

Yellow Rice Wine Production Research

Subjects: **Microbiology**

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Yellow rice wine is a traditional fermented beverage in China. The microorganisms in the brewing process play a crucial role in shaping the composition and quality of the wine through their regulation of microbial growth, fermentation metabolites, metabolic balance, and ethanol production. Yellow rice wine not only has culinary value but also possesses potential medicinal value. This is attributed to the presence of polyphenolic compounds, antioxidants, and other natural products that can provide antioxidant and other probiotic effects. The fermentation process of yellow rice wine offers potential nutritional supplementation and improved digestion. While traditional brewing techniques have long been employed, modern biotechnology helps enhance the quality and stability of the wine by selecting suitable microbial strains, optimizing fermentation conditions, and precisely controlling the fermentation process. The collection of diverse fermentation microbes and the construction of specifically designed microbiota for yellow rice wine production could expedite the production of high-quality yellow rice wine. The ultimate goal is to enhance the nutritional value, conditioning function, and overall consumption experience of yellow rice wine.

yellow rice wine

microbial fermentation

biotechnology

microbiota

synthetic biology

1. The Traditional Brewing Process of Yellow Rice Wine

Yellow rice wine is typically named based on its production area, as different regions produce variations of the wine with distinct characteristics, including Nanyang, Shaoxing [1], Huzhou [2], and Guangdong [3]. With evolving consumer preferences, the Huangjiu industry is continuously adapting, innovating, and expanding. The Nanyang basin, known for its red millet with amber-colored grains and high nutritional value, provides proper raw materials for brewing special rice wine [4]. Nanyang yellow rice wine, famous for its long history and unique flavor, is among the most famous local yellow rice wines in China. The brewing process of Nanyang yellow rice wine typically involves the following steps [5] (**Figure 1**). Firstly, select high-quality millet as the main raw material, rinse and soak it in water, steam it for later use, and incorporate auxiliary ingredients such as wheat to enhance the taste and flavor of the yellow rice wine [6]. Next, a mixture of high-quality Jiuqu and wheat flour is prepared in a specific ratio, and water and sugar are added to form Jiuqu cakes or sticks [7]. These Jiuqu cakes or sticks undergo fermentation, exposure to air, and other processes, allowing the yeast and other microorganisms in red Jiuqu to ferment and mature. Subsequently, the steamed glutinous rice is cooled, combined with the Jiuqu, and placed in a rice wine fermentation tank or vat for the fermentation cycle. The main fermentation temperature is maintained at 27–30 °C, with a maximum temperature limit of 32 °C [8]. The primary fermentation period lasts for approximately 4–5 days. Following this, the post-fermentation temperature is controlled at 15–25 °C, and the post-fermentation stage continues for 15–20 days (**Figure 1**). Throughout the fermentation process, careful control of temperature,

humidity, and time is crucial to facilitate microorganisms and their enzyme activities, promoting the transformation of the mixture into yellow rice wine.

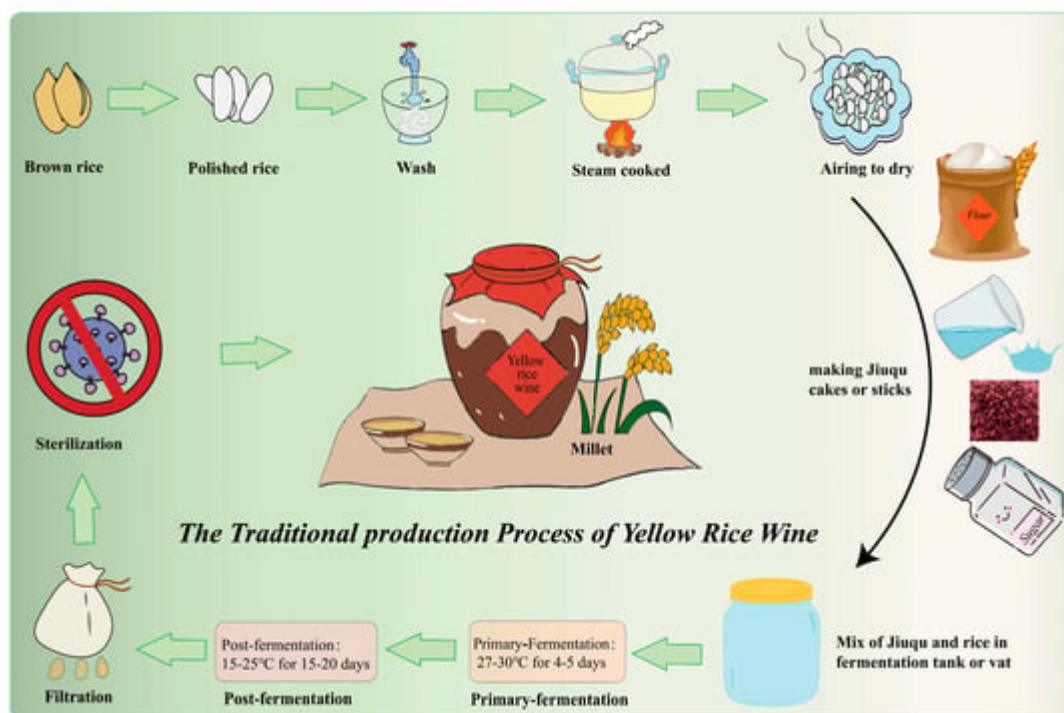


Figure 1. The traditional brewing process of yellow rice wine involves several stages. These stages include raw material preparation, Jiuqu preparation, primary fermentation, post-fermentation, residue separation, and sterilization.

After fermentation is complete, the solids in the yellow rice wine are separated using cloth bag filtration and pressing, resulting in a clear yellow wine liquid (**Figure 1**). This separated yellow wine solution is then stored in a maturing container for a specific aging period, typically ranging from several months to several years. Throughout the aging process, the yellow rice wine gradually mellows, and its aroma intensifies (**Figure 1**). The volatile components of yellow rice wine are highly complex, consisting of various aromatic compounds such as esters, alcohols, acids, and carbonyl compounds, which interact and harmonize with one another [9]. After aging, use a filter to further purify the aged yellow wine and filter out impurities and suspended solids. Purified yellow wine is usually bottled or barreled for sale and storage.

2. Microbial Improvement of the Composition and Quality of Yellow Rice Wine

Microorganisms are responsible for determining the flavor and overall quality of the final product [10]. The fermentation process of Shaoxing yellow rice wine typically involves the presence of 9 dominant genera, including *Bacillus*, *Streptococcus*, *Streptococcus*, *Vibrio*, *Thermoactinomyces*, *Pseudomonas*, *Staphylococcus*, *Enterobacterium*, and *Lactobacillus* [11]. The analysis of yellow rice wine Jiuqu from different sources revealed that

it primarily contains *Weissella*, *Lactobacillus*, *Lactococcus*, *Bacillus*, *Enterococcus*, and *Cronobacter* [12]. These microorganisms can impact the flavors and qualities of yellow rice wine through the regulation of microbial growth, fermentation metabolites [13][14][15], metabolic balance, and ethanol production [16][17][18].

During the process of fermentation, microorganisms produce ethanol and other compounds, which play a role in regulating the alcohol content and taste as well as enhancing the flavor characteristics of yellow rice wine [19]. Yellow rice wine contains various volatile substances, primarily originating from the raw materials and metabolites of microorganisms. The gas chromatography-mass spectrometry (GC-MS), in combination with full rate application (RATA) techniques, could analyze the volatile flavor compounds in yellow rice wine during fermentation. The alcohol content in yellow rice wine significantly increases during the early stages of fermentation. Alcohols have higher threshold values compared to aldehydes but still play an essential role in improving the overall flavor of yellow rice wine [20]. Throughout the brewing process of yellow rice wine, the content of alcoholic substances rises rapidly during the primary fermentation stage, leading to an increase in both the quantity and variety of flavor components within the wine [21][22]. *Aspergillus oryzae*, the mold used in yellow rice wine production, produces a range of compounds through metabolism, including esters, fatty acids, and alcohols. These substances significantly influence the taste, aroma, and overall flavor profile of the beverage and contribute to the unique flavor and aroma of yellow rice wine [23]. The *Meyerozyma* genus strain YB-12, isolated from the fermentation materials of yellow rice wine, possesses a similar ability to produce ethanol as the brewing yeast NRRL Y-567. However, the production of isobutanol and isoamyl alcohol by YB-12 is only 53.96% and 50.23% of that by NRRL Y-567, respectively [24]. Isobutanol and isoamyl alcohol are primarily responsible for the spicy, bitter, and astringent taste of yellow rice wine, as well as some potential side effects such as headaches [25][26].

During post-fermentation, the alcohol content of yellow rice wine initially decreases and then increases [27]. The majority of alcohols are produced by the breakdown of sugars or amino acids by yeast, with ethanol being generated from glucose [28][29][30]. Higher alcohols, on the other hand, are produced primarily through the breakdown of amino acids and pyruvate metabolism. Yeast also produces aromatic compounds such as esters and phenolic compounds, which give yellow rice wine its unique aroma and flavor. Some microorganisms produce beneficial metabolic substances such as amino acids, polyphenols, and vitamins, thereby increasing their nutritional value [30][31][32]. Through chromatographic analysis, amino acids, alcohols, acids, phenols, and esters are the major flavor components. The microbial genera, including yeast, *Aspergillus*, *Saccharopolysporium*, *Staphylococcus*, *Lactobacillus*, and *Lactococcus*, are most closely associated with the production of these flavor components [33]. The content and proportion of alcohol compounds produced by microorganisms in yellow rice wine are determined by the interaction between the brewing process and the microbiota [34]. Proper control of fermentation conditions, brewing temperature, and selection of appropriate strains can regulate the production of ethanol and other alcohols, thereby affecting the taste and flavors of yellow rice wine. Hence, it is possible to use specific microorganisms to regulate its fermentation products and produce yellow rice wine with different flavors and effects that highlight local characteristics.

Microorganisms regulate the metabolic balance during the brewing process of yellow rice wine [35]. Specifically, LAB are involved in the fermentation process, where they produce lactic acid and other organic acids. This helps to

maintain the appropriate acidity in the fermentation broth and regulates the taste and flavor of yellow rice wine. Yeast and LAB are the primary microorganisms responsible for controlling the pH of the liquor and the production of organic acids. LAB, in particular, are responsible for lowering the pH of the liquor by producing lactic acid, creating the acidic environment necessary for the fermentation of yellow rice wine. Additionally, yeast produces acetic acid and citric acid, which also contribute to the acidity and taste of the wine [36]. Metagenomic analysis of mechanized yellow rice wine, artificially fermented yellow rice wine, and poorly fermented artificial yellow rice wine at different fermentation stages has revealed that LAB dominate in well-fermented yellow rice wine, while *Lactobacillus brevis* dominates in spoilage yellow rice wine. The well-fermented artificial yellow rice wine mash exhibits more activity in converting malic acid to pyruvate and synthesizing lactate, while poorly fermented artificial yellow rice wine porridge tends to accumulate acetate [37]. By changing the pH of the fermentation broth, microorganisms can ensure it remains within the optimal range necessary for the yellow rice wine fermentation process, thus regulating the brewing process. Therefore, the ability of microorganisms to produce acid in yellow rice wine brewing is crucial in determining its final quality and flavor.

In the process of brewing yellow rice wine, a variety of microorganisms are selected for mixed fermentation, especially yeast [38]. During the fermentation of yellow rice wine, enzymes such as amylase and lipase are produced by the microorganisms, particularly the yeast. These enzymes aid in the hydrolysis and conversion of starch and fat in rice wine, improving its overall quality. A mixed fungal starter containing two strains, *Rhizopus oryzae* YF1 and *Aspergillus niger* YF2, was applied in wheat Qu under optimal conditions. The fermentation process led to an increase in the activities of amylase, acid protease, and cellulase. Additionally, the final fermentation products showed a 19.6% increase in short peptides and a 131.8% increase in free amino acids [39]. The synergistic metabolism between microorganisms in yellow rice wine production can impact the biochemical reactions, efficiency of substrate utilization, and overall characteristics of the final product. By designing microbial interactions, it is possible to optimize the composition and quality of yellow rice wine. Microorganisms produce antioxidants and enzymes in the brewing process of yellow rice wine, such as superoxide dismutase and glutathione peroxidase, which help to maintain the redox balance and prevent premature oxidation of the wine [3]. Yeasts, particularly *Saccharomyces cerevisiae*, are commonly used due to their high antioxidant capacity and ability to produce these enzymes [40][41]. *Pichia pastoris* is also used in brewing and fermentation processes [42].

The use of microorganisms in brewing yellow rice wine has become increasingly precise and efficient. Future understanding of the metabolic pathways and regulatory network in the brewing process, optimizing fermentation conditions and control technologies, developing new microbial strains, and improving fermentation agents will undoubtedly contribute to enhancing the quality and stability of yellow rice wine.

3. The Compounds in Yellow Rice Wine and Their Health Effects

Yellow rice wine has not only been known for its culinary uses but also for its beneficial effects on physical health. In many traditional Chinese medicine prescriptions, yellow rice wine has been used as a crucial ingredient. For instance, Taohong Siwu Tang, as documented in classic records, combines raw *Rehmannia glutinosa*, *Angelica*

sinensis, and *Safflower*, which are washed with yellow rice wine, to enhance its pharmacological effects in treating primary dysmenorrhea [43]. In recent years, the health effects of yellow rice wine have been revealed and described (Figure 2).

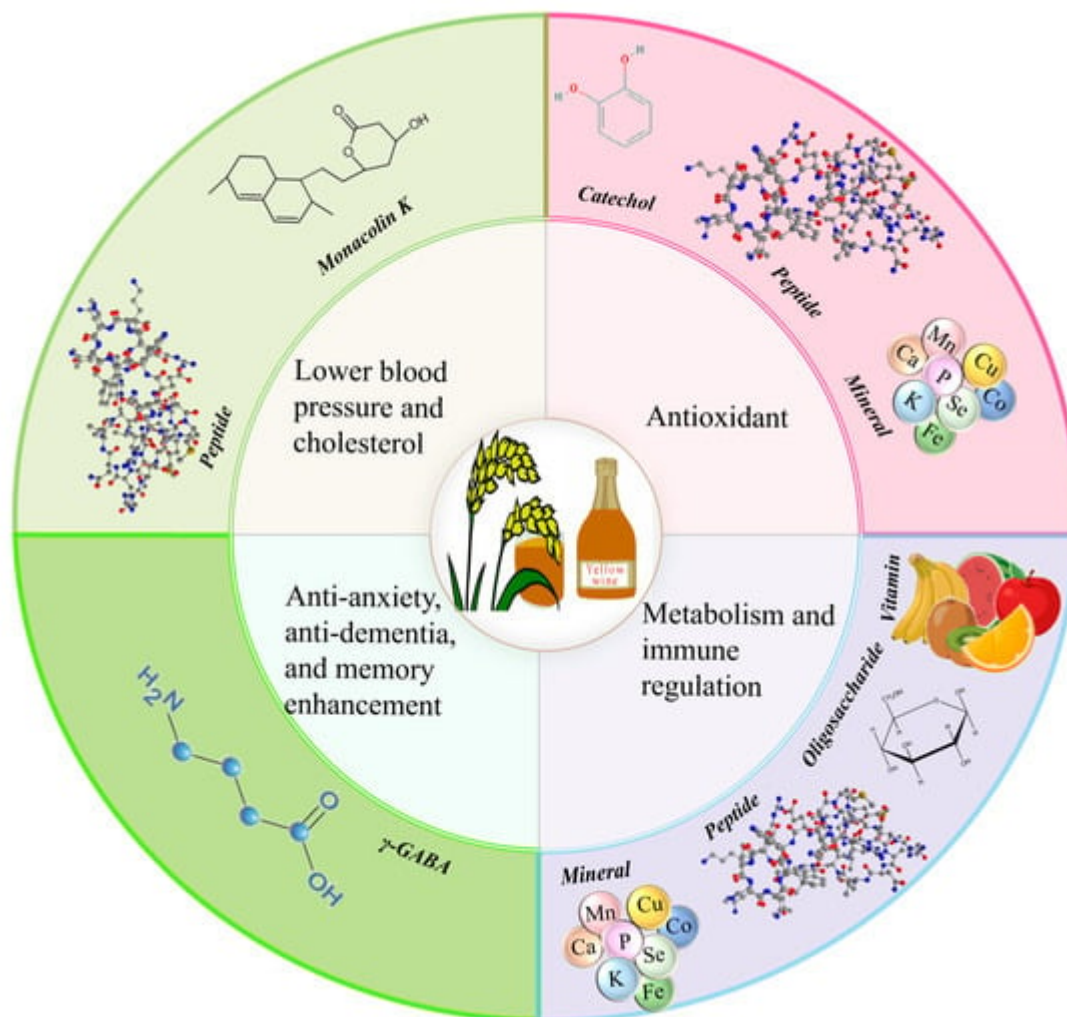


Figure 2. Yellow rice wine has a variety of components, including phenols, peptides, minerals, oligosaccharides, and vitamins, which contribute to its pharmacological effects such as blood pressure and cholesterol reduction, antioxidant, metabolism optimization, and immune regulation.

Yellow rice wine has demonstrated pharmacological effects in terms of its antioxidant properties [44][45][46], anti-aging benefits [47], and ability to support cardiovascular health [48][49]. Yellow rice wine is abundant in polyphenolic compounds such as flavonoids and anthocyanins, which have antioxidant properties. They eliminate free radicals in the body, slow down cellular oxidative damage, and help delay the aging process. The yeast and LAB present during the fermentation process of yellow rice wine contain antioxidant enzymes such as superoxide dismutase and glutathione peroxidase, which improve the body's antioxidant capacity. Additionally, various natural compounds in yellow rice wine possess anti-aging effects, enhance the body's ability to repair itself, and reduce the occurrence of cellular damage and aging processes. The active compounds in yellow rice wine promote healthy metabolism, boost the body's endurance, delay skin aging, and improve skin health. Yellow rice wine can mitigate inflammation

and mitochondrial dysfunction caused by doxorubicin (DOX) by regulating the gut microbiota and its associated metabolites. Further, antibiotic treatment increased the cardiotoxicity of DOX-treated rats and partially eliminated the anti-cardiotoxic effect of yellow rice wine on the gut microbiota, indicating that the microbiota plays a crucial role in the cardioprotective effect of yellow rice wine [50]. Moreover, yellow rice wine's ability to downregulate the TGF- β /smad3 signaling pathway in an Nrf2-dependent manner effectively reduces DOX-induced cardiac toxicity [51].

Though previous studies show that yellow rice wine has a cardiovascular protective effect [52], the exact composition and mechanism are still not fully understood. The polyphenols and peptide components in yellow wine can inhibit the proliferation and migration of vascular smooth muscle cells induced by homocysteine [53]. Additionally, yellow rice wine helps maintain a balance between metalloproteinases and tissue inhibitors of metalloproteinases [53]. The high presence of γ -aminobutyric acid (GABA) in yellow rice wine has been found to improve learning and memory in rats. Yellow rice wine, in a D-galactose-induced aging mouse model, significantly increased the activity of antioxidant enzymes and reduced levels of malondialdehyde in the brain and liver of the mice. This study showed improved cognitive impairment and anti-aging effects in mice [54]. Yellow rice wine contains peptides and minerals with antioxidant effects, as well as the ability to reduce blood pressure and cholesterol levels [55].

Yellow rice wine could promote gut health and regulate the immune system. LAB and their metabolic products present in yellow rice wine can promote a healthy balance of gut microbiota, thus maintaining gut health [56][57]. Additionally, yellow rice wine fermented with glutinous rice can relieve constipation caused by loperamide by regulating serum neurotransmitters and gut microbiota [58]. The probiotics and their metabolites, such as probiotic peptides, present in yellow rice wine have been shown to regulate the immune system and enhance the body's immune function. The organic acids in yellow rice wine, such as lactic acid and acetic acid, have beneficial effects on intestinal mucosal cells and the immune system.

Yellow rice wine can serve as a nutrient supplement and aids digestion [59]. This is because yeast and LAB generate bioactive constituents during the fermentation process; for example, the proteins can be converted into amino acids and peptides. Furthermore, yellow rice wine contains enzymes such as amylase and protease, which improve the digestion and absorption of food, offering relief from digestive discomfort. As a traditional fermented beverage, yellow rice wine shows promising medicinal properties, making it potentially valuable in healthcare and medicine. However, further understanding of the effectiveness and effects of yellow rice wine on health is necessary. Isolating and identifying the active ingredients present in yellow rice wine, exploring their related mechanisms, and conducting both preclinical and clinical trials would give insight into the health effects of yellow rice wine.

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