REVIEW ARTICLE

Advancement in sandhana kalpana and role of biotechnology: Need of research for diabetic patients

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ABSTRACT

The diabetic population of India is increasing day by day. At the meantime, diabetic patients also adopt ayurvedic treatment for their other chronic ailments. Sandhana Kalpana, which deals with hydroalcoholic oral dosage form, has a number of formulations for various ailments in ayurvedic treasures. However, ayurvedic practitioners failed to prescribe this dosage form to the diabetic patients because of more percentage of sugar content in asava-arishta. Hence, there is need to replace jaggery from other carbohydrate sources, namely, starch and cellulosic material which produce less sugar percentage in asava-arista. As *Saccharomyces cerevisiae* directly ferment the sugar molecule into alcohol, but starchy and cellulosic feedstocks required specific treatment, before the fermentation process. Different researchers have been done which revealed that uses of engineered strains are helpful in direct production of alcohol. Therefore, it is required to change the feedstock and, respectively, the strain of yeast. *Schwanniomyces castellii* and *Endomycopsis fibuligera* are some of the available strains of yeast which when used with can transform the starch directly into alcohol. Further, researchers are needed to manufacture asava-arishta from these first and second generation feedstocks with the care that therapeutic value should not to be altered.

Key words: Asava-arishta, carbohydrate feedstocks, diabetes, recombinant technology, yeasts

INTRODUCTION

In Ayurveda, the pharmaceutics is deal under the heading of Bhaishajya Kalpana (BK). BK deals with the wide range of medicinal preparations primarily Pancvidhya Kashaya Kalpana (Primary preparations), namely, Swarasa (Expressed juice), Kalka (Paste), Kashaya (Decoction), Hima (cold water infusion), and Phanta (Hot water infusion), and secondary preparations such as Churna (powder), Sneha (Medicated oils), Avaleha (linctus), and Asava and Arishta (Alcoholic preparations).[1] Among these dosage forms, *"Sandhana kalpana"* is a unique dosage form in which fermented formulations are prepared. To manufacture these medicines, liquid basic drugs (juices or decoctions) are kept for fermentation as indicated in the classics. In this process, self-generated alcohol is produced by in-source material used in pharmaceutical procedure and is not added from outside. Here, ethyl alcohol is not the only product yielded but is a part of many other organic compounds, further, alcohol is formulated and extraction of active principles of the herbal drugs is done. Thus, these formulations have a longer shelf life, quick absorption and action, and excellent therapeutic efficacy as compared to other ayurvedic herbal medicines. Therefore, the ayurvedic fraternity relies on this unique dosage form, i.e., *Sandhana kalpana* (*Asava*, *Arishta*, *kanji*, etc.) to treat diseases in routine practice[2] [Table 1]. Asava is prepared without boiling the drug in liquid media or it may be prepared by Swarasa, Hima, and Phanta. Arishta is prepared by making the decoctions of herbs. *Asava-Arishta* is considered as the best formulation in Ayurveda because they possess better keeping quality, which is likely due to the contribution of fermentation. Microbes mediate this process and enhance therapeutic properties, which may be

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Table 1: Sandhana and its importance and list of asava and arista in different disease.

System of body	Ailments	Asava-arishta used in ailments
Gastrointestinal system	Constipation	Arvindasava
	Appetizer	Arvindasava
	Stomach Pain	Viswamrita
	Indigestion	Pippalyasavam, Amritarishta, Chitrakasavam
	Vomiting	Drakshasava, Mrigamadasava
	Peptic ulcers	Kumaryasava
	Intestinal problems	Loharishta, Bilvasava, Madhukasava
	Intestinal parasites	Kutajarishta, Vidangarishta
	Diarrhea	Babularista, Bilvasava, Takrarishta
	Amebic dysentery	Kutajarishta, Mukta rishta
	Abdominal disorders	Punarnavarishta
	Jaundice	Dhatriarishta, Dasamoolarishta
	Enlargement of liver	Kumaryasava, Bringarajasava
	Liver Disorder	Punarnavarishta
Excretory system	Piles	Devadarvyarishta, Amritarishta, Chitrakasavam
	Diuretic	Balarishta, Chandanaasava, Drakshaasva
	Urinary disorders	Ashokarishta, Devdaruarishta, Abhyarishta
Nervous System	Nerve tonic	Saraswatharishta, Aswagandharista
	Paralysis	Balarishta, Vidangasava, Dasamoolarishta
	Nervous weakness	Dhanyamla
	Nervous disorders	Balarishta, Aswagandharishta,
	Improves concentration	Saras
Blood circulatory System		
	Blood purifier	Ashokarishta, Sarivadyarista, Babularishta, Panchatiktarishta
	Anemia	Lohasava, Khadirarishta, Draksharishta, Rohitarishta
	Blood nourishment	Draksharishta, Drakshasava, Kumaryasava
Reproductive System		
	Menstrual disorders	Ashokarishta
	Seminal weakness	Saraswatharishta
	Sterility in females	Dasamularishta
	Vaginal Disease	Ashokarishta, Dasamularishta
Respiratory System		
	Cough	Vasarishta, Sudarshanasava, Draksharishta
	Asthma	Shirisharishta, Kanakasava, Vasarishta, Babularishta
	Lung disease	Dasamoolarishta
	Bronchitis	Babularishta, Vasarishta, Draksharishta
Other ailments		
	Fever	Kutajarishta, Panchatiktarishta, Sudarsanasava, Amritarishta
	Diabetes	Lodhrasava, Useerasavam, Loharishta

Contd...

Table 1: (*Continued*)

System of body	Ailments	Asava-arishta used in ailments
	Weakness	Aswagandharishta, Loharishta, Draksharishta
	Health tonic	Balarishta, Aswagandharishta
	Rejuvenator	Mahamanjisthadyarishta
Excretory System		
	Rheumatoid Arthritis	
	Gout	
	Osteoarthritis	

due to microbial biotransformation of the initial ingredients (phytochemical compounds) into more effective therapeutics as end-products, i.e., alcohol–aqueous milieu. Requirements for asava-arishta preparations as per ayurvedic pharmaceutics need following materials.[3]

- Dravya (drug substance used for decoction/juice).
- Drava Dravya (liquid media).
- Madhura Dravya (sweetening agents, namely, Jaggery, sugar, honey, etc.).
- Sandhana Dravya (fermentative agents).
- Prakshepa Dravya (drugs for perfuming/additives).
- Sandhana patra (fermentation vessel).

Importance of Fermentation

Fermentation is a process by which sugars are converted to alcohol and carbon dioxide by yeast in the absence of oxygen. The sugar could be a simple sugar such as glucose or fructose or a more complex sugar such as sucrose (dimer of glucose and fructose). In any chemical reaction, including fermentation, the amount of products formed is determined by the amount of reactants used. Thus, increasing the amount of sugar used increases the amount of alcohol being produced since increasing the amount of sugar increases the amount of reactants going into the fermentation reaction. The source of the sugar can determine the flavor of the alcohol produced.

Role of Microbes in Sandhana Kalpana

Involvement of microbes in fermentation process is a compulsory trend. During fermentation process, microbes help in the generation of alcohol from sweetening agents or from carbohydrate sources. In one of the study, the effect of addition of yeast (*Saccharomyces cerevisiae*) and *Dhataki pushpa* to fermenting media was studied.[4] The study reveals that the onset and completion of fermentation process in the samples containing yeast were quick, as in these samples, fermentation started on the 2nd day and was completed within 1 month. However, in the group where yeast was not used, fermentation started on the 5th day and was completed in the 2nd month. Fermentation may be delayed because of natural growth and multiplication of yeast cells as well. To know the role of other microbes if any, in production of alcohol during fermentation process a study was conducted.[5] In this study, microbial composition at initial stages of fermentation of was assessed by culture-independent 16S rRNA gene clone library approach. At the 0 day of fermentation, *Lactobacillus* sp., *Acinetobacter* sp., *Alcaligenes* sp., and *Methylobacterium* sp. were recovered but were not detected at 8 days of fermentation. Initially, microbial diversity increased after 8 days of fermentation with 11 operational taxonomic units (OTUs), which further decreased to three OTUs at 30 days of fermentation. *Aeromonas* sp., *Pseudomonas* sp., and *Klebsiella* sp. dominated till 30 days of fermentation. The predominance of C-Proteobacteria and the presence of galloyl derivatives at the saturation stage of fermentation imply tannin degrading potential of these microbes. These studies confirm the role of different microbes from the initial stage to final stage of fermentation. Microbes play a key role in fermentation process which finally aids in conversion of carbohydrate sources into alcoholic production and degradation of phytoconstituents into therapeutics content.

Demerit for Diabetic Patients

A wide range of formulations in sandhana kalpana is described in ayurvedic treasure for the treatment of various ailments. India is now becoming the hub for diabetic patients,[6] and at the same time, Ayurveda is gaining global attention for the treatment of chronic diseases. At the meantime, diabetic patients also adopt ayurvedic treatment for their other chronic ailments. However, ayurvedic doctors failed to prescribe this dosage form to thediabetic patients because of more percentage of sugar in asava-arishta. As this dosage form has a high percentage of sugar which restricts its use among diabetic population.

Need of Less Sugar Content in Sandhana Kalpana for Diabetic Patients

It is the need of the time to replace the traditionally using sweetening agents such as jaggery and honey so that sugar can be produced in a controlled manner. There are some other feedstocks [Table 2] other than jaggery which contain less number of glucose molecule, hence after fermentation, they will produce least percentage of sugar and optimum level of

Table 2: Feedstocks other than sugar: Products which can replace the sweetening agent

Starchy feedstocks	Cellulosic feedstocks (fibrous parts of the plants)
Sweet potato, rice flour,	Barley, rice stalk
Potato, wheat flour	Maize, vegetables
Cheese, whey, invert syrup	Oat, cereals

alcohol in the final product, namely, asava and arista. Some of the researchers have been done on these feedstocks, namely, banana peel, potato flour, etc., and the result showed that in the final product there is least percentage of sugar present with optimum percentage of alcohol. Results of some more researchers are described to enlighten this fact.

FIRST-GENERATION FEEDSTOCKS

Sugar Feedstocks

Fermentation involves microorganisms that use the fermentable sugars for food and in the process produces ethanol and other byproducts. These microorganisms can typically use the 6-carbon sugars, one of the most common being glucose. Therefore, biomass materials containing high levels of glucose or precursors of glucose are the easiest to convert to ethanol. Although fungi, bacteria, and yeast microorganisms can be used for fermentation, specific yeast (*S. cerevisiae* also known as bakers' yeast) is frequently used to ferment glucose to ethanol. Theoretically, 100 g of glucose will produce 51.4 g of ethanol and 48.8 g of carbon dioxide. However, in practice, the microorganisms use some of the glucose for growth and the actual yield is <100%. Other biomass feedstocks rich in sugars (materials known as saccharides) include sugar beet, sweet sorghum, and various fruits.

In one of the researchers, a combination of *Gurmar* leaves (*Gymnema sylvestre*), *Methi* seeds (*Trigonella foenumgrecum*), *Vijayasar* heartwood (*Pterocarpus marsupium*), *Jamun* seeds (*Eugenia jambolana*), *Karela* (*Momordica charantia*), and *Dhataki pushpa* (*Woodfordia fruticosa*) are coarsely ground to small pieces and extracted twice with water using boiling pan. The extract thus prepared is transferred to a benchtop fermentor. The nutrient (invert syrup) was added in a batch-fed mechanism.[7] The medium was inoculated with baker's yeast and spicy materials such as *Piper longum, Elettaria cardamomum, Myristica fragrans,* and *Amomum subulatum* were topped over the fermentation medium. The temperature of the fermentation medium was maintained at 30°C. The samples were estimated at regular intervals for alcohol generation and residual sugar content. The amount of alcohol produced at the end of fermentation was 7–11% v/v and the residual sugars content was <1% w/w. This particular fed-batch mechanism shall have the advantage of avoiding any kind of foaming problem during fermentation. These experimental results further emphasize the effectiveness of the process and the end results of the process are same irrespective of the type of fermentation.

The production of ethanol from sweet potato flour by co-culture of *S. cerevisiae* and *Trichoderma* species in solid state fermentation. Here, the study was aimed at eliminating the enzymatic saccharification step by using a co-culture of Trichoderma sp. as an amylolytic mold along with Saccharomyces cerevisiae (strain CET).[8] The concentration of ethanol increased with the increase of fermentation of fermentation time and yeast biomass. The maximum ethanol (154 ± 4 g/kg substrate) concentration (95%) was obtained after 72 h of incubation. At the time (72 h) when the maximum concentration of ethanol was achieved, 78% of sugar consumed was converted to ethanol. Sweet potato flour is available in plenty in the Asia-Pacific regions, including in Orissa (India), but it is commercial potential for ethanol has not been fully explored. Being a cheap source of fermentable carbohydrate bioresource, it could be employed for the production of ethanol. Ethanol production ability by the coculture (*S. cerevisiae* and *Trichoderma* sp.) was 65% higher than the single culture of *S. cerevisiae* from unsaccharified sweet potato flour.

Starchy Feedstocks

Another potential carbohydrate feedstock is starch. Starch molecules are made up of long chains of glucose molecules. Thus, starchy materials can also be fermented after breaking starch molecules into simple glucose molecules. Examples of starchy materials commonly used around the world for ethanol production include cereal grains, potato, sweet potato, and cassava. Cereal grains commonly used in the US for ethanol production include maize and wheat. Starchy materials require a reaction of starch with water (hydrolysis) to break down the starch into fermentable sugars (saccharification).

Flour prepared from potato tubers (*Solanum tuberosum*) after cooking and drying at 70°C was used for ethanol production.[9] Fermentation of hydrolysates with *S. cerevisiae* HAU-1 at 30°C for 48 h resulted in production of 56.8 gl-1 ethanol. Supplementation of nitrogen sources to potato flour did not contribute significantly to ethanol yield. Simultaneous saccharification and fermentation of hydrolysate were as effective as separate hydrolysis and fermentation.

An alternative approach in place of jaggery, wheat flour is used to maintain the same percentage of alcohol by *in situ* fermentation without keeping the risk of enhancement of sugar levels in diabetic patients, almost 6.44% v/v alcohol generation was observed by this alternative formulation by keeping all the ingredients as such and unaltered.[10] Although the *in vitro* results indicated its positive effects for diabetic patients, with 3.6% of sugar in the formulated product,

compared to that of traditional product with 30% of sugar concentration, still *in vivo* experimentation will be reflecting the actual claim, and thereby it is expected to conclude its beneficial effects in diabetic cases with the clinical situation.

In a recent study in which arishta is prepared where jaggery is replaced by banana peel and yeast was replaced by mutant strains which were developed from the wild strain of *S. cerevisiae* using UV irradiation technique and by varying the exposure timings. All the mutant cultures were used for ethanol production using banana peel as a substrate in a batch fermentor. The effect of temperature, pH, and initial substrate concentration on ethanol production was studied and optimized. One mutant strain yielded a maximum ethanol production of 9 g/L under identical conditions. The conditions were optimized for this mutant strain at a temperature of 33°C, pH 4.5, and initial substrate concentration of 10% (w/v).[11]

Direct and efficient production of ethanol by fermentation of raw corn starch was achieved using the yeast *S. cerevisiae* codisplaying *Rhizopus oryzae* glucoamylase and *Streptococcus bovis*. In 72-h fermentation, this strain produced 61.8 g of ethanol/liter, with 86.5% of theoretical yield from raw cornstarch.[12] Alcohol fermentation of starch was investigated using a direct starch fermenting yeast, *S. cerevisiae* SR93, constructed by integrating a glucoamylase-producing gene (STA1) into the chromosome of *S. cerevisiae* SH1089. The glucoamylase was constitutively produced by the recombinant yeast. The ethanol concentration produced by the recombinant yeast was 14.3g/L which was about 1.5-fold higher than by the conventional mixed culture using an analytic microorganism and a fermenting microorganism. About 60% of the starch were converted into ethanol by the recombinant yeast and the ethanol yield reached its maximum value of 0.48 at the initial starch concentration of 50g/L. The amount of ethanol produced in the fed-batch culture increased about 20% compared to the batch culture.[13] Cereals might also have potential for use in grain distilleries and ethanol production. Studies of the properties of wheat, maize, sorghum, and millet showed that they had good potential for grain distilling and ethanol production.[14]

Alcoholic fermentation of cheese whey permeate was investigated using a recombinant flocculating *S. cerevisiae* and *Kluyveromyces marxianus* enabling for lactose metabolization. For cheese whey permeate having a lactose concentration of 50 gL(-1), total lactose consumption was observed with a conversion yield of ethanol close to the expected theoretical value. The use of 2 times concentrated cheese whey permeate, corresponding to 100 gL(-1) of lactose concentration, was also considered allowing for obtaining a fermentation product with 5% (w/v) alcohol.[15]

The use of microorganisms as catalyst is an alternative technology for biological treatment of cheese whey. Cheese whey constitutes an inexpensive and nutritious, rich raw material for production of ethanol by fermentation. The conclusion to be drawn from the findings is that *K. marxianus* is more promising for ethanol productions from whey permeate than *Candida kefyr*.[16]

SECOND-GENERATION FEEDSTOCKS

Cellulosic Feedstocks

Cellulosic materials are comprised lignin, hemicellulose, and cellulose and are thus sometimes called lignocellulosic materials. Cellulose molecules consist of long chains of glucose molecules as do starch molecules but have a different structural configuration. These structural characteristics plus the encapsulation by lignin makes cellulosic materials more difficult to hydrolyze than starchy materials. Hemicellulose is also comprised long chains of sugar molecules in addition to glucose (a **6**-carbon or hexose sugar) contains penthouses (**5**-carbon sugars). The conversion of lignocellulose to ethanol involves a series of enzymatic steps for hydrolysis or saccharification of the constituent polysaccharides and subsequent fermentation of the released hexose and pentose sugars.

Enzyme requirements for Lignocellulosic Feedstocks[17]

Although *S. cerevisiae* is approved industrial ethanol producer in traditional starch-based process, it will be not easy task to provide that microorganism with the ability to convert lignocellulosic biomass to ethanol. The carbohydrate components of lignocellulose (cellulose and hemicellulose) are tightly bound to lignin, making the sugars largely inaccessible to enzymes. "Before enzymatic hydrolysis, pretreatment with acid or alkali is generally needed to fully maximize the release of sugars from any lignocellulosic biomass," yeast utilized these carbon sources and can be further fermented and isolated in higher temperature for ethanol production.[18]

In addition, a pretreatment step is required to disrupt the tightly packed cellulose structure and allow access to the enzymes. The stages to produce ethanol using a biological approach are as follows**:** [19]

1. A "pretreatment" phase, to make the lignocellulosic material such as wood or straw amenable to hydrolysis.
2. Cellulose hydrolysis (that is, cellulolysis) with celluloses, to break down the molecules into sugars.
3. Separation of the sugar solution from the residual materials, notably lignin.
4. Microbial fermentation of the sugar solution.
5. Distillation to produce roughly 95% pure alcohol.
6. Dehydration by molecular sieves to bring the ethanol concentration to over 99.5%.

In 2010, a genetically engineered yeast strain was developed

to produce its own cellulose-digesting enzymes. Assuming this technology can be scaled to industrial levels, it would eliminate one or more steps of cellulolysis, reducing both the time required and costs of production.[19] Many years of research have been applied to engineer a yeast strain that can metabolize xylose as well as the hexose sugars found in biomass. Much of this research has recently focused on enhancing the fermentation performance of *S. cerevisiae* strains expressing heterologous enzymes from bacterial or fungal xylose utilization pathways.[20] Recently, special microorganisms have been genetically engineered which can ferment 5-carbon sugars into ethanol with relatively high efficiency.[21] Research laboratories around the world have been trying to solve the problem of poor xylose utilization using xylose-fermenting yeast, *Pichia stipites*.[22]

Hemicellulose to Ethanol

Sakamoto *et al*. showed the potential of genetic engineering microbes[23] to express hemicellulase enzymes. The researchers created a recombinant *S. cerevisiae* strain that was able to:

1. Hydrolyze hemicellulase through codisplaying endoxylanase on its cell surface and
2. Assimilate xylose by expression of xylose reductase and xylitol dehydrogenase.

The strain was able to convert rice straw hydrolysate to ethanol, which contains hemicellulosic components. Moreover, it was able to produce ×2.5 more ethanol than the control strain, showing the highly effective process of cell surface engineering to produce ethanol.

Metabolic Engineering Yeast Strains: A Ray of Hope for cofermentation

In ayurvedic treasures, jaggery is used as the carbohydrate source and Dhataki pushpa (*W. fruticosa) as a* fermenting agent, but nowadays, yeast *S. cerevisiae* is used for the production of ethanol in sandhana kalpana. Jaggery itself produces high concentration of sugar (approximately 30%) which is quite harmful for the diabetic patients. If jaggery is replaced by other carbohydrate sources which are having less glucose molecule then in final product least percentage of sugar produced (approximately 1%) with sufficient amount of ethanol. A number of feedstocks such as potato tuber, banana peel, wheat flour, cheese whey, and rice stalk and different feedstocks which include starch, cellulose, and hemicellulose need enzymatic reaction followed by fermentation and produce adequate amount of ethanol with least or no sugar percentage. Starch and cellulosic substrate required enzymatic process before fermentation to release sugar molecules from the substrate which is a very hectic and time-consuming process. Hence, direct fermentation from starch and cellulosic substrate can only be achieved by involvement of biotechnology. Collaboration of biotechnology with Ayurveda will help to provide engineered yeast, which can direct ferment the starch and cellulosic substrate for the production of self-generated alcohol in asava and arista preparation.

In recent years, metabolic engineering of microorganisms used in ethanol production has shown significant progress. Besides, *S. cerevisiae* microorganisms such as *Zymomonas mobilis* and *Escherichia coli* have been targeted through metabolic engineering for cellulosic ethanol production.[24] Recently, engineered yeasts have been described efficiently fermenting xylose and arabinose and even both together. Yeast cells are especially attractive for cellulosic ethanol processes because they have been used in biotechnology for hundreds of years, are tolerant to high ethanol and inhibitor concentrations and can grow at low pH values to reduce bacterial contamination.[19]

When mixed culture of mutant *Schwanniomyces castellii* and *S. cerevisiae* was used (US Patent 4769324 - Ethanol production), the *S. castellii* mutant produces the enzymes á-amylase and glucoamylase which converts starch and higher sugars in the unfermentable component of the molasses substrate to a hexose sugar, thereby making it available for conversion to ethanol by *S. cerevisiae*. The mutant strain was acclimatized and able to produce amylases in a molasses containing medium. Hence, even though these are not used up in conversion process can be used as ancillary enzymes. The National Renewable Energy Laboratory produces mutant strain of *Z. mobilis*. The engineered *Z. mobilis* now produced ethanol from xylose and continuous to produce ethanol efficiently from glucose.[25]

DISCUSSION

In the present era, the knowledge of Ayurveda is validated by a contemporary research work. The diabetic population of India is increasing day by day. For the treatment of chronic disease, patients are looking for ayurvedic treatment. At the meantime, diabetic patients also adopt ayurvedic treatment for their other chronic ailments. Sandhana Kalpana, which deals with hydroalcoholic oral dosage form, has a number of formulations for various ailments in ayurvedic treasures Sandhana kalpana. However, ayurvedic doctors failed to prescribe this dosage form to the patients because of more percentage of sugar in asava-arishta. As this dosage form has a high percentage of sugar which restricts its use among diabetic population. Hence, it is the demand of time to move ahead with classical pharmaceutical method for the preparation of asava and arista and make some advancement in sandhana kalpana related to its sugar percentage so that diabetic population can also enjoy the goodness of Ayurveda.

The feedstock of carbohydrate and fermenting agent is the necessary elements for fermentation. Carbohydrates having more number of glucose molecules are considered as good elements in the fermentation process. This results in high

percentage of sugar in final product. In the process of sandhana kalpana, this rule is also followed, which is not good for diabetic patients as it increases the sugar concentration in the medicine. To solve this major problem, there is a need to be changed in feedstock of carbohydrate from other carbohydrate sources, namely, starch, wheat, potato, and cellulosic material which will produce less sugar in medicine after completion of fermentation process. Different feedstocks are given in Table 2, which are having less number of sugar molecules for fermentation which further results in least percentage of sugar in the final product. Different researchers have been done which revealed that the use of recombinant technology on different yeast strains or engineered strains is helpful in direct production of alcohol without giving any pretreatment to these substrates, namely, starchy or cellulosic feedstocks [Table 3].

As yeast, i.e., *S. cerevisiae* directly ferment the sugar molecule into alcohol, but for the fermentation of starchy feedstock and cellulosic material, fermentation process can be proceed only after applying specific treatment to these feedstocks. Different treatment required like saccharification or enzymatic hydrolysis to get free glucose molecule for the fermentation from these feedstocks only. Therefore, it is required to change the strain of yeast which can directly fermentate the starchy feedstocks. *S. castellii* and *Endomycopsis fibuligera* are some of the available strains of yeast which when used with can transform the starch directly into alcohol.[26] The engineered *Z. mobilis* now produced ethanol from xylose and continuous to produce ethanol efficiently from glucose.

CONCLUSION

Use of other carbohydrate sources such as potato tuber, banana peel, and cellulose at the place of jaggery for self-generated alcohol in sandhana kalpana will be a boost in the field of ayurvedic pharmaceutics. With the collaboration of biotechnology, engineered yeast would be used in place of Dhataki pushpa or with/without yeast *S. cescerevisiae*. These recombinant yeast strains will help to produce ethanol directly from starchy and cellulosic feedstocks without giving any pretreatment to these feedstocks. This will help to produce the asava-arishta, which will possess least or no percentage of sugar in comparison to classical method where sugar percentage present in quite large amount. Adopt to prepare asava-arista. This novel method facilitates the production of fermented liquid orals virtually free from sugar and hence provides benefits to the large segment of the population suffering from diabetes. However, care should be taken that therapeutic valve should not to be altered and it give the same benefits. Further, researchers are needed to manufacture asava-arishta from these first and second generation feedstocks for having least sugar percentage and optimum alcohol level and giving the same therapeutic effect as described in ayurvedic texts.

Table 3: Yeast strains used for fermentation in starchy and cellulosic feedstocks

Name of strains	
Starchy feedstocks	Cellulosic feedstocks
S. cescerevisiae	*Z. mobilis*
R. oryzae	*P. stipitis*
S. castellii	*E. coli*
E. fibuligera	*C. thermocellum*
T. species	
K. marxianus	
C. Kefyr	
S. kefyr	

S. cescerevisiae: Saccharomy cescerevisiae, Z. mobilis: Zymomonas mobilis, R. oryzae: Rhizopus oryzae, S. castellii: Schwanniomyces castellii, E. fibuligera: Endomycopsis fibuligera, T. species: Trichomonas species, K. marxianus: Kluyveromyces marxianus, C. Kefyr: Candida Kefyr, S. kefyr: Streptococcus kefyr, P. stipitis: Pichia stipitis, E. coli: Escherichia coli, C. thermocellum: Clostridium thermocellum

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