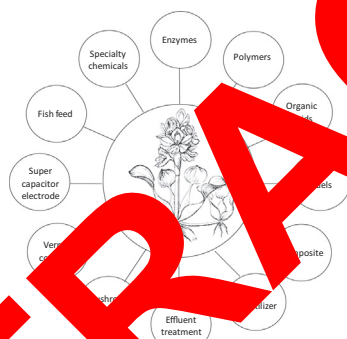
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HIGHLIGHTS

- Overview on the production of value added products from water hyacinth.
- Recent trends in water hyacinth based biorefinery.
- Strategies for renewable fuels from water hyacinth.
- Several possibilities for the generation of wealth from this weed.
- Targeting multiple products would improve economic viability of process.

GRAPHICAL ABSTRACT



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ABSTRACT

Water hyacinth a fresh water aquatic plant is considered as a noxious weed in many parts of the world since it grows very fast and depletes nutrients and oxygen from water bodies adversely affecting the growth of both plants and animals. Hence conversion of this problematic weed to value added chemicals and fuels helps in the self-sustainability especially for developing countries. The present review discusses the various value added products and fuels which can be produced from water hyacinth, the recent research and developmental activities on the bioconversion of water hyacinth for the production of fuels and value added products as well as its possibilities and challenges in commercialization.

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1. Introduction

Water hyacinth (WH) is a free floating, perennial aquatic plant originated from Amazon river basin and have distributed throughout the world. It has exhibited extremely high growth rate and the coverage of waterways by WH has created several problems, including destruction of eco systems, irrigation problems, also as a mosquito breeding place leading to increase in mosquito population (Sornvoraweat and Kongkiattakorn, 2011). It is considered as the most productive plant on earth and now considered as a serious threat to biodiversity. The invasive effects of WH lead to several research and developmental activities for the control of this notorious weed. Attempts to control this weed have high costs and labour requirements. Several biological, physical and chemical methods have been used for the control and eradication of WH but none of these strategies proved to be a permanent solution for the control of this weed.

WH contains about 20% of cellulose, 48% of hemicelluloses and 3.5% of lignin. The high hemicellulose and cellulose content of the WH can be exploited for the production of various value added products and fuels. Since the productivity is very high it could be utilized as feed stock for the production of biofuels. WH has several advantages like it can grow on water without competing against arable land for growing grains and vegetables. Several reports are available for the conversion of WH to fuel ethanol and biogas (Okewale et al., 2016; Gunja et al., 2016; Das et al., 2016; Shah et al., 2015). The commonly used strains for the production of bioethanol from WH are *Saccharomyces cerevisiae*, *Pichia stipitis* and *Zymomonas mobilis*.

Discharge of industrial effluent to the environment creates several ecological and environmental issues. Aquatic plants are well known for water purification as well as extraction of heavy metals and nutrients. Compared to other aquatic plants WH is the most suitable aquatic weed for phytoremediation. The potential of WH for the removal of pollutants is a well-established environmental protection technique and it functions like “nature's kidney” for

removal of toxic compounds from water resources of earth. The adsorption potential of WH has been exploited for the removal of various heavy metals and pigments from various industrial effluents. Growing of WH in industrial effluents leads to decrease of total suspended solids (TSD), chemical oxygen demand (COD) as well as biological oxygen demand (BOD). The mechanism involved in biosorption is by extracellular accumulation/precipitation, cell surface sorption/precipitation and intracellular accumulation (Rai et al., 2002). Phytoremediation using WH is cost effective and eco-friendly process.

The present review addresses the recent developments and advances for the production of fuels and value added products from the nuisance weed WH.

2. Current conversion strategies

Lignocellulosic biomasses are composed of cellulose, hemicelluloses and lignin as the major component. Pretreatment is to be carried out to facilitate the separation of hemicellulose, cellulose and lignin, so that complex carbohydrate molecule containing cellulose and hemicellulose can be broken down by enzymatic saccharification to simple sugars. The main objective of the pretreatment is to make the cellulose accessible for enzymatic saccharification by removing hemicelluloses and lignin. Several pretreatment strategies are available for the fractionation, solubilisation, hydrolysis and separation of cellulose, hemicellulose and lignin. This includes physical methods, chemical methods and hybrid strategies.

Several reports are available on the pretreatment of WH like acid (Satyanagalakshmi et al., 2011), alkali (Pothiraj et al., 2014; Aswathi et al., 2013), biological (Sinigani et al., 2005), hot water (Saha et al., 2014), microwave-alkali (Zhang et al., 2016), ultrasound combined alkali (Soontornchaiboon et al., 2016), catalytic hydrothermal liquefaction (Singh et al., 2015), calcium peroxide (Cheng et al., 2015), surfactant free ionic liquid microemulsions (Xu et al., 2016), thermo-chemical conversion (Huang et al.,

Table 1
Value added products from water hyacinth residue.

Product	Microorganism	Reference
Cellulose xanthogenate	–	Rodkong et al. (2016)
Cellulose xanthogenate	–	Deng et al. (2012)
Cellulose xanthogenate	–	Tan et al. (2008)
Levulinic acid	–	Girisuta et al. (2008)
Shikimic acid	–	Cardoso et al. (2014)
Biogas	Microbial consortium	Okevale et al. (2016)
Biogas	Microbial consortium	Shah et al. (2015)
Biogas	Microbial consortium	Ehiri et al. (2014)
Biogas	Microbial consortium	Putra et al. (2014)
Biogas	Microbial consortium	Shankar et al. (2014)
Biogas	Microbial consortium	Raja and Lee (2012)
Biogas	Microbial consortium	Singhal and Singh (2003)
Bioethanol	<i>Saccharomyces cerevisiae</i>	Gunja et al. (2016)
Bioethanol	<i>Clostridium thermocellum</i>	Das et al. (2016)
Bioethanol	<i>Zymomonas mobilis</i>	Singh et al. (2016)
Bioethanol	<i>Saccharomyces cerevisiae</i>	Ambo et al. (2016)
Bioethanol	<i>Pichia stipitis</i>	Bothiraj et al. (2014)
Bioethanol	–	Aswathi et al. (2013)
Bioethanol	<i>Pichia stipitis</i>	Sharma et al. (2013)
Bioethanol	<i>Candida shehatae</i>	Sharma et al. (2013)
Bioethanol	<i>Saccharomyces cerevisiae</i>	Ganguly et al. (2013)
Bioethanol	<i>Candida intermedia</i> NRRL Y-981	Mani et al. (2012)
Bioethanol	<i>Saccharomyces cerevisiae</i>	Satyana Lakshmi et al. (2011)
Bioethanol	<i>Candida tropicalis</i> TISTR 5045	Sornvoraweat and Kongkiattakajorn (2011)
Bioethanol	<i>Pichia stipitis</i>	Kumar et al. (2009)
Bioethanol	<i>Pichia stipitis</i> NRRL Y-7124	Agam (2002)
Biohydrogen	<i>Clostridium diolis</i> C32-KKU	Muanruksa et al. (2016)
Biohydrogen	–	Patra and Sittijunda (2015)
Biobutanol	<i>Clostridium beijerinckii</i>	Park et al. (2016)
Biopolymer	<i>Cupravidus necator</i>	Radhika and Murugesan (2012)
Biopolymer	<i>Pseudomonas aeruginosa</i>	Preethi and Umesh (2015)
Carbon fibre	–	Soenjaya et al. (2015)
Composite (polyethylene/WH)	–	Supri and Ismail (2010)
Biofertilizer	–	Vidya and Girish (2014)
Biofertilizer	–	Lata and Veenapani (2013)
Biofertilizer	–	Lata and Veenapani (2011)
Fish feed	<i>Bacillus subtilis</i> C-13	Saha and Ray (2011)
Substrate for mushroom cultivation	<i>Bacillus megaterium</i> C-13	Onchonga et al. (2013)
Substrate for mushroom cultivation	–	Chen et al. (2010)
High calorific value fuel	–	Lu et al. (2009)
Fuel briquette	–	Rezania et al. (2015)
Fuel briquette	–	Oroka and Thelma (2013)
Fuel briquette	–	Ighodalo et al. (2011)
Effluent treatment	–	Bathla (2016)
Effluent treatment	–	Maulion et al. (2015)
Effluent treatment	–	Elias et al. (2014)
Superabsorbent polymer	–	Pitaloka et al. (2013)
Xylitol	<i>Candida tropicalis</i> Y-27405	Kalhorinia et al. (2014)
Vermicompost	–	Gajalakshmi and Abbasi (2002)
Supercapacitor electrode	–	Kurniawan et al. (2015)

2016) and crowfoot assisted alkali-organosolvent (Das et al., 2016). Each pretreatment technology has its own merits and demerits. An ideal pretreatment strategy would remove hemicelluloses and lignin, reduce turbidity and would not generate any inhibitors of fermentation. Since the composition of biomass varies depending upon the species and variety, optimization of various process parameters affecting pretreatment to be carried out for getting better reducing sugar yield.

3. Value added products from water hyacinth

Several value added products can be produced from WH residue. This include different enzymes, cellulose xanthogenate, levulinic acid, shikimic acid, biogas, bioethanol, biohydrogen, biopolymer, biobutanol, composites, biofertilizers, fish feed, high calorific value fuel, fuel briquette, superabsorbent polymer and xylitol. In addition, WH can also be used as substrate for mushroom

cultivation and for treatment of various industrial effluents for the removal of heavy metals. Table 1 show the various value added products produced from WH.

3.1. Enzymes

Various enzymes are produced by the utilization of WH residue. This includes cellulase, β -glucosidase and xylanase. Table 2 presents different enzymes produced from WH residue.

3.1.1. Cellulase

Cellulases find potential applications in food, textiles and paper industry. Cellulase hydrolyzes cellulose into sugars which can be used for the production of bioethanol, chemicals and organic acids. Cost of carbon source is one of the significant factors affecting the cost of cellulase production. Hence utilization of low cost substrates like WH seems promising.

Table 2
Enzyme produced from water hyacinth residue.

Enzyme	Microorganism	Reference
Cellulase	<i>Aspergillus niger</i>	Pothiraj et al. (2016)
Cellulase	<i>Trichoderma viride</i>	Pothiraj et al. (2016)
Cellulase	<i>Aspergillus niger</i>	Amriani et al. (2016)
Cellulase	<i>Trichoderma reesei</i> SEMCC-3.217	Zhao et al. (2011)
Cellulase	<i>Trichoderma reesei</i>	Deshpande et al. (2009)
Cellulase	<i>Aspergillus niger</i>	Ali and El-Dein (2008)
	<i>Aspergillus nidulans</i>	
Cellulase	<i>Trichoderma reesei</i>	Mukhopadhyay and Nandi (1999)
β -glucosidase	<i>Rhizopus oryzae</i>	Karmakar and Ray (2011)
Xylanase	<i>Trichoderma reesei</i> NRRL 3652	Manivannan and Narendhirakannan (2014)
Xylanase	<i>Aspergillus flavus</i>	Saha et al. (2012)
Xylanase	<i>Trichoderma reesei</i> QM 9414	Deshpande et al. (2008)

Cellulase production in submerged fermentation using WH as carbon source was reported by Pothiraj et al. (2016) using *Aspergillus niger* and *Trichoderma viride*. The optimum conditions for cellulase production were substrate concentration of 5% (w/v) and incubation period of 72 h. *Trichoderma viride* showed an activity of 68.3 IU while *Aspergillus niger* exhibited an activity of 46.3 IU. Amriani et al. (2016) utilized WH as a substrate for cellulase production using *Aspergillus niger*. Maximum cellulase activity (1.035 IU) was observed after seven days of incubation, moisture content of 75–80%, initial pH of 5.0 and incubation temperature of 30 °C.

High yield of cellulase production in SSF by *Trichoderma reesei* SEMCC-3.217 using WH was evaluated by Zhao et al. (2011). The study revealed that addition of wheat bran, CaCl₂, tween 80 and (NH₄)₂SO₄ had significant effect on cellulase production. Statistical optimization improved the production by four fold (13.4 FPU/g). The potential of WH for cellulase production was reported by Sachin et al. (2011). The medium was enriched with WH as energy source and maximum cellulase activity was observed after 6 days of incubation, incubation temperature of 30 °C, pH 7.0 and shaking at 150 rpm.

Deshpande et al. (2009) utilized WH as sole carbon source for the production of cellulase by *Trichoderma reesei*. Various process parameters affecting cellulase production were optimized. Under optimized conditions 0.22 IU/ml of cellulase activity was observed. The study revealed that sugarification of cellulose was significantly higher by laboratory produced cellulase than commercial blends. WH is an abundantly available lignocellulosic biomass composed mainly of cellulose and hemicelluloses and can be utilized for the economic production of cellulase. The utilization of WH as a renewable resource will contribute to the solution of socioeconomic problems associated with this aquatic weed.

Ali and El-Dein (2008) reported production and partial purification of cellulase by *Aspergillus niger* and *Aspergillus nidulans* grown on WH and fortified with Czapeck-Dox medium in a 4:1 ratio. The study revealed that maximum cellulase activity was observed at an incubation temperature of 35 °C, pH 7.0, sodium nitrate as nitrogen source and incubation period of 7 and 3 days under static and shake flask conditions for *A. niger*. The optimized conditions for cellulase production by *A. nidulans* was an incubation temperature of 30 °C, pH 7.0, sodium nitrate as nitrogen source and 7 and 4 days of incubation under static and shake flask conditions.

Kurup et al. (2005) evaluated the potential of WH as a substrate for cellulase production by bacteria under solid state fermentation. Three native bacterial isolates WHB3, WHB4 and SMB3 were evaluated for cellulase production using WH. The results indicate that all the three strains produced cellulase using WH as solid support. Under optimized conditions strain WHB3, WHB4 and SMB3 produced 127.2, 110.4 and 94.8 U respectively. Addition of nitrogen

sources resulted in significant increase in cellulase yield. The study proves the potential of WH as substrate for the commercial production of cellulase.

Mukhopadhyay and Nandi (1999) reported cellulase production from *Trichoderma reesei* ATCC 27121 using a simplified medium on WH biomass. Various process parameters affecting production were optimized. The results concluded that cellulase can be produced from a cheap substrate WH supplemented with yeast extract, tween-80, KH₂PO₄, (NH₄)₂SO₄ and 4% w/v of milled WH as substrate.

3.1.2. β -Glucosidase

β -Glucosidase (EC 3.2.1.21) is the key enzyme component in cellulase. It completes the final step in cellulose hydrolysis by converting cellobiose to glucose. It is the rate limiting enzyme. Karmakar and Ray (2011) reported β -glucosidase production by *Rhizopus oryzae* by solid state fermentation (SSF) of WH. The production cost of β -glucosidase could be reduced by using WH as sole carbon source using SSF. Various process parameters affecting β -glucosidase production was optimized by adopting a central composite design. The maximum β -glucosidase activity of 137.32 U/ml was observed with a substrate concentration of 1.25%, pH of 6.6 and incubation temperature of 32.09 °C. The study revealed the potential of using WH as a substrate for cost effective production of β -glucosidase.

3.1.3. Xylanases

Xylanases (EC 3.2.1.8) are enzymes which degrade xylan randomly and produce xylooligosaccharides, xylobiose and xylose. Xylanases represents one of the largest groups of commercial enzymes. It finds applications in paper industries for bio-bleaching of paper pulp, as an additive in animal feedstock to improve the nutritive value, food additives in baking industry, as an ingredient in detergents or fabric care compositions as well as for biofuel production. Response surface strategy was adopted by Manivannan and Narendhirakannan (2014) for the co-production of cellulase and xylanase enzymes from *Trichoderma reesei* NRRL 3652. The study revealed that WH can be used as a cost effective substrate for the production of cellulases and xylanases which in turn ultimately helps to develop a cost effective process for bioethanol production. Under optimized conditions 21.47 IU/ml of xylanase was produced.

Submerged cultivation of *Aspergillus flavus* xym4 for the production of highly active and thermostable xylanase using WH as a substrate was reported by Saha et al. (2012). The xylanase production using WH as substrate is comparable to that using birch wood xylan. The optimized conditions for xylanase production were an incubation temperature of 30 °C, pH 6.5 and incubation period of 72 h. There was a two-fold increase in xylanase activity (3292 U/ml).

Deshpande et al. (2008) reported xylanase production by *Trichoderma reesei* (QM 9414) and *Aspergillus niger* by SSF using WH as a substrate. For enhanced production of xylanase a solid state cabinet fermenter (SSCF) was used. SSCF provide more space and large surface area to the substrate as well as better control of fermentation parameters like moisture, humidity, temperature and aeration. Maximum xylanase activity was observed on the seventh day. The study revealed that supplementation of additional nutrients with Toyoma Ogawa medium produced 8–9-fold increase in xylanase.

3.2. Cellulose xanthogenate

Cellulose xanthogenate showed increased capacity of heavy metal adsorption. For the preparation of cellulose xanthogenate, WH was first treated with alkali to obtain an alkali treated straw intermediate and when sulfonated with CS₂ and substituted by magnesium salt to produce magnesium cellulose xanthogenate which shows high adsorption capacities for heavy metals. Rodkong et al. (2016) reported the potential of WH for the preparation of natural fibre sorbent for oil sorption. The highest efficiency of the sorbent was observed when prepared with 18% NaOH, 25% CS₂ and sodium sulphate in a 1:1.5 ratio. Compared to conventional sorbent polypropylene, the oil sorption properties of WH sorbent was 1.23 times higher in engine oil, 1.15 times in vegetable oil and 1.43 times in diesel oil. Thus the utilization of WH as a natural fibre sorbent will be economically and ecologically viable.

Deng et al. (2012) adopted different strategies for preparation of cellulose xanthogenate using bio-degumming, CS₂ sulfonation and magnesium substitution and the study revealed that based on an environmental aspect, pectate degumming is the most promising method for preparation of heavy metal adsorbent. Wang et al. (2008) selected cellulose from WH and other plant materials based on the exchangeable capacity of copper and carbon content. The results indicate that the adsorption capacity of cellulose xanthogenate of WH to copper was higher than other plant materials and the adsorption capacity increases with increase of pH value. The study revealed that cellulose xanthogenate made from WH can serve as a potential source for the treatment of waste water polluted by copper.

3.3. Organic acids

3.3.1. Levulinic acid

Levulinic acid (LA) or 4-oxopentanoic acid is a keto acid. It is derived from agricultural residue cellulose and is a potential precursor to biofuels. LA is a building block for the synthesis of various organic compounds. γ -Aminolevulinic acid is used as a biodegradable pesticide. LA reacts with phenol to produce diphenolic acid which can be used for the production of epoxy resins, polycarbonates and other biopolymers. Esters of LA are used in flavouring and fragrance industry as well as for blending with biodiesel. Girisuta et al. (2008) developed an acid catalyzed strategy for the production of LA from WH. The various process parameters affecting LA production like temperature, sulphuric acid concentration and WH intake were optimized. The study revealed that at high sulphuric acid concentration, LA was the major organic acid produced while at low sulphuric acid concentration propionic acid is preferentially formed. Organic acid production is based on the reaction conditions. The highest yield of LA was 53 mol% based on the amount of C6 sugars in WH. This low yield is due to the relatively low amount of C6 sugars in WH leaves.

3.3.2. Shikimic acid

Shikimic acid is a naturally occurring organic compound and its anionic form shikimate is an important intermediate in the synthesis of aromatic amino acids like tyrosine, tryptophan and phenylalanine. It is a high value compound used as a key starting material for the synthesis of neuramidase inhibitor GS4104 which is used for the treatment of antiviral infections. It also plays an important role as anti-coagulant, antioxidant, antibacterial, anti-inflammatory and analgesic activities. It is also used as an additive to food and feed and injectable. It is mainly obtained from Chinese star anise and from genetically modified *E. coli*. Cardoso et al. (2014) reported shikimic acid production from WH thereby keeping it under control. The study revealed the potential of WH as an alternative renewable source of shikimic acid. HPLC analysis of the plant extracts with methanol revealed that the aerial parts of WH contains higher shikimic acid concentration (0.05–2.7% w/w) when compared to roots where the concentration was 0.05–0.09% w/w. Soxhlet extraction was found to be more efficient than ultrasonic bath and magnetic stirring and temperature does not significantly affect the extraction process.

Lenora et al. (2016) evaluated the potential of WH for the production of shikimic acid, a precursor of Tamiflu, a swine flu drug. Shikimic acid is the key compound in the manufacture of this drug and one of the main bottlenecks in the production of Tamiflu is the availability of shikimic acid. The only source for extraction of shikimic acid is from the fruits of Chinese star anise. The study revealed that n-hexane extract of WH leaves contain more shikimic acid (2.5%) than Chinese star anise (1.77%). Hence WH serves as an alternative renewable source for shikimic acid. Utilization of WH for the production of shikimic acid could be alternative strategy for the management of WH contributing to ecological and environmental problems caused by it.

3.4. Biogas

Biogas is a gas mixture produced by the anaerobic fermentation of organic materials by methanogenic bacteria. It consists of a mixture of methane, carbon dioxide, water, hydrogen sulphide and ammonia. WH serves as a potential source for biogas production. Several reports were available for the potential of WH as a raw material for the production of biogas. Anaerobic co-digestion of WH along with cow dung and elephant grass for biogas production at laboratory scale was evaluated by Okewale et al. (2016). The study revealed that co-digestion of WH and elephant grass gave a higher yield of biogas production. Process parameters like temperature, pH and retention time have a significant effect on biogas yield. The highest methane content of 62% was observed in digester 1 which contains WH, elephant grass, cow dung and water gave a yield of 2.303 L after 60 days of incubation.

Anaerobic digestion of WH and other two plants like giant reed and maize were explored for their potential for biogas production by Shah et al. (2015). WH had the highest biogas generation rate of 1000 ml/day followed by giant reed and poultry wastes. This is due to better C: N ratio and biochemical composition, WH was a successful substrate for mono-digestion which resulted in its highest biogas production rate. The cumulative biogas production during 30 days was highest for WH (25780 mL), followed by giant reed (18845 mL) and maize (15900 mL). The study revealed that utilization of WH as substrates for biogas production will overcome energy crisis in developing countries to a certain extent.

Ehiri et al. (2014) reported the possibility of producing biogas from a mixture of WH and fresh rumen residue. The effects of reaction rate and kinetics on biogas production were evaluated. The study revealed the potential of WH for biogas production in Nigeria. The maximum biogas production was observed on

seventeenth day (16.4 ml). The reaction kinetics followed a second order with a specific rate constant of 0.02878 ml/day.

The effects of hydrothermal pretreatment on biogas production enhancement rates from WH mixed with buffalo dung were reported by Putra et al. (2014). Maximum biogas production (7889 ml/day) was observed when hydrothermal pretreatment was carried for 60 min with a WH: buffalo dung ratio of 1:2. The optimum methane yield was 2856 ml/day. The study revealed that the ratio of WH to buffalo dung has a significant impact on biogas production rates.

Shankar et al. (2013) reported effect of substrate concentration on biomethanation of WH. In this study the biomethanation was carried out for 60 days using a substrate concentration of 3–11% at mesophilic condition. WH itself does not have the ability to produce biogas. Anaerobic co-digestion of WH with primary sludge, cow dung and poultry litter were evaluated. Maximum biogas production was observed with a biomass loading of 7%. In the present scenario, WH proves to be a promising renewable source of energy in the form of biogas.

The study conducted by Raja and Lee (2012) revealed the possibility to produce biogas from a mixture of WH and cow dung. The results indicate that dried and chopped WH combined with cow dung had the highest cumulative biogas yield (64%) when compared with dried and chopped WH combined with wood charcoal (60%). WH is a very good biogas producer which needs minimal pre-treatment to enhance the biogas yield. Biogas can be used as a substitute for charcoal, firewood and oil products. The compost, obtained at the end of the digestion process, is an organic fertilizer.

Singhal and Rai (2003) reported biogas production from WH. The plant grows well in diluted paper mill and highly acidic tannery effluents and takes up heavy metals and other toxic materials for their growth. Utilization of the slurry of WH used for phytoremediation produced significantly more biogas than that of plants grown in deionized water. Maximum biogas production was observed in 9–12 days.

3.5. Bioethanol

Depletion of fossil fuels and increase in energy consumption leads to search for alternative sources of energy. Utilization of lignocellulosic feed stock can be considered as a suitable biomass for production of renewable biofuels like ethanol. Production of fuels from waste biomass like WH plays an important contribution in self-sustaining society. Bioethanol production from WH involves three stages: pretreatment, hydrolysis and fermentation. Bioethanol is a renewable fuel and its importance increases due to depletion of fossil fuels, increase of crude oil price and green house effect.

Gurukul et al. (2014) observed bioethanol production from WH using a thermotolerant yeast strain isolated from a sugarcane field. The novel isolate produced 13.45 g/l of bioethanol at an incubation temperature of 45 °C. Normally fermentation is carried out with *Saccharomyces cerevisiae*, but during summer the temperature raises up to 45 °C or above at which the yeast cannot survive and it is difficult for beverages industries to maintain the temperature at 32–35 °C for maintaining the viability of yeast. Hence the use of thermo-tolerant bacteria seems promising.

Enhanced bioethanol production by statistical optimization of WH was evaluated by Das et al. (2016) adopting a Taguchi design. Various process parameters affecting simultaneous saccharification and fermentation involving *Clostridium thermocellum* hydrolytic enzymes and fermentative microbes for enhanced bioethanol production from mixed, microwave-assisted alkali and organosolvent pretreated substrate were carried out. Under optimized conditions 9.78 g/l and 13.7 g/l of bioethanol was observed under shake flask and bioreactor. This is the first report utilizing

recombinant *C. thermocellum* enzymes for simultaneous saccharification and fermentation with subsequent Taguchi optimization.

Bioethanol production from fresh and dry WH using ruminant microorganisms and ethanol producers were evaluated by Sambo et al. (2015). The study revealed the potential of bacterial and fungal isolates obtained from rumen of goat, ram and cow to digest cellulosic materials of WH. Fermentation of the WH hydrolysate was carried out with *Saccharomyces cerevisiae* and *Zymomonas mobilis*. The results indicate that *Zymomonas mobilis* produced more bioethanol than *Saccharomyces cerevisiae* as well as the fresh WH biomass produced more bioethanol than the dried WH biomass. The utilization of WH for the production of bioethanol will go a long way in reducing dependence on fossil fuel.

Pothiraj et al. (2014) observed potential of bioethanol production from lime pretreated WH by using mono and co-cultures of isolated fungal strains like *Trichoderma reesei* and *Fusarium oxysporum* along with *Pichia stipitis*. The pretreated biomass was saccharified with crude enzymes from *Trichoderma reesei* and *Fusarium oxysporum* and the hydrolysate was fermented by *Pichia stipitis*. In simultaneous saccharification and fermentation, the co-culture fermentation using *Trichoderma reesei* and *Pichia stipitis* was found to be promising with a higher bioethanol yield of 0.411 g/g after 60 h of fermentation. Separate hydrolysis and fermentation resulted in lesser bioethanol yield (0.34 g/g) after 96 h of fermentation. The study revealed that higher bioethanol production was achieved in a shorter period in the co-culture system containing *Trichoderma reesei* and the xylose fermenting yeast *Pichia stipitis*. The optimum parameters for fermentation are biomass loading of 100 g/l, incubation temperature of 35 °C and incubation time for 60 h. The use of crude fungal enzymes produced on site proved to be a cost effective strategy for enzymatic saccharification of alkali pretreated WH biomass instead of using commercial cellulases.

Improved production of bioethanol from WH by optimization of pretreatment conditions was reported by Aswathi et al. (2013). The study revealed integration of low cost pretreatments with advanced bioethanol producing microorganisms will make the process commercially viable. Among the various acid and alkali used for pretreatment, H₂SO₄ was found to be the best pretreatment agent in terms of reducing sugar yield.

Ganguly et al. (2013) reported bioethanol production from WH hydrolysate using three different strains – *Pichia stipitis*, *Candida shehatae* and *Saccharomyces cerevisiae*. WH biomass was pretreated with acid and alkali and the alkali pretreatment was found to be more effective in lignin removal thereby enhancing the cellulose and hemicellulose content of WH by removing lignin. It was observed that enzymatic saccharification was more effective than saccharification with whole cell biocatalysts. The production of bioethanol can be enhanced by using both hexose and pentose utilizing strains simultaneously. Maximum bioethanol yield was observed with *Pichia stipitis* (3.49 g/l), followed by *Candida shehatae* (3.45 g/l) and 3.13 g/l for *Saccharomyces cerevisiae*.

The study conducted by Manivannan et al. (2012) revealed that bioconversion of WH to bioethanol using two sequential steps of acid hydrolysis followed by fermentation with *Candida intermedia* NRRL Y-981 produced maximum bioethanol yield of 0.21 g/g with a productivity of 0.01 g/l/h. The yield can be improved by integration of low cost pretreatments followed by fermentation with improved bioethanol producing microorganisms will play a critical role in making the process economically viable.

Satyanagalakshmi et al. (2011) evaluated bioethanol production from acid pretreated WH by separate hydrolysis and fermentation. Fermentation of the enzymatically saccharified biomass hydrolysate with *Saccharomyces cerevisiae* yielded 0.292% v/v of bioethanol. Separate hydrolysis and fermentation of WH leaves for fermentation has been evaluated by Sornvoraweat and

Kongkiattikajorn (2011). Dilute H_2SO_4 pretreated leaves were used for hydrolysis using an enzyme cocktail consisting of cellulases, xylanases and pectinases. Study revealed that fermentation using co-culture of *Saccharomyces cerevisiae* TISTR5048 and *Candida tropicalis* TISTR5045 showed highest bioethanol concentration (3.42 g/l) and yield (99.9%).

Bioconversion of lignocellulosic fraction of WH hemicellulosic hydrolyzate to bioethanol by *Pichia stipitis* was reported by Kumar et al. (2009). Fermentation of detoxified hydrolyzate yielded 0.425 g/g of bioethanol. Nigam (2002) evaluated the potential of WH hemicellulosic acid hydrolyzate to motor fuel bioethanol by xylose fermenting yeast, *Pichia stipitis* NRRL Y-7124. Fermentability of the hydrolyzate was considerably improved by boiling and over-liming up to pH 10.0. The optimum conditions of fermentation were pH of 6.0, incubation temperature of 30 °C and an aeration rate of 0.2vvm. The bioethanol yields of treated and untreated hydrolyzate were 0.19 g/g and 0.35 g/g respectively. The acetic acid present in the hydrolyzate decreased the bioethanol yield and productivity.

3.6. Biohydrogen

Due to depletion of fossil fuels and increase in energy demand scientists all over the world are searching for alternative strategies of energy. Biohydrogen is an alternative energy since its combustion generates only water and heat as well as has high energy yield of 122 kJ/g. Traditionally hydrogen is produced by chemical process involving electrolysis of water and steam reforming. These processes are not economically viable since it requires high energy input and high reaction temperature. Biohydrogen production is eco-friendly and can be produced from mixed or pure culture. Many anaerobic microorganisms can produce biohydrogen from organic wastes. *Clostridia* species produce hydrogen gas by exponential growth phase *Clostridia* produce hydrogen by reversible reduction of protons accumulated during fermentation to hydrogen and is catalyzed by the enzyme hydrogenase. *Clostridium* species are highly sensitive to oxygen and their hydrogenase activities will be inhibited by even traces of oxygen. The addition of a reducing agent like L-cysteine in the medium is responsible for biohydrogen production.

Muanruksa et al. (2016) reported direct biohydrogen production from WH using *Clostridium diolis* C32. Cellulose and hemicelluloses presented in WH is directly fermented by cellulolytic bacterium *Clostridium diolis* 2-KKU to biohydrogen. Various process parameters affecting biohydrogen production were optimized for both static and shaking modes of cultivation. The study revealed that shaking mode was more effective than static mode for biohydrogen production. Maximum biohydrogen production (19 mL) was observed at pH 6.5 and WH biomass loading of 19 gdw/l. The results clearly indicate that direct biohydrogen production from WH could be a feasible approach.

Pattra and Sittirakul (2015) optimized various process parameters affecting acid hydrolysis of WH for the production of biohydrogen. Response surface methodology (RSM) and central composite design (CCD) were adopted for the optimization studies. The optimum conditions for acid hydrolysis of WH were observed as reaction time of 7.73 h, dilute H_2SO_4 concentration of 1.31% (v/v) and stirring speed of 264.41 rpm yielded maximum total reducing sugar of 13 g/l. Fermentation of the hydrolyzate yielded 127.6 mM H_2 .

3.7. Biobutanol

Acetone-butanol-ethanol (ABE) fermentation was an important industrial process and was first reported for butanol production by Louis Pasteur in 1869. Increase in crude oil prices, depletion of fos-

sil fuels and increasing concerns of environmental issues has increased the demand for fermentative production of butanol. To tide over the limitations of conventional ABE fermentation several research and developmental activities were going on throughout the world for utilization of renewable and low cost feed stocks, metabolic engineering of solventogenic microbes as well as development of novel fermentation and downstream processing strategies.

Biobutanol have more merits than bioethanol in terms of chemical and physical properties. It could replace gasoline for transportation and its demand is increasing drastically. Park et al. (2016) reported biobutanol production from WH using *Clostridium beijerinckii*. The pretreated and enzymatically saccharified hydrolyzate of WH serves as an excellent medium for biobutanol production by *Clostridium beijerinckii*.

3.8. Biopolymer

Poly-3-hydroxybutyrate (PHB) is a polyhydroxyalkanoate which is one of the most important biodegradable plastics. It is produced by various microorganisms like *Bacillus*, *Pseudomonas*, *Ralstonia*, *Methanobacterium* etc and is produced when the nutrients are limited. PHB is a primary product of carbon assimilation and produced as energy storage molecule and assimilated when other carbon sources are not available. Biosynthesis of PHB takes place by the condensation of two molecules of acetyl CoA to give acetoacetyl CoA which is reduced to hydroxybutyryl CoA and later polymerize to form PHB. Production cost of PHB is very high when compared to commercially available polyesters. More than 70% of the total production cost is contributed by the carbon

source. Patra and Murugesan (2012) reported PHB production from WH hydrolysate of WH as sole carbon source. Acid pretreated and enzymatically saccharified WH hydrolyzate was used for PHB production by *Cupravidus necator*. The study revealed that addition of WH enzymatic hydrolyzate in the minimal mineral media gave higher PHB production (4.3 g/l). Fermenter studies gave a higher yield (7 g/l).

Polyhydroxyalkanoate (PHA) production from *Pseudomonas aeruginosa* using WH as a potential substrate was reported by Preethi and Umesh (2015). Acid pretreatment was carried out for breaking down of complex sugars in the WH to easily fermentable sugars. WH hydrolysate supplemented with glucose, peptone, yeast extract and NaCl was used for PHA production by *Pseudomonas aeruginosa*. Extraction of PHA from the fermentation media yielded 65.51% of PHA after 72 h of incubation.

3.9. Carbon fibre

Carbon fibre has been used as a precursor for the preparation of composite materials. Most of commercially available carbon fibres are prepared from organic polymers like rayon, polyacrylonitrile or petroleum pitches. The major drawback of commercial carbon fibres is the high cost of the precursors. This can be overcome by using renewable polymers as precursors for carbon fibre production (Zheng et al., 2014).

Soenjaya et al. (2015) prepared carbon fibre from WH liquid tar. Chemical compositional analysis of WH liquid tar revealed that it contains significant amount of phenolic compounds. High content of phenolic compounds indicates that the WH tar is a suitable material for carbon fibre production. The carbonization of WH was carried out at 900 °C yielded 29% of carbon fibre. Characterization by SEM, XRD and FTIR revealed that the properties are comparable to commercial carbon fibre. The carbon fibre obtained from WH is non-graphite in nature (Soenjaya et al., 2015).

3.10. Composite

Natural fibres are reinforced with polymer composites for the production of low cost materials of engineering. The natural fibres present in WH can be used as reinforcement materials in polymer composites due to their interesting characteristics like high cellulose content and low cellulose diameter. [Supri and Ismail \(2010\)](#) developed a low density polyethylene/ WH composite by melt blending. The study revealed that polyethylene/WH composite prepared by coupling with NCO-polyol showed higher values of tensile strength and water absorption resistance. The modified fibre exhibited better thermal stability than unmodified WH fibre.

[Ramirez et al. \(2015\)](#) reported composite production from WH and polyester resin. Composite production from cheap natural fibre of WH is economically viable. Composites of WH and polyester resin were prepared using solution impregnation and hot curving methods. Blending improved the thermal and mechanical properties of the composite. The composites which contain 5 to 10% of WH showed better properties. The integration of WH fibre into polyester resin generated composite of low molecular weight with superior acoustic insulation when compared to polyester resin. The study revealed WH can replace conventional materials such as glass, carbon and plastic fibres.

3.11. Biofertilizers

Biofertilizers are organic material of natural origin and which provides one or more nutrients to plants essential for their growth. One of the mostly available strategies for soil fertility remediation is the use of weeds. The study conducted by [Vidya and \(2014\)](#) revealed that WH can be used as a biofertilizer when incorporating to soil increased the performance of wheat plant. In this study wheat crop was treated with compost derived from WH and were grown for 15 days. Control experiment were carried out without WH compost. Physical and chemical parameters were studied. The physical parameters like percentage germination, length of root, length of shoot, biomass content and C/N ratios were studied. Chemical parameters like chlorophyll, reducing sugar and protein content were also evaluated. The study revealed that both physical and chemical parameters had higher values as compared to control. WH is a good absorber of N, P and K from the water and can be used as a compost material. The results indicate the potential of WH as organic manure.

Response of WH manure on yield and growth attributes in *Coriandrum sativum* was reported by [Lata and Veenapani \(2013\)](#). Addition of organic manure along with WH manure in various combinations resulted to have a positive impact on performance of crop plants as a result of increase in nutrient availability. The growth and productivity of *Coriandrum sativum* was more pronounced with 100% WH manure treatment.

Response of WH manure on yields and growth attributes in *Brassica juncea* was reported by [Lata and Veenapani \(2011\)](#). The study revealed that addition of WH manure in various combinations into soil have a pronounced influence in the performance of crop plants as a result of increased nutrient availability when compared to control. The growth of *Brassica juncea* were more pronounced with addition of 50% of WH manure and WH manure combined with farm yard manure on the growth behaviour of seedlings when compared to control.

3.12. Fish feed/animal feed

Several studies are going on for the development of supplementary feed for cost effective substitution of high cost fish meal with cost effective protein source. Aquatic macrophytes have been used as supplementary feed in fish farming. WH can be used as a source

for fish feed. Nutritional value of WH leaves fermented with *Bacillus subtilis* CY5 and *Bacillus megaterium* CI3 were reported by [Saha and Ray \(2011\)](#). The bacterial strains having cellulolytic and amylolytic activity were used for fermentation of WH leaves for 15 days at 37 °C. The study revealed that fermentation of the WH leaves resulted in reduction of crude fibre, cellulose and hemicellulose contents, anti-nutritional factors, tannin and phytic acid. The free amino acids and fatty acids were increased in the fermented WH leaf meal. Both the inclusion level and type of WH leaf meal significantly affected the growth performance of Rohu (*Labio rohita*). The study revealed that WH leaf meal fermented with fish gut bacteria extracellular enzyme activity as a dietary ingredient in diets of *Labio rohita* fingerlings up to 40% and replacing fish meal without any adverse effects on growth of the fish to produce cost effective fish feed. [Mohapatra \(2015\)](#) reported utilization of WH meal as partial fish protein replacement in the diet of *Cyprinus carpio* fry. WH at different levels (0%, 10% and 30%) was prepared to feed *Cyprinus carpio* in place of fish meal. The study revealed that growth performance decreases as the level of WH increases. WH based weeder is cheaper as compared to the conventional feed. Supplemental use of WH in carp diets would make the process economically viable. [Sam \(2015\)](#) reported utilization of WH as animal feed. High protein content in the leaves and rapid growth has made WH potential for use as fodder for cows, goats, pigs etc.

3.13. Mushroom cultivation

Mushroom cultivation is normally carried out in crop residues, paddy straw, waste cotton, sugarcane bagasse, maize stalks and other crop stalks. Availability of most of these crop residues was not uniform throughout the year. WH serves as a potential raw material for mushroom cultivation. [Onchonga et al. \(2013\)](#) reported utilization of WH as an alternative substrate for mushroom farming. The study revealed that the yield of mushrooms increased substantially when WH was combined with saw dust than when WH used alone (control). [Murugesan et al. \(1995\)](#) developed a strategy for oyster mushroom cultivation using WH. The study revealed that WH serves as a good substrate for mushroom cultivation and a good yield was achieved due to ideal C/N ratio and low lignin content. Since WH is available free of cost, this technology is economically viable and also helps in the eradication of this troublesome aquatic weed. [Chen et al. \(2010\)](#) reported mushroom cultivation using biogas fluid soaked WH and saw dust. The study revealed that among the different strategies adopted the greatest yield and highest amino acid content was obtained when the proportions of WH and sawdust in the medium were equal.

3.14. High calorific fuel

[Lu et al. \(2009\)](#) prepared high calorific value (HCF) from WH by deoxy-liquefaction. The maximum yield of HCF was 12.6 w% and the dominant components were alkanes, benzene derivatives and phenol derivatives. Elemental analysis data reveal that the residual content of hydrogen was too low to produce HCF and deoxy-liquefaction was reported as an effective way to remove oxygen and to utilize carbon and nitrogen in WH more effectively. The empirical formula of HCF from deoxy-liquefaction was CH_{1.7000}O_{0.38N0.026}. The optimum temperature for deoxy-liquefaction was observed as 623 K. The study revealed that temperature played an important role on the product distribution of HCF. With increase of temperature, the yields of alkanes and benzene derivatives increased. This strategy shows the potential of WH as an energy supplement.

3.15. Fuel briquette

Increase in energy demand has raised concerns about the economic and environmental impacts of power generation based on each nation's energy source. Briquetting of abundant biomass is one of the possible solutions to overcome the local energy shortages in the country. WH will be an ideal source for preparation of fuel briquettes. Compared to wood or other fuel briquettes WH has a lesser cultivation and preparation cost. The possibilities of WH conversion to briquettes have been reported by [Rezania et al. \(2015\)](#). [Oroka and Thelma \(2013\)](#) investigated the properties of fuel briquette obtained by mixing WH and cow dung. Mixing of WH: cow dung was carried out in four different ratios such as 100:0, 90:10, 80:20 and 70:30. The results indicate that the briquettes produced with 80:20 and 70:30 WH: cow dung ratio exhibited largest relaxed density on drying with values of 1296 kg/m³ and 1157 kg/m³ respectively. The durability of the briquettes exceeded 85%.

[Ighodalo et al. \(2011\)](#) processed WH biomass into briquette and observed that WH along with cassava starch as binder can be used as wood burner for cooking purposes. Addition of some binders can improve the properties of briquettes. These studies reveal that WH based fuel briquettes serves as a potential alternative to fire wood and charcoal.

3.16. Effluent treatment

The discharge of effluents from various industries adversely affects soil fertility, water resources and integrity of ecosystems. Phytoremediation using aquatic macrophytes seems as a promising strategy for the removal of pollutants and contaminants from various natural sources. WH has been widely used for the rapid removal of various kinds of pollutants from water due to its easy availability, effectiveness as well as its capability to remove a wide range of pollutants. Phytoremediation of metals and dyes in distillery effluent using WH was observed by [Bath et al. \(2016\)](#). The study revealed that heavy metals like iron, zinc, sodium, calcium, magnesium and calcium in the effluent were reduced after treatment with WH. Maximum reduction was observed with 2% WH concentration after 15 days. WH could be economically used to remediate distillery effluent since it is widely available and has high metal absorption capacities. Phytoremediation of WH could be used as an economically viable strategy for the treatment of distillery effluent.

[Maulion et al. \(2015\)](#) investigated the removal of hexavalent chromium in simulated wastewater using WH. Current technologies available for hexavalent chromium removal from waste waters are too costly and difficult to operate efficiently. The study revealed that WH serves as an excellent source for hexavalent chromium removal. Maximum chromium removal was observed at twelfth day. This indicates the potential of WH for phytoremediation of hexavalent chromium contaminated waste water.

[Elias et al. \(2014\)](#) reported the potential of WH for the bioremediation of ceramic industry waste water. Ceramic industry produces large amount of waste water which contains heavy metals like cadmium, chromium, copper, zinc, iron and boron. The study revealed the potential of WH in removing these heavy metals and they are translocated in roots, leaves and shoots of WH and the concentration of heavy metals is ten times higher in roots than in leaves and shoots of WH. In this study WH was able to remove 70% of heavy metals from waste water.

3.17. Superabsorbent polymer

WH for superabsorbent polymer material synthesis was reported by [Pitaloka et al. \(2013\)](#). One of the organic materials used

for superabsorbent is carboxymethyl cellulose (CMC). In this study CMC was produced from WH with a maximum degree of substitution (DS) of 0.72 with isopropyl alcohol as solvent. Synthesis of superabsorbent material from CMC results in highest absorption capacity. WH based CMC with a higher degree of substitution are expected to be more valuable. Preparation of superabsorbent material from CMC can be conducted by using citric acid as a cross linking agent which is environmentally friendly.

3.18. Xylitol

Xylitol is used as sugar substitute in many foods. It gained importance in food and pharmaceutical industries for the preparation of confectionaries, chewing gum, etc. It also prevents ear and upper respiratory tract infections and benefits pregnant and nursing women ([Prakasham et al., 2009](#)). [Chorini et al. \(2014\)](#) evaluated the potential of WH for xylitol production using *Candida tropicalis* Y-27405. Dilution and hydrolysis of WH was carried out with 2% H₂SO₄. The hydrolysate was concentrated using a rota vapour and detoxified by overliming followed by activated charcoal treatment. The sugars were measured using *Candida tropicalis* Y-27405 for xylitol production. 3.0 g/l of xylitol was obtained after 48 h of fermentation with a yield and productivity of 0.65 g/g and 0.67 g/l/h respectively.

3.19. Vermicompost

Vermicompost is produced by composting vegetables and food waste using various worms like earth worms, white worms and red wigglers. Vermicompost acts as an excellent nutrient rich organic fertilizer and soil conditioner. Vermicomposting seems to be a highly preferred option for large scale utilization of WH as well as for its ultimate disposal. [Gajalakshmi and Abbasi \(2002\)](#) reported the effect of application of WH compost/vermicompost on the growth and flowering of *Crossandra undulataefolia*. The study revealed that *Crossandra* saplings grown on vermicompost showed significant improvement in the growth and flowering of *Crossandra* when compared to the untreated plants.

[Patil et al. \(2012\)](#) reported vermicomposting of WH with poultry litter using rotary drum reactor. High organic content of WH makes it a potential source for vermicompost production. The study revealed the potential of converting WH to vermicompost with poultry litter as supplement. The vermicompost obtained after 45 days contain 30% moisture, 9.67% carbon, pH 7.2 and NPK values were 0.72%, 0.51% and 0.60% respectively. The main advantages of this strategy is that the vermicompost can be produced in 45 days which is half the time consumed by conventional strategies like wedge, bed and bin method as well as predigestion of WH has resulted in exemption of thermophilic stage during vermicomposting process thereby providing suitable environment for the persistence of earth worms.

[Patidar et al. \(2013\)](#) reported vermicompost production using WH by thermophilic composting using *Streptomyces viridosporus*, *Aspergillus niger* and *Moraxella osloensis*. Vermicomposting of jatropha seed cake with 2:1 ratio of WH and cow dung was tested. The study revealed that concentration of vermicompost above 20% had inhibitory effect and cause phytotoxicity.

[Blessy and Prabha \(2014\)](#) evaluated application of WH vermicompost on the growth of *Capsicum annum*. Two types of vermicompost were prepared – V1 and V2. One composted by using *Eudrilus euginae* and the other was prepared with cellulose from WH was hydrolyzed enzymatically and then composted by using *Eudrilus euginae*. The study revealed the potential of *Eudrilus euginae* in the degradation of WH leaves and to convert waste into a vermicompost. Macronutrients like potassium, phosphorous and nitrogen were higher in V2 when compared to V1. Micronutrients

copper and iron is higher in V2. The study revealed that V2 method is more efficient compared to V1 method when *Capsicum annuum* was treated with vermicompost of WH.

3.20. Supercapacitor electrode

Activated carbon has been widely used for the production of electrodes for supercapacitors. Pore structure and surface chemistry of the activated carbon plays an important role on the electrochemical and capacitance performance of the carbon electrode and it depends on the method of preparation and type of precursors. Kurniawan et al. (2015) exploited the potential of low cost biomass WH as the precursor for the preparation of carbon microspheres. The WH was hydrolyzed to sugars by dilute H₂SO₄ under subcritical water conditions for 2 h. Then the sugar solution was carbonized under subcritical conditions to produce carbon microspheres. The highest yield of carbon microspheres was 0.1019 g/g dry WH. Chemical and physical treatments of carbon microspheres were carried out using KOH solution and microwave to increase the specific surface area and porosity of carbon microspheres. Electro-capacitive studies of carbon microspheres revealed that the carbon microspheres activated at impregnation ratio of 1:1 and microwave power of 630 W showed the highest specific capacitance and excellent electrochemical stability.

4. Conclusion

The utilization of WH for the production of fuels and value added products will contribute significantly for the reduction of socio-economic problems associated with extensive growth of proliferative weed. Development of improved strains by genetic engineering for the production of value added products, process integration as well as media engineering for the improvement of product yield would develop an economically sustainable strategy, thereby making the process commercially viable. Several research and developmental activities are going on in this direction throughout the world for the conversion of WH to value added products leading to a sustainable management of this noxious weed.

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