The study presented fermentation characteristics and microbial dynamics of primary and secondary fermentation of koozh and the process was standardised using starter culture technology. Primary fermentation study was conducted with four different grains viz., Finger millet, Pearl millet, Maize and Sorghum. The fermentation process involved various groups of microorganisms viz., aerobic mesophillic bacteria, starch hydrolysing bacteria, Enterobacter, LAB and yeasts occurring in succession. Fungal growth was not observed during the fermentation period. During initial hours, starch hydrolysing bacteria population was high compared to other bacterial groups but significant increase in their population was noticed only in finger millet and pearl millet. Concurrent to the increase in starch hydrolyser population, significant increase in reducing sugar level was observed in finger millet and pearl millet while in maize and sorghum no such change was noticed. Enterobacter was present in the initial hours of fermentation in all the grains with an increase till 10 h of fermentation. Significant rise of LAB population from 10 h of fermentation was observed in all the grains with concurrent increase in acidity. Changes in microbial profile of the grains affected sensory quality of the product. Sensory evaluation results showed that Koozh prepared with maize and sorghum was not acceptable and finger millet received high scores with respect to odour and taste. Based on the results obtained from sensory tests and considering wide usage and previous published results, finger millet was selected for secondary fermentation studies.

Secondary fermentation of finger millet was initiated by survivors of thermal treatment mainly starch hydrolysing bacteria. However, starch hydrolyser population decreased after 15 h of fermentation and LAB became the quantitatively dominant group in all the samples. Enterobacter population was found to be influenced by processing and handling conditions. Coliforms were observed during initial hours but as fermentation proceeded they reduced to undetectable levels. Consistent increase in yeast population was noticed in all the samples. Microbial composition of samples collected from home and market was similar in microbial profile. But Enterobacter population was more in market samples than the household samples exceeding the limits given by HPA guidelines. Hence to analyse the safety of the product...
Enterobacter isolates were randomly picked up from the plates and organisms were identified as Enterobacter cloacae, Enterobacter gergoviae, Citrobacter freundii, Enterobacter taylorae, Proteus mirabilis, Enterobacter aerogenes and Aeromonas sp. The results indicated the need for standardization of the fermentation process for which biochemical changes and predominant microbial strains involved in the process were studied.

Lactic acid and acetic acid were the major acids formed during fermentation. Reducing sugar content increased during 15 h and 45 h with intermittent decrease at 30 h which depicted microbial succession during fermentation. Amylase production was observed during the release of sugars. Sucrose, maltose, glucose and xylose were the sugars found during 0 h of fermentation. As the fermentation proceeded, sucrose and xylose were metabolized and at the end of fermentation only glucose and maltose were found. Ethanol production was also observed during fermentation. There was no significant change in calorific value of the food but the availability of some of the minerals such as calcium and iron significantly increased during fermentation.

Secondary fermentation of finger millet bought significant changes in concentration of bioactive compounds thus affects antioxidant property of the substrate. Significant increase in phenols and flavonoids was observed during 45 h of fermentation with concurrent rise in reducing activities of finger millet. But scavenging activities of the millet did not show any variation during secondary fermentation. The changes recorded can be attributed to the activities of microorganism by comparing to controls of sterilized samples. Ferulic acid and gallic acid were identified as the main phenol groups in the fermented extracts. Vanillic acid was detected in 30 h of fermentation. Some of the catechin derivates namely gallocatechin, epicatechin were also found in 15 h and 30 h fermented samples.

As starch being major substrate for fermenting microbes the fermentation process influenced starch fractions concentration with respect to time. Resistant starch content increased during 15 h of fermentation due to gelatinisation of starch after which resistant starch content decreased due to microbial activities. Simultaneously, digestible starch content increased significantly from 30 h of fermentation.
Despite the improvement in sensorial qualities and nutraceutical properties there were safety and quality concerns in natural fermentation which resulted in variations in product quality and decreased shelf life of the product. Hence for controlled fermentation identification of predominant strains of starch hydrolysing bacteria and LAB occurring during fermentation process was carried out.

Starch hydrolysing bacterial isolates from primary fermentation was identified as *Bacillus cereus*, *Bacillus subtilis* and *Bacillus* sp. Representative LAB isolates collected from primary fermentation were identified as *Weisella confusa*, *Weisella cibaria* and *Weisella* sp. Other than *Weisella* species *Lactobacillus* genera was also found at lower proportions during primary fermentation.

*Bacillus cereus*, *Bacillus megatherium*, *Bacillus subtilis*, *Bacillus arybhattai*, *Bacillus* sp., *Staphylococcus aureus* and *Acinetobacter baumanii* were the predominant starch hydrolysing bacterial isolates identified from secondary fermented substrate. Majority of the LAB isolates of secondary fermentation belonged to *Enterococcus*, *Pediococcus*, *Weisella* and *Bacillus*. Representative isolates were identified as *Enterococcus durans*, *Enterococcus hirae*, *Enterococcus lactis*, *Enterococcus faecalis*, *Enterococcus faecium* and *Enterococcus* sp., *Bacillus coagulans*, *Pediococcus pentosaceus* and *Weisella confusa*. Studies were conducted to assess use of these strains as starter cultures.

All LAB isolates were subjected to series of tests which are prerequisites for starter culture selection. The tests included growth characteristics, acid production, bacteriocin production, enzyme activities and probiotic properties. All the isolates showed rapid growth in MRS broth except *Bacillus coagulans*. Of all the isolates *Weisella confusa* showed more acid production. *Pediococcus pentosaceus* exhibited broad spectrum antimicrobial activity and also showed weak amylolytic activity. All the isolates exhibited weak proteolytic activity. *Enterococcus durans* and *Pediococcus pentosaceus* showed potential probiotic properties such as tolerance to acidic conditions and bile salts. *Bacillus coagulans* showed good adhesion properties. All isolates were negative for biogenic amine production with varied susceptibility to antibiotics.

Depending on the functional properties and antibiotic susceptibility profile *Pediococcus pentosaceus* and *Enterococcus durans* were selected as starter cultures.
for controlled fermentation. Starter cultures were used singly and in combination and the results of starter treatments were compared with uninoculated control. Starter culture treatments reached desirable pH quicker compared to control samples. Of the starter culture treatments, dual culture inoculated samples took 6-8 h to reach desirable pH. Microbial analysis showed that pathogen count was minimum in starter culture inoculated samples compared to control samples. Though safety was ensured in all starter culture inoculated samples, shelf life of the product varied among the treatments which was analysed by sensory evaluation test and product viscosity. Of all the treatments *Pediococcus pentosaceus* inoculated samples had shelf life of 15 days which is three times higher than control samples. With respect to dual culture treatments samples deteriorated close to control. Thus *Pediococcus pentosaceus* as single starter was able to improve the shelf life of the traditional food compared to control. Also as a positive note, the calorific value of the food remained same even after 15 days of storage in *Pediococcus pentosaceus* inoculated samples.