In Asia amylolytic starter culture prepared from the growth of filamentous fungi and yeasts on raw or cooked cereals are more commonly used (Haard et al. 1999; Tamang 2016). The use of mixed amylolytic starters might have its origins during the time of Euchok, the daughter of the legendary king of Woo of China, known as the Goddess of rice-wine in Chinese culture in 4000 BC (Lee 1984; Lee and Kim 2016). The first documentation of chu, a Chinese amylolytic starter, is very similar to marcha of the Himalayas (Tamang 2010a), was reported in Shu-Ching document written during Chou dynasty (1121-256 BC), in which it is reported that chu is essential for making alcoholic beverages (Haard et al. 1999). According to the text Chhi Min Yao Shu, written by Chia Ssu-Hsieh of Late Wei kingdom between 533 and 544 AD, many methods of preparation of chu were described (Yoon 1993; Huang 2000). The use of chu, a Chinese amylolytic starter for rice-based alcoholic beverage production was commonly practiced in the Spring and Fall and Warrior Periods of China during 6th to 7th centuries B.C. and the beginning of the Three Nations’ Periods in Korea during 1st century BC to 2nd century AD (Lee 1995). It might have transferred from Korea to Japan in the 3rd century AD according to Kojiki, or Chin, whose memorial document is kept in a shrine at Matsuo or Matsunoo, Taisha, Kyoto, Japan (Lee 1995). The process of cereal alcohol fermentation using mold starters was well established in the year of 1000 BC and forty three different types of cereal wines and beers were described with detailed processing procedures in Chhi Min Yao Shu (Haard et al. 1999). According to this document chu was prepared from barley, rice and wheat (Yoon 1993). Ten different types of chu were described in Chhi Min Yao Shu (Yoon 1993; Huang 2000), all of which were used for the fermentation of alcoholic beverages in China. Cake type ping-chu is similar to nuruk of Korea, and granular type san-chu is similar to koji of Japanese (Yoon 1993). According to Yokotsuka
(1985), *chu* Chinese starter may either be white probably due to *Rhizopus* and *Mucor* or yellow (*huang*) possibly due to *Aspergillus oryzae*. *Nu-chu* is prepared by using cooked rice, which is further shaped into a cake and then cultured with molds (Yokotsuka 1985). Wheat *chu* starter originated from the Northern of China and the Korean Peninsular, while rice *chu* starter originated in the South China (Haard et al. 1999). The word *ragi* of Indonesian was first time noted on an ancient inscription called the Kembang Arum, near Yogyakarta in Java of Indonesia around 903 AD (Astuli 1999). In Asia production technique of ethnic starter cultures to make alcoholic beverages is usually kept secret and the indigenous knowledge of processing is not easily passed on. However, the protected hereditary right of making ethnic mixed starters is passed to daughter by mothers, and she carries the indigenous knowledge to in-laws after marriage. Traditionally preparation of ethnic mixed starters is done exclusively by women, *marcha* is prepared by the Limboo and Rai castes of the Nepali, *ragi* by Indonesian, *loog pang* by ethnic Thai, *nuruk* by ethnic Koreans, and *bubod* by the Filipino (Tamang 2010a). Asian ethnic people traditionally prepare three major types of mixed amylolytic starters to convert cereal starch to sugars and subsequently to alcohol and organic acids are practiced in Asia (Steinkraus 1983; Hesseltine et al. 1988; Fleet 1998; Tamang and Fleet 2009).

**Type I**: Traditional practice of sub-culturing by back-sloping for preservation of essential native microbiota consisting of consortia of yeasts, molds and bacteria, in the form of dry, flattened, or round balls amylolytic starters (related to conversion of starch to sugar), for alcoholic beverages production in South-East Asia including the Himalayan regions of India, Nepal, Bhutan, and China is the worth wisdom of the ethnic people for centuries (Tamang 2010a). Consortia of mycelia or filamentous molds, amylolytic and alcohol-producing yeasts and lactic acid bacteria
(LAB) with rice or wheat as the base in the form of dry, round to flattened balls of various sizes. The starter is inoculated with previous starter. This mixed flora is allowed to develop for a short time, then dried, and used to make either alcohol or fermented foods from starchy materials. Ethnic starters have different vernacular names such as marcha in India and Nepal, ragi in Indonesia, bubod in Philippines, chiw/chiu in China and Taiwan, loogpang in Thailand, nuruk in Korea, and men in Vietnam (Tamang et al. 1996; Dung et al. 2007), which are used as starters for a number of fermentations based on rice and cassava or other cereals in Asia. There are several major types of ethnic amylolytic mixed starters in dry and ball-flatted discs shaped sold in local markets in India, Nepal, Bhutan, China, Thailand, Myanmar, Cambodia, Laos, Malaysia, Indonesia, Korea, Japan, Singapore, Taiwan, etc.

Calmette (1892) was the first to report the presence of several wild yeast species accompanied by Amylomyces, Mucor, Aspergillus and 30 different bacteria in starters used in China.

**Type II:** A combination of Aspergillus oryzae and A. sojae are used in the form of starter called koji in Japan to produce alcoholic beverages including saké. Koji also produces amylases that convert starch to fermentable sugars, which are then used for the second stage yeast fermentation to make non-alcoholic fermented soybean product called miso and shoyu, while proteases are formed to break down the soybean protein.

**Type III:** Whole-wheat flour with its associated flora is moistened and made into large compact cakes, which are incubated to select certain desirable microorganisms. The cakes are used to inoculate large masses of starchy material, which is then fermented to produce alcohol. This type of starter contains yeasts and filamentous molds, and is mostly used in China for alcohol production. A list of
common traditionally prepared amylolytic starters and their alcoholic products of Asia is shown in Table A.

<table>
<thead>
<tr>
<th>Starter Culture</th>
<th>Substrate</th>
<th>Nature and use</th>
<th>Area</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amou/pe rokkushi</td>
<td>Rice and wild herbs</td>
<td>To ferment rice into alcoholic beverage-jou</td>
<td>Bodoland, Assam, India</td>
<td>Das et al. (2017)</td>
</tr>
<tr>
<td>Bakhar</td>
<td>Rice flour, ginger</td>
<td>To ferment rice into alcoholic beverage-pachwai</td>
<td>India (Himachal Pradesh)</td>
<td>Hutchinson and Ram Ayyar (1925)</td>
</tr>
<tr>
<td>Balam</td>
<td>Roasted wheat flour and spices</td>
<td>To ferment alcoholic beverage-jaan</td>
<td>India (Uttarakhand)</td>
<td>Roy et al. (2004)</td>
</tr>
<tr>
<td>Banh men</td>
<td>Rice, wild herbs, spices</td>
<td>To ferment rice/maize/cassava into alcoholic beverage-ruou nep chan</td>
<td>Vietnam</td>
<td>Dung et al. (2007)</td>
</tr>
<tr>
<td>Bubod</td>
<td>Rice, wild herbs</td>
<td>To ferment sugar cane into alcoholic beverage-basi</td>
<td>The Philippines</td>
<td>Hesseltine and Kurztman (1990)</td>
</tr>
<tr>
<td>Chiu/chu, yao qu and hong qu</td>
<td>Rice, wild herbs</td>
<td>To ferment rice into alcoholic beverage-Hong qu. Glutinous rice wine, Shaoxing rice wine and Shandong Jimo millet wine.</td>
<td>China and Taiwan</td>
<td>Lv et al. (2013)</td>
</tr>
<tr>
<td>Dhehlí</td>
<td>Herbal mixture of 36 herbs and roasted barley flour</td>
<td>Starter to ferment alcoholic beverage-sura</td>
<td>India (Himachal Pradesh)</td>
<td>Thakur et al. (2004)</td>
</tr>
<tr>
<td>daqu</td>
<td>Glutinous rice, wild herbs</td>
<td>Starter to ferment alcoholic beverage-fen</td>
<td>China</td>
<td>(Chen et al. 2014).</td>
</tr>
<tr>
<td>Hamei</td>
<td>Rice, wild herbs</td>
<td>To ferment rice into alcoholic beverages-atingba</td>
<td>India (Manipur)</td>
<td>Jeyaram et al. (2009) and Singh and Singh, 2006.</td>
</tr>
<tr>
<td>Humao</td>
<td>Rice, barks of wild plants</td>
<td>Dry, flat, cake-like starter for judima production</td>
<td>India (Assam)</td>
<td>Chakrabarty et al. (2014)</td>
</tr>
<tr>
<td>Ipoh/Siye</td>
<td>Rice and powder of seeds and bark</td>
<td>Starter to ferment alcoholic beverages - apong and ennog</td>
<td>India (Arunachal Pradesh)</td>
<td>Tiwari and Mahanta (2007)</td>
</tr>
<tr>
<td>Plant Name</td>
<td>Source</td>
<td>Description</td>
<td>Country, Region</td>
<td>Reference</td>
</tr>
<tr>
<td>--------------------</td>
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<td>-----------------------------------------------------------------------------</td>
<td>------------------------------------------------------</td>
<td>------------------------------------</td>
</tr>
<tr>
<td>Keem</td>
<td>Wheat; plants</td>
<td>Starter to ferment alcoholic beverages - soor</td>
<td>India (Himachal Pradesh)</td>
<td>Rana et al. (2004)</td>
</tr>
<tr>
<td>Khekhrii</td>
<td>Germinated rice</td>
<td>Starter to ferment alcoholic beverages - zutho/zuchu</td>
<td>India (Nagaland)</td>
<td>Jamir and Rao (1990), Jamir and Deb (2014)</td>
</tr>
<tr>
<td>loogpang</td>
<td>Rice, wild herbs</td>
<td>Khao-maak, krachae, nam khoa, ou, sato</td>
<td>Thailand</td>
<td>Vachanavinh et al. (1994)</td>
</tr>
<tr>
<td>Maae/domba e/buh/puh</td>
<td>Rice, Spices, herbs</td>
<td>To ferment rice into alcoholic beverage-sombai.</td>
<td>Cambodia</td>
<td>Chay et al. (2017) and Chim et al. (2015)</td>
</tr>
<tr>
<td>Medombae</td>
<td>Rice, Spices, herbs</td>
<td>To ferment rice into alcoholic beverage-sombai.</td>
<td>Cambodia</td>
<td>Savitri and Bhalla (2007)</td>
</tr>
<tr>
<td>Malera/treh</td>
<td>Wheat flour</td>
<td>Starter to ferment bhatooru/chilra</td>
<td>India (Himachal Pradesh)</td>
<td>Deori et al. (2007)</td>
</tr>
<tr>
<td>Mod pitha</td>
<td>Rice and 31 plant materials</td>
<td>Starter to ferment alcoholic beverages - sujen</td>
<td>India (Assam and Arunachal Pradesh)</td>
<td></td>
</tr>
<tr>
<td>Marcha</td>
<td>Rice, wild herbs, spices</td>
<td>Dry, mixed starter to ferment alcoholic beverages</td>
<td>India (Darjeeling hills, Sikkim, North East)</td>
<td>Tamang and Sarkar (1995)</td>
</tr>
<tr>
<td>Nuruk</td>
<td>Rice, herbs</td>
<td>Takju, sojo, yakju</td>
<td>Korea</td>
<td>Jung et al. (2012)</td>
</tr>
<tr>
<td>Pham/phab</td>
<td>Rice and leaves of Solanum khasianum</td>
<td>Starter to ferment alcoholic beverages - themsing, chhang, arrak, kinnauri</td>
<td>India (Arunachal Pradesh, Jammu and Kashmir, Himachal Pradesh)</td>
<td>Singh et al. (2007), Angmo and Bhalla (2014)</td>
</tr>
<tr>
<td>Ragi</td>
<td>Rice, herbs</td>
<td>To ferment cassava/rice into mild-alcoholic and sweet beverage- tapé-kekan, brem</td>
<td>Indonesia</td>
<td>Surono (2016)</td>
</tr>
<tr>
<td>Ranu dabai</td>
<td>Rice, herbs</td>
<td>Starter to ferment alcoholic beverages- jhara or haria</td>
<td>India (West Bengal)</td>
<td>Ghosh and Das (2004)</td>
</tr>
<tr>
<td>Ranu goti</td>
<td>Rice, herbs</td>
<td>Starter to ferment alcoholic beverages - handia and mahua</td>
<td>India (Central India)</td>
<td>Kumar and Rao (2007)</td>
</tr>
</tbody>
</table>
AMYLOLYTIC STARTERS

Amoulperokkushi

Amoulperokkushi is amyloolytic starters of Assam for preparation of rice-based alcoholic beverage in Assam, by the Deori and Bodo communities, respectively (Das et al. 2017). They identified the amyloolytic fungi, based on the sequencing of their internal transcribed spacer (ITS) regions, as Amylomyces rouxii and Rhizopus oryzae, and both the strains showed the ability to breakdown and saccharify starch (Polysaccharides). The glucoamylase activity was considerably high in A. rouxii (14.92 mmol/min) as compared to R. oryzae (1.41 mmol/min), whereas a-amylase activity was observed to be closely related, i.e. 7.02 and 6.09 unit/mL, respectively. They used SDS-PAGE to determine molecular size of the glucoamylase enzymes revealed the production of two distinct units of 59 kDa and 31 kDa by A. rouxii, and one unit of 72 kDa by R. oryzae (Das et al. 2017).

Bakhar

Bakhar is a starter culture used to make pachwai, rice wine in eastern part of India and contains Rhizopus sp., Mucor sp., and at least one species of yeast (Hutchinson and Ram-Ayyar 1925). Ginger and other plant materials are dried, ground and added to rice flour. Water is added to make a thick paste and a small round cake of 1.0-1.5 cm in diameter are formed and inoculated with powdered
cakes from previous batches. The cakes are then wrapped in leaves, allowed to ferment for 3 days and then sun-dried (Hutchinson and Ram Ayyar 1925). Ray (1906) reported the presence of *Saccharomyces cerevisiae* in *bakhar*.

**Balam**

*Balam* is traditionally prepared wheat based amylolytic starter of Uttaranchal used for preparation of *jann*, during the preparation of *balam* first the raw wheat is washed and sun dried, then this is ground into flour and then it is roasted over fire and removed before it becomes turns brown in color. The roasted wheat flour is then mixed properly with various plants spices like *Cinnamomum zeylanicum*, *elachi* (*Amomum subulatum*), *Piper longum* (*kalimirch*), seeds of *Ficus religiosa* (*papal*) and leaves of wild chilies (*mirchi-ghash*). In this mixture, old powder of *balam* is also added. The addition of old *balam* starter powder is a must, without addition of this old starter production of fresh *balam* is not possible. The whole mixture, which is prepared, is now thoroughly mixed with the required amount of water and a thick paste is prepared. This prepared mass is then pressed between palms to make *balam* balls of the different required size. These different sized wet balls are dried in shade and then stored for future use for a long period of time (Roy et al. 2004).

**Banh men**

*Banh men/men* is the traditionally prepared amylolytic starers of Vietnam (Dung et al. 2007). The diversity of yeasts (*Candida tropicalis, Clavispora lusitaniae, Pichia anomala, Pichia ranongensis, Saccharomycopsis fibuligera, Sacch. cerevisiae, Issatchenkia* sp.); filamentous molds (*Absidia corymbifera, Absidia yeasts*).
Amylomyces rouxii, Botryobasidium subcoronatum, Rhizopus oryzae, Rhi. microsporus, Xeromyces bisporus); LAB (Ped. pentosaceus, Lb. plantarum, Lb. brevis, Weissella confusa, Weissella paramesenteroides); amylase-producing bacilli (Bacillus subtilis, B. circulans, B. amyloliquefaciens, B. sporothermodurans); and acetic acid bacteria (Acetobacter orientalis, A. pasteurianus) were present in men, a starter culture of Vietnam (Dung et al. 2006, 2007; Thanh et al. 2008). The diversity of fungi and bacteria associated with Vietnamese ethnic amylolytic starters, banh men was studied by PCR-DGGE. The fungal population of the banh men was consistent with little variation among samples. It mainly consisted of amylase producers (Rhizopus oryzae, R. microsporus, Absidia corymbifera, Amylomyces sp., Saccharomycopsis fibuligera), ethanol producers (Saccharomyces cerevisiae, Issatchenkia sp., Pichia anomala, Candida tropicalis, P. ranongensis, Clavispora lusitaniae), and opportunistic contaminants (Xeromyces bisporus, Botryobasidium subcoronatum). The bacterial population of starters was highly variable in species composition and dominated by lactic acid bacteria (LAB). The most frequent LAB were, Lactobacillus plantarum, L. brevis, Pediococcus pentosaceus, Weissella confusa and W. paramesenteroides. Species of amylase-producing Bacillus (Bacillus subtilis, B. circulans, B. amyloliquefaciens, B. sporothermodurans), acetic acid bacteria (Acetobacter orientalis, A. pasteurianus) and environment contaminants/plant pathogens (Burkholderia ubonensis, Ralstonia solanacearum, Pelomonas puraquae) (Dung et al. 2006; Thanh et al. 2008).
**Bubod**

*Bubod* is used as a starter in the Philippines (Tanimura et al. 1977; Elegado 2016). Rice and ginger are powdered, and mixed thoroughly with enough water to have a consistency that permits rolling the material into a ball and flattening it. The discs are coated with 1-3 month old *bubod* and incubated in rice straw for 36 h at room temperature and sun-dried. Tanimura et al. (1977) reported that *Mucor, Rhizopus* and filamentous yeasts in *bubod*. Kozaki and Uchimura (1990) reported the presence of *Mucor circinelloides, M. grisecyanus, Rhizopus cohnii, Saccharomyces cerevisiae* and *Saccharomycopsis fibuligera* in *bubod*. Sanchez (1986) reported that the molds present in *bubod* ranged from $10^3$ to $10^5$ cfu/g, yeasts from $10^7$ to $10^8$ cfu/g, and lactic acid bacteria from $10^5$ to $10^7$ cfu/g. Hesseltine and Kurtzman (1990) reported that *Saccharomycopsis fibuligera* was dominant in *bubod*. Lim et al. (2006) reported *Sacchromycopsis fibuligera, Saccharomyces cerevisiae, Hansunela anomala* from Philippine ethnic amylolytic starter, *bubod* by Genetic DNA Fingerprinting (PCR-RAPD) of yeast isolates.

**Chiu-yueh**

*Chiu-yueh* or *peh-yueh* is a gray-white ball-like starter for *lao-chao*, fermented rice product of China. Wei and Jong (1983) isolated yeasts and moulds from *chiu-yueh* and tested the ability of these microorganisms to convert steamed glutinous rice into a good quality *lao-chao*.

**Chou or Chu**

*Chou/Chu* is ball, cake or brick (20×22×4.5 cm) shaped and made from moistened raw rice, wheat, sorghum or barley flour (Campbell-Platt 1987). The principal
amylolytic enzyme producers of chu are Rhizopus and Mucor (Yokotsuka 1991). Microbiota in wheat-based chu were Rhizopus japonicus, R. hangchon, R. chinensis, Absidia, Mucor, Monilia, Aspergillus, Lactobacillus and Acetobacter (Otani 1973; Iizuka 1979).

**Dhehli**

Herbal mix or dhehli preparation is an annual community effort, in which elderly people go to forests on the 20th day of Bhadrapada month (usually 5 or 6th September) and collect approximately 36 fresh herbs (Thakur et al. 2004). Some of the important herbs used in dhehli preparation are Pistacia integerrima (kkakar shinga), Solanum xanthocarpum (katari), Clitoria ternatea (kkayal), Aegel marmelos (bhel), Viola cinerea (banaksa), Cannabis sativa (bhang), Trachyspermum copticum (ajwain), Micromeria biflora (chharbara), Spiranthes australis (bakarshingha), Saussurea sp. (bbacha), Bupleurum lanceolatum (nimla), Drosera lunata (oshtori), Salvia sp. (kotugha), Arisaema helleborifolium (chidi ri chun), Fragaria sp. (dudlukori). The collected herbs are crushed in stone with a large conical cavity (ukhal) using a wooden bar (mussal) and the extract as well as the plant biomass are added in to the flour of roasted barley and are roughly kneaded. This is put in to a wooden mould, to give the shape of a brick and dried, is called dhehli (Thakur et al. 2004; Savitri and Bhalla 2007).

**Daqu**

Study of daqu Chinese amylolytic starter revealed the presence of filamentous fungal community associated with Chinese wine making process (Chen et al. 2014). *Paecilomyces variotii, Aspergillus oryzae* and *Asp. terreus* were reported
from this starter (Chen et al. 2014). The Next generation sequencing (NGS) results of amylolytic starter *daqu* revealed the microbial community including *Saccharomycetaceae* (60%), *Saccharomycopsidaceae* (29%), *Saccharomycodaceae* (2%), *Dipodascaceae* (1%), *Trichocomaceae* (<1%), *Candida* (7%), and *Pleosporaceae* (<1%) which play an important role during fermentation of *fen*, Chinese rice wine (Li et al. 2011).

**Hamei**

*Hamei* is an ethnic amylolytic mixed dry, round to flattened starter of Manipur in India which is very similar to *marcha* (Tamang 2010a). *Hamei* an ethnic amylolytic starter of Manipur is used for the preparation of alcoholic beverage from glutinous rice is very interesting because of its unique flavor and aroma. Yeast communities of *hamei* were identified by phenotypic (biochemical characterization) and molecular tools such as restriction digestion pattern generated from PCR amplified internal transcribed spacer region along with 5.8S rRNA gene (ITS1-5.8S-ITS2) which included yeasts *Saccharomyces cerevisiae*, *Pichia anomala*, *Trichosporon* sp., *Candida tropicalis*, *Pichia guilliermondii*, *Candida parapsilosis*, *Torulaspora delbrueckii*, *Pichia fabianii* and *Candida Montana* (Jeyaram et al. 2008). The genetic diversity of industrially important *S. cerevisiae* group isolated from *hamei* was investigated using Pulsed Field Gel Electrophoresis (PFGE) (Tamang et al. 2007; Jeyaram et al. 2008).
Huamo

Huamo is traditionally prepared rice based ethnic amylolytic starter of Assam and is commonly used for the preparation or fermentation of *judima* (Tamang 2010a). *Humao* is prepared by using locally available glutinous rice, bark, leaves and roots of wild plants (Chakrabarty et al. 2014). During the preparation of *huamo* rice is first washed and powdered in a wooden *okhari along* with the bark, leaves and roots of wild plants parts and few old *humao* starters are mixed properly with clean water to make paste. Then the paste is used to make different sized round to flat, cake-like starters on mat or carpet, fermented for 1-2 days and sun-dried and then stored at room temperature for further use.

Hongqu/yaoqu

*Hongqu* and *yaoqu* are two popular traditionally prepared amylolytic starters of China (Lv et al. 2012). These traditionally prepared amylolytic starters investigated using a combination of culture-dependent and culture-independent molecular methods. using restriction fragment length polymorphism (PCR-RFLP) analysis of the internal transcribed spacer region ITS1-5.8S-ITS2 and sequencing of the D1/D2 domain of the 26S rRNA gene and generated 12 different genera of yeasts *Pichia, Saccharomyces, Candida, Saccharomycopsis Cryptococcus, Sporobolomyces, Rhodosporidium* and *Rhodotorula* (Lv et al. 2012). On the other hand, the yeast diversity associated with these starters was also investigated through culture-independent method using PCR-DGGE patterns and sequencing of the DNA bands and found almost the same as that of culture-dependent methodology (Lv et al. 2013). The PCR-DGGE fingerprints revealed that *Rhizopus oryzae, R. microsporus* and *Aspergillus* sp. were the most frequent
species in yaoqu, while Monascus sp. dominated in hongqu and non-Saccharomyces yeasts (Saccharomycopsis fibuligera, Pichia guilliermondii and Pichia farinose) were also detected in some starter samples (Lv et al. 2012). Xu et al. (2012) reported the bacterial DGGE profile targeting the V3 region of the 16S rRNA gene showed that the bacterial composition of starters dominated by Bacillus sp., including B. ginsengihumi, B. megaterium or B. aryabhattai, B. subtilis, B. methylotrophicus and B. amyloliquefacien (Xu et al. 2012). Lactic acid bacteria including Weissella paramesenteroides, Pediococcus pentosaceus and Pediococcus acidilactici were also detected in some fermentation starters (Xu et al. 2012). Rhizopus oryzae, R. microsporus and Aspergillus sp. were the most frequent species in yao qu, while Monascus sp. dominated in hong qu (Xu et al. 2012).

**Ipoh**

Ipoh is traditionally fermented amylolytic starter of Arunachal Pradesh used for the traditional fermentation of apong and ennog, popular mild alcoholic beverages (Tamang 2010a). It is prepared through various processes of washing, drying and grinding of glutinous rice into fine powder and mixing powder of leaves, bark, seeds of locally available plant species, viz. Veronia cinerea and Clerodendron viscosum. Then this mixture is mixed properly in a large container (dekchi) and made into paste by using previously stored rice water, spread on clean bamboo mats and made into circular, disc shaped small cake like or biscuit shaped. The cakes are then carefully kept to dry out completely either in the attic above the fireplace of traditional houses or kept in a cool dry place for 4–5 days for fermentation and sun-dried, after drying it is stored for further use. The major
microorganisms involved in *ipoh* are yeast populations (Tiwari and Mahanta 2007).

**Khekhri**

It a traditionally prepared ethnic unique type starter of Nagaland used to prepare local alcoholic beverage *zutho* (Tamang 2010a). During the traditional preparation, unhulled glutinous rice is washed, soaked into water for 2-5 days, kept and covered with *Khreihenyii* leaves and allowed to germinate for 3-4 days in summer and 5-6 days in winter. After partial germination when the rice sprout is about half inch in length, the sprouted rice is exposed to sun for drying and powdered and again sun dried and stored for further use (Jamir and Rao 1990; Jamir and Deb 2014).

**Koji**

*Koji* is mold-culture and is prepared from steamed-cooked cereal (Kitamura 2016). The substrate is usually rice, or sometimes steamed legume beans. The steamed substrate is spread on trays usually made of bamboo strips to depth of 5-7 cm, which are stacked with gaps of about 10 cm in between to allow air circulation. It is followed by inoculation with 0.1 % mold spores, *tane-koji* and incubated at 23-25° C. The rise in temperature due to the growth of mould is kept within the range 35-45° C by stirring and turning *koji* top to bottom on trays at about 20 h and 40 h, normally fermented for 3 days, when mould mycelium spread throughout mass, and before sporulation (Lotong 1985). The mould used is *Aspergillus oryzae*, which is used for starch saccharification in *saké* manufacture (Inoue et al. 1992; Kitamura 2016). Since *koji* is not cultivated in a closed system,
koji is a mixture of several microorganisms. At an early stage of cultivation, yeast grows on steamed rice grain and after that, about 20 h after inoculation of seed koji, koji mold begins to grow. Koji usually contains $10^2$/g saké yeast, $10^2$ to $10^5$ g film-foaming yeasts, $10^2$/g lactic acid bacteria, $10^4$ to $10^6$ /g micrococi, $10^7$/g bacilli, etc. Kodama and Yoshizawa (1977) studied the biochemical changes occurring in koji and found the increase of reducing sugar from 0.2 % to 21.4 %.

Tanaka (1982) studied enzyme activity of steamed or unsteamed glutinous rice-koji inoculated with Aspergillus oryzae and Rhizopus javanicus and found that α-amylase was 1527 U/g in Aspergillus and 100 U/g in Rhizopus in steamed rice koji, whereas 1255 U/g and 100 U/g in unsteamed rice koji, respectively. A combination of A. oryzae and A. sojae is used in koji in Japan to produce alcoholic beverages including saké (Zhu and Trampe 2013). Koji (Chinese chu, shi, or qu) also produces amylases that convert starch to fermentable sugars, which are then used for the second stage yeast fermentation to make non-alcoholic fermented soybean miso and shoyu (Sugawara 2010). A. awamori, A. kawachii, A. oryzae, A. shirousamii, and A. sojae have been widely used as the starter in preparation of koji for production of miso, saké, shoyu, shochu Suganuma et al. (2007).

**Keem**

Keem is traditionally prepared barley based ethnic amylolytic starter of Himachal Pradesh and is commonly used for the preparation of soor which is commonly consumed as mild alcoholic beverages during various occasions (Rana et al. 2004). During traditional preparation chopped fresh twigs of Cannabis sativa (8 kg), 5 kg leaves of Sapindus mukorossi and 10-15 kg in total of different plant
species are dried in the shade for few days and then powdered, mixed properly with about 50 kg of barley flour. To the desired quantity of above dry mixture is added a sufficient quantity of Jayaras (a compound prepared by keeping finely cleaved leaves and tender parts of \textit{(Dicliptera roxburghiana, Zanthoxylum armatum, Leucas lanata and Melia azedarach)}, in a bigger vessel for overnight night and dough in to a round, circular cake of about 1-2 kg weight. Many oval-shaped cakes are prepared and kept on plant bed (\textit{sathar}) made up of with 15 different tender shoots of \textit{Pinus roxburghii} and \textit{Cannabis sativa} alternately between the cakes incubated in a closed room. The prepared starters are allowed to remain undisturbed for 24 days. On 25th day of incubation, the room is opened and the cake is placed upside down and allowed them to remain there for another 12 days for fermentation. Cakes of \textit{keem} are now taken out, sun-dried, and are used preparation of local alcoholic beverages known as \textit{soor} (Rana et al. 2004).

\textbf{Loogpang}

\textit{Loogpang} is the starter commonly used in Thailand to prepare alcoholic drink and vinegar (Vachanavinich et al. 1994; Krusong 2014). In \textit{loogpang}, organisms are grown on bran (Steinkraus 1996). The main ingredient of this starter is rice flour with the addition of different type of spices and microorganisms. The microorganisms are originated from the inoculum or surrounding place of preparation of previous batch (Vachanavinich et al. 1994). Pichyangkura and Kulprecha (1977) found that the molds \textit{Amylomyces}, \textit{Rhizopus}, \textit{Aspergillus}, \textit{Mucor}, and \textit{Absidia} in \textit{loogpang}. Dhamcharee (1982) showed that the molds present in \textit{loogpang} from different places in Thailand were \textit{Rhizopus}, \textit{Mucor}, \textit{Amylomyces}, \textit{Penicillium}, and \textit{Aspergillus}, and the main yeast genera were
*Endomycopsis* (*Saccharomycopsis*), *Hansenula*, and *Saccharomyces*. Sukhumavasi et al. (1975) isolated a strain of *Endomycopsis* (*Saccharomycopsis*) *fibuligera* from *loogpang* with high glucoamylase activity. Uchimura et al. (1991) reported the presence of *Saccharomycopsis fibuligera* and *Pediococcus* sp. in *loog-pang*. Most studies found *Saccharomycopsis fibuligera* as common yeast in *Loog-Pang* (Limtong et al. 2002). Saelim et al. (2008) reported the Saccharification of cassava starch by *Saccharomycopsis fibuligera* isolated from *Loog-Pang*. Kanlayakrit et al. (1989) Kanlayakrit and Booranasawettatham (2004, 2005) reported that *Aspergillus*, *Penicillium*, *Rhizopus* and *Mucor* were dominant fungi found in *Loongpang*.

**Mod pitha**

*Mod pitha* is traditionally fermented amylolytic starter of Assam and Arunachal Pradesh used for the traditional preparation of *sujen* which is consumed as mild alcoholic beverages (Deori et al. 2007). For the traditional preparation of *mod pitha* glutinous rice grains (*saol*), a handful each of cleaned leaves, fronds, barks, roots and bulb of the various plant parts are put in a round bamboo trey (*saloni*) and exposed for sun drying for a day. The rice grains (*saol*, 4-5kg) is soaked in water for 2 h, cleaned and mixed with the dried plant parts and grounded in a wooden grinder (*dheki*). The grounded rice powder is taken out, sieved in a round bamboo trey (*saloni*) and the coarse part is returned to the wooden grinder (*dheki*) for grinding and this process is continued until a fine powder is obtained. Old *mod pitha* (2 to 3) are added to the mass during grinding. Powdered glutinous rice is put into a utensil (*soriya*), then water is added to make a sticky paste and small round to flattened cakes (2-3 cm in diameter and 0.1 to .04 cm in thickness) are
prepared. Rice cakes are then placed on clean, dry paddy husk spread on a round bamboo trey (kula) and again covered with paddy husks. A round bamboo bucket is then kept on a dhua sang tied about 1 m above a fireplace in the traditional house kitchen for drying. This procedure of drying the yeasts cake continues for a couple of weeks until this pitha becomes harder. Pitha is now ready for use in sujen fermentation. Mod pitha can be stored traditionally for 2-4 months and can be used for the traditional fermentation to make alcoholic beverage sujen (Deori et al. 2007).

**Malera/treh**

Malera/treh is traditionally prepared wheat flour based ethnic amylolytic starter of Himachal Pradesh and is commonly used for the preparation or fermentation of bhatooru/chilra which is commonly consumed as staple diet in rural parts of Himachal Pradesh during various occasions (Tamang et al. 2016b). These are prepared with wheat/buckwheat flour dough or slurry fermented with the addition of malera which mainly consists of lactic acid bacteria and yeasts (Savitri and Bhalla 2007).

**Medombae**

There are different types of traditional ethnic amylolytic starters found in Cambodia are medombae, buh, praa, mesraa, dombae, krow, paeng and poo (Yamamoto 2016). The starter culture for rice fermentation is known as medombae in Cambodia. Spices, herbs, and a sweetener are ingredients commonly added also for dried starter preparation. Water is also added to the mixture and the previous starter was used as a source of inoculum at the rate of 1
to 2%. After mixing thoroughly, the mixture is being shaped into balls manually and placed on layers of rice husks or dried rice straw for 3 days at room temperature, sun-dried, and used as a starter for the production of alcoholic beverages such as rice wine (Sombai). Cultural morphological and biochemical identification studied revealed that the isolated representative mold strains were as *Mucor* sp. and *Rhizopus oryzae* and yeasts *Candida tropicalis* and *Saccharomyces cerevisiae* reported by Chay et al. (2017) and Chim et al. (2015).

**Mana**

*Mana* is a granular type starter prepared from wheat flakes in Nepal (Tamang 2010a). During its production, wheat grains are soaked in water overnight, steamed for 30 min and is transferred to a bamboo basket, drained and grounded into lump. The floor is cleaned, straw is spread on ground, and wheat lump is placed over it, covered with paddy straw or straw mat, and fermented for 6-7 days. After 7 days, green mold appears on the wheat grains and is dried in the sun to get *mana* and stored. *Mana* contains $10^6$ cfu/g of mucorales (*Rhizopus* sp.), $10^7$ cfu/g of aspergilla (*Aspergillus oryzae*), $10^3$ cfu/g of yeasts and $10^5$ cfu/g of LAB (Nikkuni et al. 1996; Shrestha et al. 2002).

**Manapu**

*Manapu* is an ethnic amylolytic starter of Nepal similar to *marcha*, which is prepared from rice flour and millets in Nepal (Tamang 2010a). Rice or millet is milled to get flour, and is mixed with 20% old *manapu*, 5% *manawasha* (white flower of a wild plant), and 5% black pepper. It is then needed to prepare a cake and placed on straw, which is then covered by straw and fermented at 30-33°C for
5-7 days. Freshly fermented dough is sun dried to get manapu microorganisms present in manapu are *Saccharomyces cerevisiae*, *Candia versatilis*, *Rhizopus* sp. and *P. pentosaceus* (Shrestha et al. 2002).

**Marcha**

*Marcha* is a ball-like amylolytic starter, used to ferment starchy materials into fermented beverage in Nepal, Bhutan and the Darjeeling hills and Sikkim in India (Tamang and Sarkar 1995; Tamang 2010a). During its preparation, glutinous rice is soaked, excess water discarded, pounded, wild herbs, old *marcha* (~1 %) are added, mater thick paste by adding water, and kept on wild fern leaves and fermented for 1-2 days, sun-dried and stored for a year or more (Tamang et al. 1996). Kobayashi et al. (1961) reported *Rhizopus oryzae*, *Mucor praini* and *Absidia lichtheimi* in *marcha* samples collected from Sikkim. Hesseltine et al. (1988) isolated *Mucor* and *Rhizopus* sp. in *marcha*. Tamang and Sarkar (1995) identified the microorganism in *marcha* of the Darjeeling Hills and Sikkim as *Pediococcus pentosaceus*, *Saccharomyces fibuliger*, *Pichia anomala*, *Mucor circinelloides*, and *Rhizopus chinensis*. Batra and Miller (1974) reported *Hansenula anomala* var. schneggii (*Pichia anomala*) in *marcha*. In Bhutan, *marcha* is called chang-poo, in which *Saccharomyces*, *Penicillium* sp. and *Aspergillus* sp. were reported (Uchimura et al. 1990). Microbial profiles of amylolytic starters of India, Nepal, and Bhutan are filamentous molds like, *Mucor circinelloides*, *Mucor hiemalis*, *R. chinensis*, and *R. stolonifer* variety *lyococcus* (Tamang et al. 1988); yeasts *S cerevisiae*, *S bayanus*, *Saccharomyces fibuliger*, *Sm. capsularis*, *Pichia anomala*, *P burtonii*, and *Candida glabrata*;

**Nuruk**

*Nuruk* is the starter for preparing Korean alcoholic drink *yakju, takju, makgeolli*, etc. (Jung et al. 2012; Shin et al. 2016). Historically the substrate for *nuruk* was rice but presently it is wheat (Park et al. 1977; Lee and Kim 2016). Generally, *nuruk* is prepared by natural inoculation of molds, bacteria, and yeasts; however, it can be prepared by inoculation with *Aspergillus usamii*. Traditionally *nuruk* is prepared by moistening wheat flour, kneaded and molded into a ball [0.8-1.6 kg (dry weight)] and fermented for 17 days at 30° C to 45° C, dried for 2 weeks and cured for 1-2 months at room temperature (Park et al. 1977). Kim (1968) isolated *Aspergillus oryzae* (10⁷ cfu/g), *A. niger* (10⁷ cfu/g), *Rhizopus* sp (10⁶ cfu/g), anaerobic bacteria (10⁷ cfu/g), aerobic bacteria (10⁶ to 10⁷ cfu/g) and yeasts (10⁵ cfu/g) from *nuruk*. Recent advances in high-throughput sequencing technologies such as DNA microarrays and next-generation sequencing (NGS) are rapidly changing the way microbial communities are studied (Roh et al. 2010). The Next Generation Sequencing result represents simple and rapid method of studying microbial ecology that permits the analysis of hundreds of thousands of nucleotide sequences. The phyla *Ascomycota* and *Zygomycota* were the predominant phyla in all samples of *nuruk*, constituting 85.4% (±31.1) and 14.3% (±30.9) of the fungal populations, respectively and *Basidiomycota* at a rate of 0.01%. NGS results of *nuruk*, showed dominance of *Saccharomycopsidaceae, Trichocomaceae, Mucoraceae* and *Saccharomycetaceae* at family level, constituting 99.6% (±0.6) of the fungal sequences (Jung et al. 2012). Yang et al.
(2013) reported that *Aspergillus oryzae* strains isolated from traditional Korean amylolytic starter, *nuruk* improves fermentation properties and rice beverage quality. Bal et al. (2016) identified the dominant *Aspergillus oryzae* mold from *nuruk* by using molecular (ITS-PCR) and biochemical characterization. They also reported the α-amylase, gluco-amylase as well as acid protease activity. The α-amylase and gluco-amylase activity were higher than the acid protease activity of *Aspergillus oryzae*. The α-amylase activity was positively correlated with glucoamylase activity. Fungal diversity in wheat-based *nuruk* by NGS and the fungal ITS database, revealed differences in mycobioime composition of the different samples of *nuruk*. Members of both *Ascomycota* and *Zygomycota* dominant in some *nuruk* samples whereas *Zygomycota* dominated some other samples of *nuruk* Bal et al. (2016). In comparison to the domestic samples, the commercial samples dominated by mostly genera of *Pichia*, *Wickerhamomyces*, unclassified members of *Saccharomycetales* Bal et al. (2016).

**Phab/dheli**

*Phab* and *dheli* are traditional ethnic amylolytic starters of Himachal Pradesh mostly North West Himalayas used for the preparation of *chhang*, *jau chhang* and *sura*, alcoholic beverages (Tamang et al. 2016c). The study revealed that yeasts and lactic acid bacteria are the major microflora of these amylolytic starters. Yeasts were identified by sequencing of D1/D2 26S rDNA regions as *Saccharomyces cerevisiae*, *Saccharomyces fibuligera*, *Pichia kudriavzevii* and *Candida tropicalis* (Thakur et al. 2015). The dominant lactic acid bacteria (LAB) were *Lactobacillus plantarum*, *Lactobacillus casei*, *Pediococcus* and *pentosaceus*
Enterococcus faecium identified on the basis of comparison of the sequence of 16S rRNA genes (Thakur et al. 2015).

**Ragi**

Ragi is an amylolytic starter culture of Indonesia where rice is used as a substrate (Saono et al. 1974; Surono 2016). During production of ragi, mainly rice or millet or cassava or other starchy bases are milled, mixed with herbs and spices, roasted together, sieved, water added and starter (ragi) from previous batch is mixed and shaped into balls. These are incubated at 25-30° C for 72 h in humid environment. Balls are dried in the sun and used as inoculum for the various fermentations. Went and Prinsen-Geerligs (1896) found *Monilia javanicus* (*Pichia anomala*) and *Saccharomyces cerevisiae* as principal yeasts in ragi. Dwidjoseputro and Wolf (1970) reported the yeasts *Candida parapsilosis*, *C. melinii*, *C. lactosa*, *Hansenula subpelliculosa*, *H. anomala* and *H. malanga* in ragi. Addition of spices to some ragi contributes other microorganisms or may inhibit the growth of undesirable microorganisms (Soedarsono 1972). Saono et al. (1974) conducted studies on mycoflora of ragi and products fermented by ragi such as tape keté la, tapé ketan hitam, oncom hitam and oncom mérâh from various places in West Java and reported that *Candida* sp. was dominating among yeasts, *Mucor* sp. and *Rhizopus* sp. were dominating among moulds. Kato et al. (1976) studied the properties of glucoamylase from ragi isolates of *Saccharomycopsis fibuligera*. Saono and Basuki (1978) reported thirteen species of *Candida* from ragi of Indonesia. Hadisepoetro et al. (1979) reported that population of yeast in three ragi was $5.6 \times 10^6$ to $1.4 \times 10^7$, bacteria was $3 \times 10^4$ to $1.8 \times 10^5$ and mould was $3.2 \times 10^4$ to $4 \times 10^4$. Ardhana and Fleet (1989) reported only single yeast *Candida*
pelliculosa and one mould Amylomyces rouxii in four samples of *ragi*. Yokotsuka (1991) reported the presence of mixed cultures in *ragi* mainly *Rhizopus* and *Mucor* among molds; other organisms such as *Amylomyces*, *Aspergillus*, *Fusarium*, *Candida*, *Saccharomyces*, *Hansenula*, *Endomycopsis* (*Saccharomycopsis*). Ishimaru and Nakano (1960) isolated *Streptococcus faecalis*, *Lactobacillus plantarum* and *Pediococcus pentosaceus* in *ragi* in the range of $10^5$ to $10^8$ cfu/g. Hesseltine and Ray (1988) reported that most of the bacteria isolated from *ragi* belong to *Pediococcus pentosaceus* and *Streptococcus faecalis*, which may produce secondary products from the glucose formed by the amylolytic yeasts and moulds always found in the starters. Ardhana and Fleet (1989) reported the presence of bacteria in all four samples studied, which included *Bacillus coagulans*, *B. brevis*, *B. stearothermophilus* and an unidentified species of *Acetobacter* at the level of $10^3$ to $10^4$ cfu/g. Saono et al. (1984) prepared *ragi* by using pure cultures of the selected molds and yeasts, *Amylomyces rouxii* and *Saccharomyces cerevisae* strains and pure culture of *Rhizopus formosaensis* and also prepared brem from this improved *ragi*. Elegado and Fujio (1993) isolated two polygalacturonase producing strains of *Rhizopus* spp from *ragi* and studied the enzyme stability in wide range of pH from 2-11 and tolerance at 50º C for 20 min. Uchimura et al. (1991) revealed that there is a higher variability rate of *Pediococcus pentosaceus* in older *ragi* than younger ones and the result suggested that rod-shaped bacteria cannot survive for a long time under dry conditions in *ragi*. Sujaya et al. (2010) reported bacterial diversity of Indonesian *ragi* and their dynamics during the fermentation as investigated by PCR-DGGE the result revealed that lactic acid bacteria were the predominant bacterial flora of *ragi* such as *Pediococcus pentosaceus*, *Enterococcus*,
Lactobillus sp, Lactobacillus sp., Enterococcus sp., Weissella sp., and some other bacterial populations were also reported such as Clostridium perfringent Eubacterium moniliforme, Clostridium sardiniensis, or Clostridium baratii Pediococcus, Weissella. Barus and Steffysia (2013) reported the genetic diversity of yeasts from Ragi tape “starter for cassava and glutinous rice fermentation from Indonesia” by using Internal Transcribed Spacer (ITS) region they reported that yeasts Pichia jadinii and Pichia kudriavzevii are dominant in Ragi.

Ranu dabai

Ranu dabai is an amylolytic stater of Assam (Ghosh and Das (2004). During the perpetration of ranu dabai six steps are involved: Washing of rice and storing of wash-water. After cleaning then glutinous rice on a soop, (a flat traditionally prepared tray generally made up of sliced bamboo) it is taken in a vessel (made of metal/clay) for washing. Clean water is poured in it, mixed and drained off. The discarded wash-water is stored in a container future use. Mixing and grinding: In this step traditional wooden husking machine dhiki is used for grinding purpose. The freshly collected plant materials grains are chopped and ground properly and taken out on a soop. Glutinous rice is taken in dhiki and partially powdered and 3–4 ranu dabai large old tablets are added for 10 kg of rice. After some time, paste of various plants is also added to it and mixes properly. The powdered mixture is now taken in a large dekchi and made into paste using the previously collected washed rice water. Clean gunny bags are then spread on the floor under shade. These tablets are completely handmade. The standard size is about 4.5–7 cm in diameter, which is kept in rows on the gunny bags, where these tablets are kept for 40–60 min. The sized ranu cakes vary from 1.5–15 cm in diameter.
Incubation: It is done inside a large bamboo basket made. Clean and dry straw is spread on the bottom of the bamboo basket and some old ranu tablets are kept on it and full basket is covered with the newly prepared ranu tablets, after filling of bamboo basket with tablets the basket is covered with polythene sheet or gunny bags and incubated in a dark and warm place and fermented for 2 to 3 days in summer and 4–6 days in winter season. They are taken out from the bamboo basket and are arranged in single layer on circular flat bamboo basket called dagra and kept for the sun drying for 7–8 days. After complete drying the ranu dabaĩs ready for storage and for further use for preparation of local alcoholic beverage, haria (Ghosh and Das 2004)

Ranu goti

Ranu goti prepared by some ethnic communities of Central India for the preparation of alcoholic beverage such as handia (Kumar and Rao 2007). During the perpetration of ranu goti firstly the glutinous rice washed, soaked and excess water is drained off and then powdered with help of dhiki. The rice powder is now mixed with powdered roots, leaves, bark, rhizomes; seeds of about 20-25 plants species in ratio of 2:1 with clean water and small pieces of cakes were made. These goti are kept for incubation in bamboo basket under closed conditions after incubation the ranu goti are taken out from the bamboo basket and are exposed to sun for drying for 7–8 days and are used for preparation of local alcoholic beverage, handia (Kumar and Rao 2007).
**Thiat**

*Thiat* is an amylolytic starter of Meghalaya used to ferment alcoholic beverage—*kiad* (Tamang 2010a). During the preparation of *thiat* firstly the glutinous rice washed, soaked and then powdered. The rice powder is now mixed with powdered, *khaw-tang/hawiang* plants leaves with clean water and small pieces of cakes were made in size ranging from 4-5 cm in diameter and 0.8-1.0 cm in thickness and are kept for fermentation in *malieng* and covered by *sla-pashor* after fermentation the *thait* are sun-dried and used as dry starters for alcohol production (Samati and Begum 2007).

**Vekur pitha**

*Vekur pitha* is traditionally prepared ethnic amylolytic starter of Assam and is commonly used for the traditional preparation of *ahom* which is consumed as mild alcoholic beverages during various ceremonies (Saikia et al. 2007). For the traditional preparation of amylolytic starter, *vekur pitha* glutinous rice grains (*saol*) and leaves of few wild plants are used. The plants ingredients and additive ingredients, which serve as source of, yeast *Saccharomyces cereviceae*. The leaves of plants are collected from the wilderness and exposed to natural sunlight for 2-3 days. Sun dried leaves are powdered and mixed with the powder of rice grain in a vessel containing few ml of clean water. Here, the powder old *pitha* 8 commonly called *ghai pitha* is mixed with freshly prepared *pitha* as source of yeast microflora. The semi-solid *pitha* is mixed with required ingredients and rolled into plate-disc shaped, wrapped with fresh leaves of *Musa paradisiaca* and kept in anaerobic environment over fire heat. The fire heat is maintained at 90-180 cm height for 5-6 days dry till it gets harder. Oval shaped dried *pitha*
containing yeast inoculum, rice powder and plant material is known as *vekur pitha*, which is preserved in natural conditions for future use for preparation of various alcoholic beverages. *Saccharomyces cerevisiae* is the major yeast, which plays vital role in fermentation of *vekur pitha* (Saikia et al. 2007).

**Xaj-pitha**

Bora et al. (2016) reported that *xaj-pitha*, a rice based ethnic amylolytic starter culture of Assam used to prepare the local alcoholic beverages. The microbial community of *xaj-pitha*, by NGS approach revealed the amylase producers, such as *Rhizopus delemar*, *Mucor circinelloides*, and *Aspergillus* sp. Ethanol producer’s viz., *Candida glabrata*, *Debaryomyces hansenii*, *Ogataea parapolymorpha*, *Wickerhamomyces ciferrii*, *Saccharomyces cerevisiae*, *Meyerozyma guilliermondii* and *Dekkera bruxellensis* (Bora et al. 2016). Some opportunistic contaminants were also reported from *xaj-pitha*. The bacterial population was dominated by LAB as *Lactobacillus plantarum*, *Lactobacillus brevis*, *Weissella cibaria*, *Lactococcus lactis* *Leuconostoc lactis*, *Weissella para mesenteroides*, *Leuconostoc pseudomesenteroides*, etc. (Bora et al. 2016).

**Some Ethnic Alcoholic beverages**

**Atingba**

*Atingba* is one of the popular traditionally prepared alcoholic beverages of Manipur, prepared from rice (Jeyaram et al. 2009). The Meitei community of Manipur mainly consumes it as food beverage on the several occasions. For preparation of *atingba*, rice is cooked first and then its water is allowed to remove and then cooled to room temperature. The powdered *Hamei* (starter culture for
Atingba) mixed properly with cooked rice with at the ratio of 5 cakes/10 kg of rice. The mixture is then placed within earthen pots, which are covered with hangla leaves (Alocasia sp.) and then mature is allowed for 3–4 days fermentation in summer and 6–7 days in winter season. This process is then followed by 2–3 days of submerged fermentation in earthen pot to produce the final alcoholic product atingba. It is then distilled to give a clear-liquor alcoholic beverage called yu in the Manipur. There are various types of yeasts and filamentous fungi that are responsible for fermentation of rice (substrate) to Atingba (product) yeasts, Saccharomyces cerevisiae, C. Pichia anomala, montana, C. parapsilosis, P. guilliermondii, Torulaspora delbrueckii, P. fabianii, Trichosporon sp., Candida tropicalis and molds like Mucor sp. and Rhizopus sp; whereas some important LAB are Pediococcus pentosaceus, Lactobacillus brevis are playing vital role in flavor and texture development (Tamang et al. 2007; Jeyaram et al. 2008).

Bhaati Jaanr

Bhaati jaanr is an ethnic rice-based mild alcoholic food beverage fermented by marcha in the Eastern Himalayan regions of Nepal, India and Bhutan (Tamang 2010a). During preparation, first rice is sccharified for 1-2 days in an earthen pot at room temperature and once the saccharification is achieved the vessel is made airtight and is allowed for fermentation for 2-3 days in summer and 7-8 days in winter season. The major microflora involved in Bhatti Jaanr saccharification and fermentation are filamentous fungi (Rhizopus chinensis, M. hiemalis, Mucor circinelloides, R. stolonifer and var. lyococcus) and yeasts (Candida glabrata, Saccharomyces cerevisiae and S. bayanus), and Lactic acid bacteria like
(Pediococcus pentosaceus, Lactobacillus bifermentans, and Lb. brevis) (Tamang and Thapa 2006). This microflora is responsible for development of flavor and acidity of the product. pH, titrable acidity, ethanol content and moisture content of the Bhaati jaanr is 3.5, 0.24%, 5.9%, and 83.4%, respectively. Bhaati jaanr is consumed as a staple food directly in Sikkim and Darjeeling (Tamang 2010a).

**Chhang**

Chyang or lugri is a mild alcoholic, foamy and translucent beverage, which is prepared by traditional fermentation. It is prepared by using the substrate barley (Hordeum nulum) locally known as sherokh in Ladakh (Bhatia et al. 1977). Chyang having a sweet-sour taste and aromatic flavor (Batra and Millner 1976; Batra 1986). During the chhang preparation, first Barley grains are cooked over a slow fire in the water just sufficient for absorption it and then after cooking the mixture is spread on blanket or burlap mat to remove the access water. The cooked barley grains at lukewarm stage are mix with starter culture, phab using in ratio of 1g/kg of barley. These mixtures are filled in drill bags, mostly in 20-kg batches, and then tightly packed. These mixtures are then packed by gunny bags to maintain the temperature around 30°C–35°C which is required for fermentation of barley it to Chyang after 7–8 days of fermentation (Bhatia et al. 1977). Microorganisms that plays significant role in the fermentation process of Chhang are yeasts Saccharomyces cerevisiae and S. uvarum (Batra 1986). Chyang is one of the popular mild alcoholic beverages traditionally prepared and consumed by the people of Ladakh (Bhatia et al. 1977).
**Kodo ko Jaanr**

*Kodo ko jaanr* is one of the most popular ethnic fermented finger millet (*Eleusine coracana*) beverages of the Himalayan regions of India with mild-alcoholic (4.8 %) and sweet taste (Tamang 2010a). *Kodo ko jaanr* has several synonyms as used by different ethnic groups of the Himalayan people such as *chyang* (Tibetan, Ladakhi, Drupka), *mandokpenaa thee* (Limboo), *mong chee* (Lepcha) (Tamang et al. 2016b). During its production, finger millet seeds are cleaned, washed and cooked for about 30 min, excess water is drained off and cooked millets are spread on a bamboo mat for cooling. About 1-2 % of powdered *marcha* is sprinkled over the cooked seeds, mixed thoroughly and packed in a bamboo basket lined with fresh fern (*Thelypteris erubescens*) and then covered with sack cloths, and fermented at room temperature for 2-4 days. The saccharified mass is transferred into an earthen pot or bamboo basket, made air-tight and fermented for 3-4 days during summer and 5-7 days in winter at room temperature for alcohol production. Freshly fermented *kodo ko jaanr* is filled into a bamboo-made vessel locally called *toongbaa*, and lukewarm water is added up to its edge and leave it for 10-15 min. Then, the milky white extract of *jaanr* is sipped through a narrow bamboo straw called *pipsing* which has a hole in a side near the bottom to avoid passing of grits. Water is added twice or thrice after sipping of the extract. Consumption of fermented finger millet beverages in exclusively decorated bamboo or wood-made vessel called *toongbaa* is unique in the Himalayas (Tamang et al. 1996). *Kodo ko jaanr* liquor is believed to be good tonic for ailing persons and post-natal women. After consumption, residual or grits of *kodo ko jaanr* are used as fodder for pigs and cattle. This is a good example of total
utilization of substrate as food and fodder, and also the discarded grits contain nutrient used as animal feed.

Marcha used as amylolytic starter supplements all functional microorganisms in kodo ko jaanr fermentation (Thapa and Tamang 2004). Mycelial molds have roles only in the initial phase of fermentation mostly in saccharification of the substrates. Yeasts *Pichia anomala*, *Saccharomyces cerevisiae*, *Candida glabrata*, *Saccharomycopsis fibuligera*, and LAB *Pediococcus pentosaceus* and *Lactobacillus bifermantans* have been recovered in kodo ko jaanr samples. Population of filamentous molds, which were originated from marcha, declines daily during *in situ* fermentation of kodo ko jaanr and finally disappears after fifth day (Thapa and Tamang 2006). *Sm. fibuligera* and *R. chinensis* saccharify and liquefy millets starch into glucose and produce alcohol *in situ* fermentation of kodo ko jaanr. Fermentation of finger millet enhances bio-enrichment of minerals such as Ca, Mg, Mn, Fe, K, P, contributing to mineral intake in daily diet of rural people (Thapa and Tamang 2004). Ailing persons and post-natal women consume the extract of kodo ko jaanr to regain the strength due to high calorie in jaanr.

**Sujen**

It is a mild alcoholic beverage is popular among the Deori, an ethnic community of Assam (Deori et al. 2007). It is also considered as pure and used as a holy water by the Deoro priests during various festivals and ceremonies. During sujen preparation, first the preparation of the natural starter called *mod pitha* is done and then the fermentation of sujen (Deori et al. 2007). Several types of plants species used for the preparation of *mod pitha* starter. Five kg of glutinous rice is soaked for about 2 hours in water, cleaned then mixed properly with the dried plant parts in a grounded in
dheki, a wooden grinder along with old mod pithas starters. The grounded starter powder is taken in a vessel for fermentation for duration of 7-15 days. After fermentation it is diluted for consumption (Das et al. 2012).

**Lao-Chao**

*Lao-Chao* is one of the famous alcoholic fermented beverages of China (Steinkraus 1996). During preparation, rice is boiled and then allows it for cooling on a mat, and then mixed properly with yeast cultures grown on rice and nosan leave. The yeast inoculated rice is then poured into a cone-shaped bamboo basket and an earthen pot is placed under the cone for the collection of the liquefied rice as it ferments. The fermented product (juice) is collected and transferred to new boiled rice for about 3 or 4 times in succession. The dominant microorganisms consists of filamentous fungi mainly *Rhizopus, Mucor*, yeasts and lactic acid bacteria. The final alcohol content of the product ranges from 12 to 14% (v/v) with pH 3.9 (Wang and Hesseltine 1970; Wei and Jong 1983).

**Poko**

*Poko* is traditionally prepared rice fermented alcoholic beverage of Nepal (Shrestha et al. 2002). It is very similar to *Bhatii ko Jaanr* an alcoholic beverage of Sikkim and Darjeeling Himalayas. It is generally consumed and served during the festive seasons and various ceremonies by the people of Nepal. The dominant micro-biota which plays important role during fermentation of *poko* are mainly *Rhizopus* and yeasts like *Saccharomyces cerevisiae, Candida versatile* and lactic acid bacteria, *Pediococcus pentosaceus* also playing very significant role in the product and flavor development.
This traditionally prepared ethnic alcoholic beverage of Nepal has strong socio-cultural significance (Shrestha et al. 2002).

**Tapé ketan**

*Tapé ketan* is a traditionally fermented, sweet/sour, alcoholic beverage of Indonesia (Steinkraus 1996). The cassava (*tapé ketella*) and glutinous rice (*tapé ketan*) are most common substrate used for *tapé ketan* fermentation. During preparation of *tapé ketan* the glutinous rice is washed and soaked for 1 h in water then cooked well, spread over a bamboo tray and then allowed to cool to room temperature. Then powdered *ragi*, amylolytic starter culture is sprinkled and mixed properly with rice and then placed in an earthenware pot for traditional fermentation. The sticky rice is converted to a soft, juicy mass with a sweet/sour; alcoholic flavor within 2 to 3 days of fermentation at room temperature now the product is ready for consumption. The *Tapé ketan* is acceptable for consumption even after one week of fermentation (Cronk et al. 1977). With the long fermentation the product becomes more liquid. The product gets ready for consumption after 36 to 48 h of fermentation at 30°C (Cronk et al. 1977). Malaysian *tapai* is also alcoholic beverage contains 27% of total sugar, 5% of ethanol (v/v), 23% of reducing sugar and pH of the product is 3.9 is acidic (Steinkraus 1996). *Tapé ketan* must be sweet to be edible and acceptable hence, the final product must be consumed between 3 to 4 days when the content of the reducing sugars in the product are highest (Merican and Yeoh 1977).
Saké

Saké is a national drink of Japan and is one of the most popular traditional non-distilled alcoholic drinks in the world (Jin et al. 2005). It is prepared from rice using koji and is clear, pale yellow, containing 15 to 20% alcohol. Polished rice is washed, steeped in water and steamed for 30-60 min, and then cooled, mixed with koji, water and a selected yeast starter culture for alcoholic fermentation. Main fermentation takes place in open tanks in cool conditions, starting at about 10°C, increasing to about 15°C. After fermentation, the liquid material called moromi is separated from the solids to give the clarified saké, which is settled, re-filtered, pasteurized and blended and diluted with water before bottling (Yoshizawa and Ishikawa 1989). Unique strains of *S. cerevisiae* have evolved to conduct those fermentations generating products with high ethanol content (12-20%), attractive flavor and aroma and odor (Kodama 1993). The first organisms developed in the mash under traditional fermentation conditions are nitrate-reducing bacteria such as *Pseudomonas*, *Achromobacter*, *Flavobacterium*, or *Micrococcus* spp. (Murakami 1972). These are followed by *Leuconostoc mesenteroides* variety saké and *Lactobacillus saké* and yeasts (Kodama and Yoshizawa 1977). The highly refined saké brewed by the most skillful brewers using very highly polished rice at low temperatures of 9 to 11°C for 25 to 30 days is known as *gonjoshu* (Kodama and Yoshizawa 1977). Most LAB that spoil saké are homofermentative rods and are more tolerant to ethanol and acid than non-spoilers (Inoue et al. 1992).

Difference in responses to osmotic stress between the laboratory and saké-brewing strains of *Saccharomyces cerevisiae* at the translational level was compared and found that enhancement of glycerol formation due to enhancement of the translation of proteins Hor2p, is required for growth of *S. cerevisiae* under high osmotic pressure.
condition (Hirasawa et al. 2009). *Saccharomyces cerevisiae* strains with disrupted ubiquitin-related genes produced more ethanol than the parental strain during *saké* brewing (Wu et al. 2009). Several researchers have reported on improved strains of *Aspergillus oryzae* for *saké* production in industrial scale (Hirooka et al. 2005; Kotaka et al. 2008; Hirasawa et al. 2009).

**Tapuy**

It is a highly acidic but alcoholic, sweet, aromatic and flavored rice beverage of Philippines (Steinkraus 1996). It is also known by other names as. In the process of preparation of *tapuy*, glutinous or ordinary white rice or a mixture of the two is soaked, cleaned then ground in a stone mill. The mash is mixed with pureed ginger and/or wild herbal root and starter culture, *bubod* from previous batches, incubated for three days, and dried (Sakai and Caldo 1983). *Saccharomycopsis fibuligera, Saccharomyces uvarum* is the major yeast flora playing vital role during fermentation of the *tapuy* (Sakai and Caldo 1985). Sakai and Caldo (1985) were reported that the enzyme glucoamylases were the primary amylases produced by *S. burtonii, S. fibuliger*, and Mucor molds helping in saccharification (conversion of polysaccharides to monosaccharide’s) fermentation as well as product and flavor development. The ethanol concentration of the final product is 4.93 % (v/v) on day 2 of fermentation and reached up to level of 15.5% v/v on day 14 of fermentation. Sanchez et al. (1985) reported that eight different varieties of *bubod*, yielded 12.9 to 17.3% (v/v) of ethanol in *tapuy*, with final pH of 3.9 to 4.5.
Zutho

Zutho is a mild alcoholic beverage popular among the Mao community in Nagaland (Tamang 2010a). In the preparation of zutho, firstly the rice is washed, soaked in water overnight, water is drained off, grinded in to powdery form and this is put in to bamboo bucket and mixed properly with warm water, then allow it for cooling, after cooling the powdered amylolytic starter which is locally known as khekhrii (Mao and Odyuon 2007) mixed properly and brewed for 7-8 h. After proper mixing the whole mass is poured in to earthen pot and more fresh water is added up to neck. Now this earthen pot kept for 3-4 days fermentation (Mao 1998). Nchiangne is another similar alcoholic beverage is prepared from glutinous rice in Nagaland (Tamang et al. 2012). The physiochemical profile of zutho showed the pH of the product is about 3.6, alcohol contents 5.1% and acidity of 5% Teramoto et al. (2002).