Cocoa and Coffee Fermentations

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Biotechnological potential of coffee pulp and coffee husk for bioprocesses

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Abstract

Advances in industrial biotechnology offer potential opportunities for economic utilization of agro-industrial residues such as coffee pulp and coffee husk. Coffee pulp or husk is a fibrous mucilagenous material (sub-product) obtained during the processing of coffee cherries by wet or dry process, respectively. Coffee pulp/husk contains some amount of caffeine and tannins, which makes it toxic in nature, resulting the disposal problem. However, it is rich in organic nature, which makes it an ideal substrate for microbial processes for the production of value-added products. Several solutions and alternative uses of the coffee pulp and husk have been attempted. These include as fertilizers, livestock feed, compost, etc. However, these applications utilize only a fraction of available quantity and are not technically very efficient. Attempts have been made to detoxify it for improved application as feed, and to produce several products such as enzymes, organic acids, flavour and aroma compounds, and mushrooms, etc. from coffee pulp/husk. Solid state fermentation has been mostly employed for bioconversion processes. Factorial design experiments offer useful information for the process optimization. This paper reviews the developments on processes and products developed for the value-addition of coffee pulp/husk through the biotechnological means.

Keywords: Coffee pulp; Coffee husk; Submerged fermentation; Solid state fermentation; Biotechnological applications

1. Introduction

In recent years, there has been an increasing trend towards efficient utilization and value-addition of agro-industrial residues such as coffee pulp and husk, cassava bagasse, sugar cane bagasse, sugar beet pulp, apple pomace, etc. [1–6]. Several processes have been developed that utilize these as raw material for the production of bulk chemicals and value-added fine products such as ethanol, single-cell protein (SCP), mushrooms, enzymes, organic acids, amino acid, biologically active secondary metabolites, etc. [1,7–13]. Application of agro-industrial residues in bioprocesses on one hand provides an alternative substrates, and on the other side helps solving pollution problem, which otherwise their disposal may cause. With the advent of biotechnological innovations, mainly in the area of enzyme and fermentation technology, many new avenues have opened for their utilization. In the present paper, we intend to discuss the biotechnological potential of the coffee industry sub-products — coffee pulp and husk for value addition.

2. Coffee

Coffee (*Coffea* sp.) is one of the most important agricultural commodities in the world. *Coffea arabica* and *Coffea robusta* are the two principal varieties of the genus cultivated all over the world for commercial production. Coffee has traditionally been grown under the canopy of the towering forest trees or inter-cropped fruit trees such as bananas, citrus, or leguminous pod trees, that produce other valuable food, timber, fuel wood and fodder crops. This shade canopy also sustainably supports the coffee plants themselves. However, since the 1970s, coffee plantation has dramatically changed with regard to crop pattern and practises, mainly to meet the high demand of coffee, and to tackle the leaf fungus. With the development of hybrid coffee varieties, ‘shade-coffee’ has

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Table 1
Production and consumption of coffee in the world[a]

Country	Production (million sacs)	Country	Consumption (kg per year per person)
Brazil	28[b]	Finland	15
Colombia	15[b]	Sweden	15
Indonesia	7[c]	Denmark	11
Uganda	5.5[c]	Norway	11
India	5[c]	Holland	11
Cote d'Ivoire	5[c]	Luxembourg	8
Guatemala	5[b]	Germany	8
Vietnam	5[c]	Austria	8
Mexico	4[b]	Belgium	8
Ethiopia	4[b]	France	6
		Switzerland	6
		Spain	5
		USA	4.5
		Italy	4.5
		Canada	4
		England	2.5
		Japan	2

[a] Source: Cafe-A la decouverte du Cafe, Le Comite Francais du Cafe, Adexquation Publicite, Paris, 1997.
[b] *Coffea arabica*.
[c] *Coffea robusta*.

turned to 'sun-coffee'. Currently about one million tons of coffee is produced yearly in more than 50 countries [14]. At different stages from harvesting to the processing and consumption, several residues, viz. coffee pulp or husk, leaves and spent-ground are generated in more than two million tons quantity yearly [15]. Brazil is the largest producer of coffee in the world. During 1998, about 30 million sacs of green coffee were produced [14]. Table 1 shows the production and consumption pattern of coffee in the world.

3. Industrial processing of coffee cherries

Fig. 1 shows the general steps involved in the processing of coffee cherries. Industrial processing of coffee cherries is done to isolate coffee powder by removing shell and mucilagenous part from the cherries. There are two methods: dry and wet processing. Depending upon the method of coffee cherries processing, i.e. wet or dry process, the solid residues (sub-products) obtained are termed as pulp or husk, respectively. In Brazil, the coffee cherries are generally processed by the dry method, resulting in coffee husk, which is rich in organic nature and nutrients. It also contains compounds such as caffeine, tannins, and polyphenols. The composition of coffee husk is different from coffee pulp. Coffee leaves, which are mostly collected during harvesting, are generally not considered as residue, but their volume during and after the harvesting creates difficulty in manipulation of the crop. It also facilitates epidemic of pathogens and pests. Coffee spent-ground, the residue obtained during the processing of raw coffee powder to prepare 'instant coffee', is the another residue obtained from the coffee industry. This also contains caffeine, tannins and polyphenols, although in lesser quantity. Due to the presence of these compounds (caffeine, tannins and polyphenols), these organic solid residues show toxic nature and thus have not been utilized beneficially. This has also led to the problem of environmental pollution [16–19].

4. Composition of coffee pulp and husk

Table 2 shows the composition of coffee pulp and coffee husk as reported by some authors. The composition of coffee pulp differs from that of coffee husk, although the nature of the compounds present in both are largely similar. There may be difference in percent composition of the constituents, depending upon the processing mode and efficiency, crop variety, cultivation conditions such as soil type, etc. [19].

Caffeine is an active compound, one of the nature's most powerful and addictive stimulants. It is the principal substance causing the mild stimulation effect of coffee. It is also present in coffee pulp and husk at about 1.3% concentration on dry weight basis. Tannins are generally thought to be an anti-nutritional factor and prevent coffee pulp from being used at greater than 10% of animal feed. The anti-nutritional effects of tannins in animal feed have been discussed by Alzueta et al. [20] and Terrill et al. [21]. Information on coffee pulp tannins is some times contradictory and such data as are available have some times been difficult to interpret because non-specific analytical methods had been used [22–24]. Depending upon the type of cultivar, the tannin contents may differ also. For example, coffee pulp from a yellow-fruited cultivar was significantly richer in condensed tannins (proanthocyanidins) than pulp from the associated red-fruited cultivar [22]. Colmenares et al. [22,25] presented data on condensed tannins (or proanthocyanidins) content of coffee pulp. These authors isolated several proanthocyanidins from coffee pulp. Proanthocyanidins are polymeric polyphenols, which inhibit the germination of *Hemileia vastatrix* race uredospores in vitro.

In contrast to several reports, interestingly, Clifford and Ramirez-Marinez [23] did not find any hydrolysable tannins in coffee pulp derived from five samples of coffee beans. There are contradictions among various authors describing the pectin contents of coffee cherries also [26].

Ramirez-Martinez [27] studied the composition of phenolic compounds in coffee pulp. Chlorogenic acid (5-caffeoylquinic acid) was the main constituent (42.2%). Epicatechin (21.6%, isochlorogenic acid I, II and III, 5.7, 19.3, 4.4%, respectively), catechin (2.2%), rutin (2.1%), protocatechuic acid (1.6%), and ferulic acid (1.0%) were the other compounds. No qualitative or quantitative differences were detected between cultivars of coffee plants resistant and susceptible to coffee leaf rust. The author noted that these values should not be considered as the absolute values, as the contents of the compounds in coffee pulp could vary from time to time. He also added that it was pointless

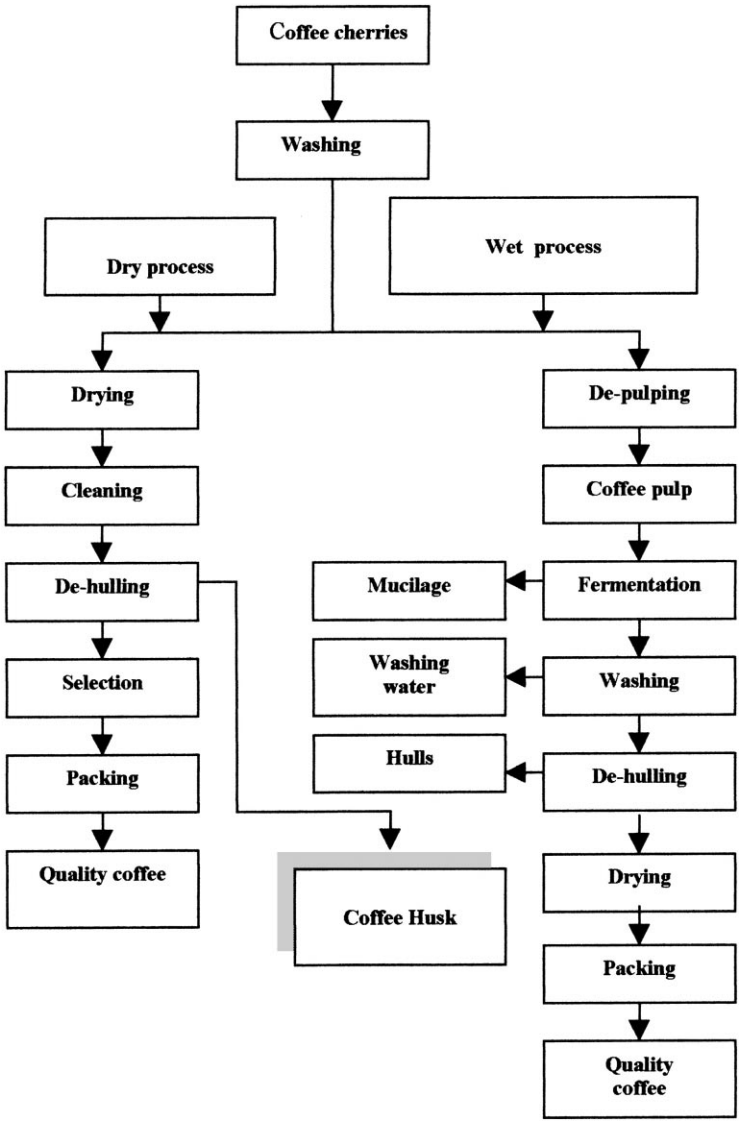

Fig. 1. Industrial processing of coffee cherries.

Table 2
Composition of coffee pulp and husk (% dry weight basis)

Components	1[a,d]	2[b,d]	3[c,e]
Carbohydrates	50	44	57.8
Proteins	10	12	9.2
Fibres	18	21	–
Fat	2.5	–	2
Caffeine	1.3	1.25	1.3
Tannins	1.8–8.56	–	4.5
Polyphenols	–	1	–
Pectins	–	–	12.4

[a] [19].
[b] [87].
[c] [1].
[d] Coffee pulp.
[e] Coffee husk.

to try to establish quantitative comparisons among coffee pulps from different cultivars.

5. Natural microbial flora isolated from coffee pulp/husk

Although several bacteria, yeasts and fungi have been cultivated on coffee pulp and husk for various purposes, filamentous fungi, especially basidiomycetes are the preferred choice and have most widely been employed. Roussos et al. [28] evaluated the natural micro-flora present in coffee pulp and husk, which revealed the presence of a wide variety of micro-organisms, including bacteria, yeasts and fungi. In coffee husk, the population of fungi was slightly higher than bacteria and yeasts but in coffee pulp all the three showed

Table 3
Micro-organisms cultivated on coffee pulp and husk

Micro-organism	Purpose	References
Aspergillus sp.	Protein enrichment	[30]
A. niger[a]	Pectinase production	[41]
A. niger[a]	Pectinase production	[39,40]
A. niger	Pectinase production	[29]
A. niger NRRL 2001	Citric acid	[10]
Ceratocystis fimbriata	Fruity aroma	[36]
F. moniliforme	Gibberellic acid	[52]
G. fujikuroi	Gibberellic acid	[52]
L. edodes	Mushroom production	[43]
Methanogenic thermophilic cultures	Biogas production	[56]
P. tannophilus	Aroma compounds	[53]
P. verrucosum	Caffeine degradation	[85]
Pleurotus sp.	Mushroom production	[47]
P. ostreatus	Mushroom production	[48]
P. ostreatus	Mushroom production	[44]
P. ostreatus[b]	Caffeine degradation	[45]
Penicillium sp.	Pectinase production	[29]
P. chrysosporium	Detoxification	[35]
Rhizopus sp.	Detoxification	[35]
Trichoderma sp.	Growth	[91]
T. harzianum	Growth	[92]
T. viride	Growth	[92]
Volvariella volvacea	Mushroom production	[78]

[a] Mutants.
[b] On coffee husk extract.

almost similar distribution. Boccas et al. [29] isolated and screened 248 fungal cultures from coffee plants and the soil samples from coffee plantation areas. They adopted a three-step selection technique. After the second step, a total of 13 isolates (seven *Aspergillus* and six *Penicillium* sp.) were selected, which were subjected to the third step. Based on enzyme production in SSF, three strains of *Aspergillus niger* and one strain of *Penicillium* sp. were found most suitable for cultivation on coffee pulp (Table 3).

6. Bioprocesses involving coffee pulp and husk

6.1. Cultivation system

The processes involving cultivation of microbes on coffee pulp and husk can broadly be classified into two groups: processes based on liquid submerged fermentation (SmF), and processes based on solid state fermentation (SSF). SSF has generally been preferred over SmF with an intention to reduce the cost of bioprocessing [18,30–33].

6.2. Bioprocess optimization using factorial design experiments

One of the approaches on the optimization of bioprocess parameters using coffee husk and pulp has been to use factorial design and surface response experiments. In this, the experiments generally involved optimization of physical, chemical and biochemical factors such as pH and moisture of the substrate, incubation temperature and period, inoculum size, addition of additional carbon, nitrogen or other nutrients etc. Thus, with several factorial design experiments, the most significant factors could be selected for optimization in the given range. The data obtained are subjected to analysis of variance (ANOVA). In the first step, the process parameters effects on production of experimental compounds is performed. The value of each variable at three levels (two levels and one medium point, -1, 0, and $+1$) is selected. The experimental matrix may consist of several parameters such as pH, initial water contents, temperature of incubation, concentration of sugars, and inoculum size and several runs are performed. The data can be expressed in Pareto chart for each effect, which has been described as a useful tool for identifying which estimated effects are the most important [35]. The Pareto chart of effects is shown in Fig. 1. Plotting the surface graphics at various levels of selected parameter shows increased or decreased response with change of the parameter. From the chart, statistically significant parameters can be selected. Then surface graphics are plotted at various levels to determine the response.

In order to test the influence of the process parameters on the culture to produce volatile compounds, two statistical experimental designs were performed by Medeiors et al. [35] for optimization of production of aroma compounds by *Kluyveromyces marxianus* in solid state fermentation using factorial design and surface response. The parameters studied were the substrate pH, addition of glucose, cultivation temperature, initial substrate moisture and inoculum size. Using 2^5 factorial design, addition of glucose and initial pH of the substrate were found statistically significant for palm bran. Although this experimental design did not show addition of glucose as a significant factor with cassava bagasse, 2^2 factorial design revealed that glucose addition was significant at higher concentrations. Brand et al. [34] also used similar design for their studies on biological detoxification of coffee husk using filamentous fungi in solid state fermentation. Figs. 2 and 3 show the Pareto chart of standardized effects and fitted surface variables, respectively. The first step of SSF using *Aspergillus* sp. for the detoxification of coffee husk consisted in the utilization of a 2^{3-0} factorial design with three factors studied: pH and moisture of the substrate and temperature of fermentation. Results (Fig. 2) were submitted to ANOVA and by eliminating no-significant effects, the data obtained for detoxification of coffee husk showed that the significant factors at the confidence level of 5% were the pH of the substrate and the temperature of fermentation [34]. In the second step of optimization, only two factors, the pH of the substrate (3.0–5.0) and the temperature of fermentation (26–30°C). The results were submitted to ANOVA. Variation of the pH led to an increase in the degradation rate at level of 5%. Fig. 3 shows the surface response for the degradation of caffeine and tannins as

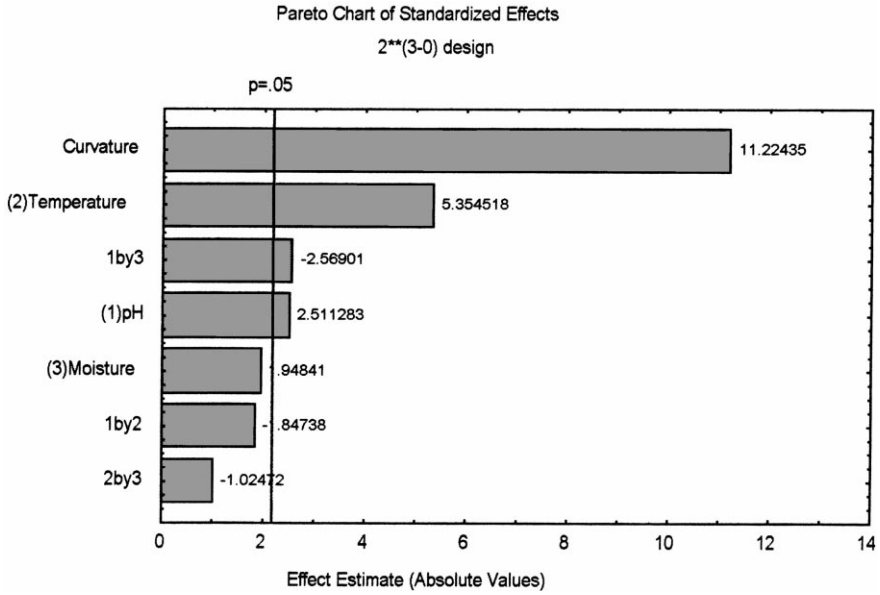

Fig. 2. Pareto chart of standardized effects for biological detoxification of coffee husk.

a function of these two factors, i.e. pH of the substrate and temperature of fermentation [34].

Soares et al. [36] used factorial design experiments for aroma compounds of volatile nature production in SSF. The raw data obtained were analyzed using the Gompertz model, a logistic like equation. Christen et al. [37] and Meraz et al. [38] also used similar model for their studies. The model described the dynamics of the production with respect to time as follows:

$$\mathrm{TV} = \mathrm{TV}_{\max} \exp\{-b \exp(-kt)\}$$

where TV is the total volatile concentration at time t (mmol/l g), $\mathrm{TV}_{\max}$ the maximum production of total volatile at time t (mmol/l g) and b is the fitting parameter. It is used to obtain the time of maximum production rate ($t_{\max}$) as follows:

$$t_{\max} = \frac{\ln b}{k}$$

where k is the production rate (h^{-1}).

Data integration and non-linear Gompertz regression were made with Kaleida Graph program (Abelbeck Software, USA).

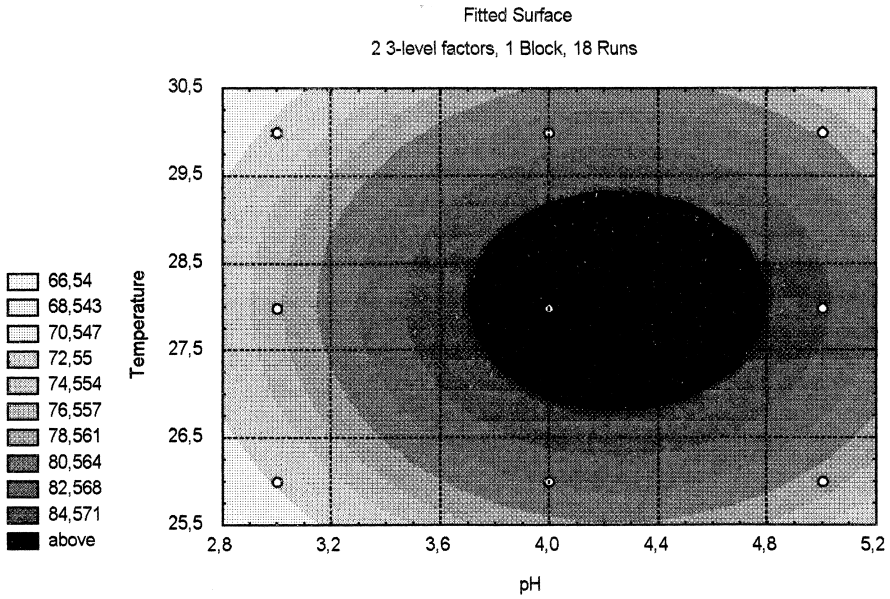

Fig. 3. Surface response graphics of fitted variables for biological detoxification of coffee husk.

6.3. Products and applications

Traditionally, coffee pulp and husk had found only a limited application as fertilizers, livestock feed, compost, etc. These applications utilized only a fraction of available quantity and were not technically very efficient. Recent attempts have focused on its application as substrate in bioprocesses and vermi-composting. Attempts have also been made to detoxify it for improved application as feed, and to it use as an efficient substrate for producing several value-added products such as enzymes, organic acids, flavour and aroma compounds, mushrooms, etc. Since these sub-products contain a good amount of fermentable sugars, these constitute an appropriate substrate for the cultivation of moulds and yeasts.

6.3.1. Production of enzymes

One of the earliest approaches on the applications of coffee pulp and husk has been for the production of enzymes such as pectinase, tannase, caffeinase, etc. [3,29,39]. Boccas et al. [29] carried out SSF using coffee pulp for the production of pectinase. One of the strain, *A. niger* V22B35, produced four times more enzyme than the reference strain of *A. niger* CH4. Pectinase production in SSF and SmF (of coffee pulp) using hyper-producing mutants of *A. niger* and depressed mutants were also reported by Antier et al. [40] and Minjares-Carranco et al. [41]. Water activity level was found to play an important role in culture's efficiency. Special selective media helped to obtain strains adapted for SSF or SmF of coffee pulp.

Boopathy [42] studied the metabolism of protein, carbohydrates and lipid during anaerobic fermentation of coffee pulp. Cellulases, protease, β-amylase, β-glucosidase and lipase were produced in the digester and the enzyme activity was higher in coffee pulp than compared to that in a cow-dung digester.

6.3.2. Production of mushrooms

Due to their good organoleptical properties of flavour and aroma, and good nutritional and therapeutically values, mushrooms cultivation has increased dramatically all over the world. Several agro-industrial residues offer good potential in this regard [1,11–13]. Although, first attempts on mushroom cultivation on coffee industry residues were made more than 10–15 years ago, not much has been done still and only little published information is available [43–48]. Recently, Fan et al. conducted a systematic study on cultivation of *Lentinus edodes*, *Pleurotus* sp. and *Volvariella* sp. using different residues such as coffee husk, leaves and spent-ground, individually or in mixture [43–45]. Studies on the cultivation of *L. edodes* showed that after 20 days growth on untreated coffee husk, the mycelia regressed and humidity was augmenting. There was no fruiting body. Treatment of coffee husk with hot water (cooked in water and filtered) was found useful for its utilization as the substrate by the fungal culture. This resulted in good mycelial growth and good fruiting bodies were obtained. The biological efficiency reached 85.8%. When a strain of *Pleurotus* sp. was used, the first fructification occurred after 20 days of inoculation. The biological efficiency reached 96.6%. It was concluded that coffee husk could be used for the cultivation of these mushrooms. Growth of these fungal species resulted in an increase in the protein content and a decrease in the fibres contents of coffee husk. The biotechnological advances could lead to an effective and economical way to utilize this otherwise unused residue and improve the economy of the coffee industry.

6.3.3. Citric acid and gibberellic acid

Coffee husk has been used to produce citric acid in solid state fermentation using strains of *Aspergillus* sp. [49–51]. It was shown that coffee husk resulted in higher citric acid production (g/kg substrate) than wheat bran, rice bran and de-oiled rice bran. Yields on the basis of amount of starch/sugar consumed were almost similar for sugarcane press-mud and coffee husk. Vandenberghe et al. [51] used coffee husk admixed with a cassava bagasse hydrolysate. Citric acid production reached 12.72 g/kg dry matter. Addition of mineral ions such as zinc, copper, iron and magnesium has been found useful for fungal growth and activity to produce citric acid. These studies showed the feasibility of using coffee husk as substrate in SSF.

In a recent work, Machado et al. [52] reported the production of gibberellins (plant hormones) in SmF and SSF utilising coffee husk as the carbon source. Five strains of *Gibberella fujikuroi* and one of its imperfect state, *Fusarium moniliforme* were used for comparison. Results showed the production of gibberellic acid (GA_3) in all the fermented samples. SSF appeared superior to SmF.

6.3.4. Aroma compounds

A novel approach on value addition of coffee husk has been to use it as substrate for the production of aroma compounds for food industry application with yeast and fungi [36,37,53]. There has been a remarkable shift in the consumer's choice for naturally produced food and flavour compounds (in comparison to synthetic ones) for human use globally. Thus, production of aroma compounds through fermentative means has a promising future. Soares et al. [53] used the yeast *Pachysolen tannophilus* in SSF for synthesizing aroma compounds. Results on using coffee husk and coffee husk extract showed superiority of steam treated coffee husk. The yeast culture produced a strong alcoholic aroma with fruity flavour. Along with ethanol, which was the major compound produced, acetaldehyde, ethyl acetate, isobutanol, isobutyl acetate and ethyl-3-hexanoate and isoamyl acetate were also produced by the culture, giving a strong pineapple aroma. When leucine was added to the medium, a strong banana odour was found with increased amounts of isoamyl alcohol and isoamyl acetate.

6.3.5. Production of biogas

Attempts have been made to use coffee industry residues, mainly coffee pulp and husk for biogas production in anaerobic digestion [54–58]. According to the estimates, from one ton of coffee pulp, about 131 m^3 biogas could be produced by anaerobic digestion which would be equivalent to 100 l of petrol in fuel value [54]. Kida et al. [55] studied anaerobic digestion of coffee waste solids in one- or two-phase system. They found that treatment in the two-phase methane fermentation could be useful using coffee waste alone as the substrate. Kostenberg and Marchaim [56] aimed to develop biogas technology for the treatment of coffee industry wastes and evaluation of digested material as a growth medium for horticulture.

A case study was performed in Tanzania to study the feasibility of using agro-industrial residues such as coffee industry solid wastes (pulp), sisal pulp, sugar filter, maize bran, etc. for biogas and electricity production and tackle the energy demands of the country [59]. Robusta and arabica coffee solid residues generated about 650 and 730 m^3 CH_4 per ton volatile solids, respectively, which were highest in comparison to any other agro-industrial residues (230, 400 and 450 m^3 for sugar filter, sisal and maize bran, respectively).

6.4. Production of feed

6.4.1. For cattle

One of the current applications which coffee pulp finds is its use as food for animals [60–62]. With its average contents of about 50, 10, 2.5 and 18% carbohydrate, protein, fat, and fibres, respectively, coffee pulp appears to be a useful feed supplement for animals. Several studies have been carried out to evaluate it from an animal nutrition point of view. Low feed intake, protein digestibility and nitrogen retention are major factors limiting the use of coffee pulp as animal feed. These effects appear to be due to the presence of caffeine, tannins and other polyphenols in coffee pulp. Coffee pulp could be best considered as possible feed ingredient if the anti-physiological (anti-nutritional) factors could be eliminated, or neutralized or at least degraded to a minimal level. In a recent review Aregheore [63] discussed about the application of several unconventional feed stuffs including coffee pulp. It was advocated that these make the ration unpalatable and unacceptable to animals. They also interfere with the nutrients bioavailability and utilization. Among the various means to detoxify these, fermentation could be an attractive way.

Demeke [64,65] evaluated the nutritive value of forage ensiled coffee pulp supplemented with sugarcane stems, sugarcane tops, elephant grass (*Mennisetum purpureum*), or sesbania sesban. On the basis of quality and palatability, pasteurized ensiled coffee silage were superior.

6.4.2. Feed for other purposes

Studies have also been made to use the coffee pulp as a feed ingredient for fishes etc. Experiments conducted to determine the efficacy of coffee pulp as a constituent of a feed used in the culture of common carp (*Cyprinus carpio* L.) and catfish (*Clarias mossambicus* Peters) resulted in decreased growth rates. The authors claimed that these results could be applied to a cost structure forecast of a fish farm [66,67]. There are several other reports describing the application of coffee pulp in feed [68–72].

6.5. Composting/vermi-composting

One of the other traditional applications of coffee pulp has been to use it as compost [73]. Generally an open piles system has been employed which is not an efficient method as it leads to a product with poor desirable characteristics. The main cause for this is that the macro- and micro-fauna such as *Acarida*, *Coleoptera*, *Collembola*, *Diptera*, *Eisenia*, *Oligochaeta*, *Perionyx*, *Thysanoptera*, etc. generally grow on the upper layers of the piles, without much deeper penetration. Vermi-composting, which takes the advantages of biological and physiological capabilities of the growing macro-fauna in the solid residues has been suggested as an attractive alternative to this. There are several species of earthworm found in coffee pulp such as *Amynthas gracilis* (Kinberg, 1867), *Dichogaster* sp., *Eisenia andrei*, *Eisenia fetida*, *Perionyx excavatus* (Perrier, 1872), etc. These could be beneficially exploited for vermi-composting. Orozco et al. [74] used the earthworm, *E. fetida* for vermi-composting and showed that the process was very efficient. Ingestion of the pulp by the earthworm resulted an increase in available P, Ca, and Ng but a decrease in K contents. In a recent review, Arnanda and Barois [75] examined different examples of coffee pulp vermi-composting. They concluded that coffee pulp vermi-composting has techno-economical feasibility and usefulness in handling this huge and major solid waste of coffee industry.

6.6. Biological detoxification of coffee pulp and husk

Due to the presence of anti-physiological and anti-nutritional factors, coffee pulp and husk is not considered an adequate substrate for bioconversion processes. Consequently, most of the pulp and husk remains unutilized or poorly utilized. If these toxic constituents could be removed, or, at least degraded to a reasonably low level, it would open new avenues in their utilization as substrate for bioprocesses. With this in mind, several authors have worked on detoxification of coffee pulp and husk through various means. This includes physical, chemical and microbial methods [17,33,35,76–78]. Some of the physical and chemical methods, although successful, were expensive and did not provide economic feasibility. This led focus on developing biological methods.

SSF has been frequently used for the biological detoxification of coffee husk using fungal strains [30,35,78]. By selective screening on an agar medium coffee husk, three strains of *Rhizopus* sp., which showed good growth

(radial growth and biomass production), were compared with two strains belonging to basidiomycetes, viz. *Phanerochaete chrysosporium* for the degradation of caffeine and tannins in coffee husk [35]. Both the cultures, i.e. *Rhizopus* sp. as well as *P. chrysosporium* grew well on coffee husk. *Rhizopus* sp., however, appeared superior to *P. chrysosporium* as it resulted higher caffeine and tannin degradation in relatively shorter period. Under the optimized conditions of substrate pH, initial moisture, inoculum size, temperature and aeration, *Rhizopus* sp. degraded 87 and 65% caffeine and tannins in comparison to 70 and 60%, respectively, by *P. chrysosporium*. Fan et al. [78] studied degradation of caffeine in coffee husk using a strain of *P. ostreatus* LPB 09. Media supplemented with caffeine showed caffeine tolerance by the strain. Mycelial growth rate and biomass were 16 and 14.5%, and 65.6 and 7.6% less with 100 and 1000 g/l caffeine concentration. At 2500 g/l concentration, there was no fungal growth at all. Mycelial analysis revealed the presence of caffeine in it, showing that *P. ostreatus* actually did not degrade caffeine but accumulated it (0.575% caffeine in dry mycelia). In SSF, caffeine concentration in the residue after fruitification diminished by 85.4%, however, the fruit-body contained 0,157% caffeine. It was claimed that *P. ostreatus* LPB 09 could be used for bioremediation of residues containing caffeine.

Some bacterial and fungal strains such as *Bacillus coagulans*, *Pseudomonas aeruginosa*, *P. putida*, *Penicillium rouquifortii*, *Penicillium curtosum*, and *Pleurotus* sp. have been stated to have the capacity of degrading caffeine [33,79–85]. Schwimmer and Kurtzman [84] achieved total degradation of caffeine by a fungal strain of *P. curtosum* in roast coffee infusions. Roussos et al. [85] studied caffeine degradation by *Penicillium verrucosum* in solid state fermentation of coffee pulp with and without external nitrogen supplementation. Results indicated that in spite of the limited growth of the culture without any external nitrogen, caffeine degradation was almost complete. Addition of nitrogenous compounds rather inhibited caffeine degradation. Hakil et al. [86] achieved considerable degradation of caffeine using several strains of filamentous fungi. Porres et al. [87] studied the degradation of caffeine and polyphenols in coffee pulp through silage. Under different processing conditions, a reduction of 13–63, 28–70 and 51–81% in caffeine, total polyphenols and condensed tannins, respectively, was achieved. It was concluded that silage presented an ideal method to reduce the anti-physiological compounds contents in coffee pulp.

6.7. Hydrolysis of coffee husk

One another approach on coffee husk utilization has been to hydrolyse it and use the hydrolysate for various purposes. Hydrolysis could be performed using dilute acids or steam treatment. Urbaneja et al. [88] used dilute sulphuric acid for hydrolyzing the coffee pulp. They obtained xylose, arabinose, fructose, glucose, sucrose and maltose. Arabinose was produced in highest concentration followed by glucose. Overall efficiency of the hydrolysis was 64 and 67% for total and reducing sugars, respectively. Woiciechowski et al. [89] compared hydrolysis of coffee husk with and without mineral acids. The water-soluble fraction composed mainly sugars from the hemicellulose. Best results (48.18 g/l of reducing sugar) were obtained when the hydrolysis was carried at 121°C for 15 min, without addition of any acid. The hydrolysate was used for the production of lactic acid [90].

7. Conclusions

Coffee pulp and husk offer potential opportunities to be used as substrate for bioprocesses. Recent studies have shown their feasibility for the production of a host of products, such as enzymes, aroma compounds, mushrooms, etc., thus adding value to the sub-product. However, much remains to be done in these areas. Bioprocess optimization using factorial design and surface response experiments could be useful in selecting production parameters. Traditional application of coffee pulp and coffee husk as livestock feed and compost could also be improved using efficient biotechnological methods. Vermi-composting offers an attractive alternative. Biological detoxification of coffee pulp and husk holds promise as the removal (degradation) of anti-physiological and anti-nutritional factors such as caffeine and tannins would open new avenues for the utilization of these sub-products. Coffee husk hydrolysate also offers good opportunities to be used as substrate for bioprocesses. However, it would be necessary to look into the economic feasibility of such processes.

Application of coffee pulp and husk in bioprocesses on the one hand may provide alternative substrates, and on the other help in solving pollution problems, which their disposal otherwise causes.

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