

Research Progress on Flavor and Quality of Chinese Rice Wine in the Brewing Process

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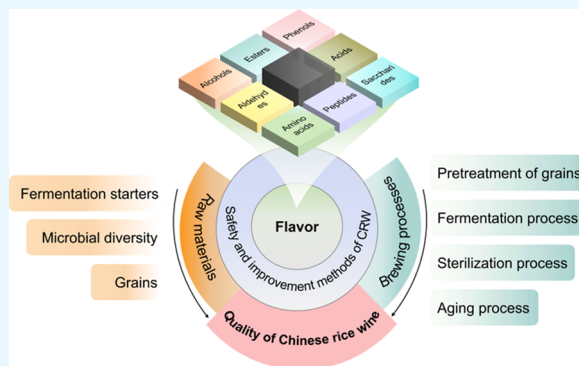
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ABSTRACT: Chinese rice wine (CRW) is a traditional and unique alcoholic beverage in China, favored by many consumers for its rich aroma, unique taste, and complex ingredients. Its flavor is primarily composed of volatile and nonvolatile compounds. These flavor compounds are partly derived from grains and starters (Qu), while the other part is produced by microbial metabolism and chemical reactions during the brewing process. Additionally, ethyl carbamate (EC) in CRW, a hazardous chemical, necessitates controlling its concentration during brewing. In recent years, numerous new brewing techniques for CRW have emerged. Therefore, this paper aims to collect aroma descriptions and thresholds of flavor compounds in CRW, summarize the relationship between the brewing process of CRW and flavor formation, outline methods for reducing the concentration of EC in the brewing process of CRW, and summarize the four stages (pretreatment of grains, fermentation, sterilization, and aging process) of new techniques. Furthermore, we will compare the advantages and disadvantages of different approaches, with the expectation of providing a valuable reference for improving the quality of CRW.



1. INTRODUCTION

Chinese rice wine (CRW), also known as yellow rice wine and Huangjiu, is a traditional alcoholic beverage with a history of more than 9,000 years.¹ CRW contains many necessary nutrients for the human body, such as proteins, vitamins, and microelements.² These nutrients are combined with alcohols, acids, esters, amino acids, phenols, aldehydes, and sugars to give CRW its unique flavor and nutritional value.³ The flavor of CRW is a crucial factor affecting the quality of CRW and consumers' preferences. Therefore, CRW is popular around the world. According to statistics, China has become a major exporter of CRW globally. However, CRW shares a food safety problem common to all fermented foods and alcoholic beverages: high levels of ethyl carbamate (EC), which is a substance with genotoxicity and strong carcinogenicity. In 2007, EC was classified as a 2A carcinogen by the International Agency for Research on Cancer.⁴ Therefore, the safety problem of CRW is a concern. This has not only restricted the export of CRW but also raised concerns about its long-term safety. Therefore, ensuring the flavor and safety of CRW is an ongoing problem that researchers and brewers want to solve.

CRW is made from grains that have been soaked, cooked, and fermented with starters.⁵ The unique flavor and taste of CRW are created through the use of different grains and starters combined with complex techniques. The flavor compounds of CRW mainly include volatile and nonvolatile

compounds.⁶ The flavor compounds of CRW are mainly derived from the flavor compounds of grains and starters, as well as the metabolism of protein and starch by microorganisms.⁷ In addition, the different techniques used in each brewing stage also affect the flavor of CRW. Although these flavor compounds belong to trace elements, they play an important role in determining the style and quality of CRW.⁸ We also collected aroma descriptions and the threshold of flavor compounds in CRW in Table 1. The traditional brewing process mainly involves pretreatment of grains (soaking and steaming), a fermentation process, a sterilization process, and an aging process. However, there are long brewing cycles and high production costs associated with handcrafted traditional brewing in the spring and fall seasons. To overcome the limitations of traditional production methods, researchers have developed a series of new brewing techniques to replace them. This paper mainly describes the relationship between the brewing process of CRW and flavor formation, as well as alternative new brewing technologies. Additionally, a solution

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Table 1. Aroma Description and Threshold of Flavor Compounds in CRW^a

No.	Compound	Aroma description	Threshold (mg/L)
Alcohols			
1	1-Propanol	Apple-like odor; bitter taste.	160,000
2	1-Butanol	Banana-like, alcoholic, sweet odor.	150,000
3	1-Octanol	Fresh, orange-rose, sweet, oily, herbaceous, fruity odor.	1,100
4	1-Dodecanol	Fatty odor; unpleasant at high concentrations, but floral on dilution.	/
5	1-Hexanol	Fragrant, woody, oily, aromatic, fatty odor; a pungent, spicy taste.	159.6
6	1-Nonanol	Fat, loral, oil odor.	45.5
7	1-Octen-3-ol	Powerful, sweet, herbaceous, reminiscent, lavender, rose, hay, herbaceous-like odor.	40
8	1-Pentanol	Fruity, balsamic aroma.	16,220
9	1-Penten-3-ol	/	/
10	1-Heptanol	Fragrant, woody, oily, faint, aromatic, fatty odor; pungent, spicy taste.	5.4
11	1,6-Octadien-3-ol	Pleasant floral odor.	0.22
12	2-Ethyl-1-hexanol	/	/
13	2-Methyl-1-propanol	Nail-polish-like odor.	360
14	2,3-Butanediol	Buttery, creamy, and fruity odor.	120,000
15	2-Ethyl-1-hexanol	Mild, oily, sweet, floral, rose, fatty-floral, fruity.	8000
16	2-Heptanol	/	/
17	2-Methylbutanol	Alcoholic, nail-polish-like odor.	69.6
18	2-Nonanol	/	/
19	2-Octanol	/	/
20	2-Furan methanol	Mild, warm, oily, burnt, cooked sugar odor.	2000
21	2-Methylpropanol	Nail-polish-like, solvent-like odor.	651
22	2-Vinyl-ethanol	/	/
23	3-Ethoxy-1-propanol	/	/
24	3-Methylbutanol	Alcoholic, nail-polish-like odor.	/
25	3-Methylbutanol	Alcoholic, nail-polish-like odor.	30,000
26	3-Ethylpentan-2-ol	/	/
27	3-Furan methanol	/	/
28	3-Methyl-1-butanol	Whiskey, malt odor.	1,000
29	3-Methyl-1-pentanol	/	/
30	3-Methylthiopropanol	Sulfur-like, rotten cabbage odor.	1,000
31	3-Octenol	/	/
32	4-Methyl-1-pentanol	/	/
33	7-Methanoazulen- 6-ol	/	/
34	Citronellol	Rose-like, peach-like odor; bitter taste.	62–2200
35	Cyclohexanol	/	/
36	Ethanol	Pleasant, fruity odor; slightly pungent, sweet taste.	1,100
37	Gentanol	/	/
38	Geosmin	Earthy, moldy odor.	10
39	Hexyl alcohol	/	/
40	Isoamyl alcohol	Pungent, balsamic odor.	6,600
41	Isopentyl alcohol	/	/
42	Isopropanol	Alcoholic, unpleasant odor; burning taste.	/
43	Geraniol	Rose-like odor.	8.54
44	L-menthol	/	810
45	Phenyl methanol	Pleasant, fruity odor; slightly pungent, sweet taste.	20,000
46	Phenyl ethanol	Flowery, honey-like odor; bitter taste.	1,100
47	Butan-1-ol	Wine, medicine, fruit-like odor.	/
Acids			
48	2-Methylpropanoic acid	Acidic taste.	2,300
49	2-Oxobutyric acid	/	/
50	3-Methylbutanoic acid	Smelly odor; acidic taste.	33.4
51	3-Methylbutanoic acid	Rancid, acidic odor.	/
52	Acetic acid	Strong, pungent, vinegar odor.	200,000
53	Butanoic acid	Cheese-like odor; sour taste.	173
54	Benzoic acid	/	/
55	Citric acid	Odorless or pleasant sour taste.	350
56	Decanoic acid	Fatty, unpleasant, rancid odor.	/
57	Fumaric acid	Odorless or tart, acidic-sour taste.	/
58	Heptanoic acid	Disagreeable rancid, sour, sweat-like, fatty odor.	3,000

Table 1. continued

No.	Compound	Aroma description	Threshold (mg/L)
Acids			
59	Hexanoic acid	Sickening, sweaty, sour, pungent, cheesy, unpleasant odor; acrid taste.	420
60	Lactic acid	Odorless or sour taste.	400
61	Lauric acid	Fatty odor.	/
62	Malic acid	Odorless or faint, acrid odor; a bit of sharp sour taste.	350
63	Myristic acid	Faint, waxy, oily odor.	12,000
64	Nonadecanoic acid	Fatty acid taste.	/
65	Nonanoic acid	Fatty, characteristic odor; corresponding unpleasant taste.	/
66	Octanoic acid	Sweat, cheese-like taste	500
67	Oxalic acid	Odorless or sour, tart taste.	/
68	Palmitic acid	Odorless or slight odor and taste.	/
69	Pentadecanoic acid	Waxy aroma.	/
70	Pentanoic acid	Cheese-like; acidic taste.	/
71	Propanoic acid	Pineapple-like, banana-like odor.	/
72	Pyruvic acid	Sour odor; pleasant, sour taste with a burning, somewhat sweet note.	/
73	Stearic acid	Tallow-like taste.	/
74	Succinic acid	Sour, astringent, umami taste	200
75	Tartaric acid	Sour taste.	300
76	Tetradecanoic acid	/	/
77	<i>trans</i> -Cinnamic acid	Cinnamon-like	/
Esters			
78	2-Butenoic acid ethyl ester	/	2.5
79	2-Hydroxy-ethyl propionate	/	/
80	2-Hydroxy- γ -butyrolactone	/	/
81	2-Methylpropyl acetate	Fruity odor.	1,605
82	2-Phenylethyl acetate	Rose-like, floral odor.	250
83	3-Methylbutyl acetate	Banana-like odor; sweet taste.	30
84	3-Methylbutyl lactate	/	/
85	Bis(2-methylpropyl) hexanedioate	/	/
86	Dehydromevalonic lactone	/	/
87	Di(<i>sec</i> -butyl) 2-methylbutanedioate	/	/
88	Diethyl butanedioate	/	/
89	Diethyl octanedioate	/	/
90	Diethyl suberate	/	/
91	Diethyl succinate	Faint, pleasant odor.	75,000
92	Diisobutyl phthalate	/	/
93	Ethyl (9 <i>E</i>)-9-octadecenoate	/	/
94	Ethyl 2-hydroxy-4-methylvalerate	/	/
95	Ethyl 3-phenylpropionate	/	/
96	Ethyl (9 <i>Z</i> ,12 <i>Z</i> ,15 <i>Z</i>)-9,12,15-octadecatrienoate	/	/
97	Ethyl 2-butenoate	/	/
98	Ethyl 2-hydroxy-2-methylpent-4-enoate	Sweet berry-like odor with rum note.	/
99	Ethyl 2-hydroxy-3-methylbutanoate	/	/
100	Ethyl 2-hydroxybenzoate	/	115
101	Ethyl 2-hydroxybutanoate	/	/
102	Ethyl 2-hydroxyhexanoate	/	/
103	Ethyl 2-methylbutanoate	Floral odor.	18
104	Ethyl 2-methylpropanoate	Fruity-like, sweet odor.	15
105	Ethyl 2-phenylacetate	Rosy, honey-like odor.	35,000
106	Ethyl 3-phenylpropanoate	Ethereal, rum, fruity, floral odor.	/
107	Ethyl 3-ethoxy-propanoate	/	/
108	Ethyl 3-methylbutanoate	Sweet, fruity, apple-like odor.	30
109	Ethyl 3-pyridinecarboxylate	/	/
110	Ethyl 4-hydroxybutanoate	/	/
111	Ethyl 4-methylbenzoate	/	/
112	Ethyl acetate	Solvent, fruity odor.	7,500
113	Ethyl benzoate	Fruity odor.	1,430
114	Ethyl butyrate	Pineapple, sweet odor.	300
115	Ethyl caprate	Grape-like odor.	5
116	Ethyl caprylate	Pleasant, fruity, floral odor.	5

Table 1. continued

No.	Compound	Aroma description	Threshold (mg/L)
Esters			
117	Ethyl cetylate	Mild, waxy, sweet odor; creamy-like taste.	/
118	Ethyl cinnamate	Cinnamon-like odor.	/
119	Ethyl decanoate	/	23
120	Ethyl heptadecanoate	/	/
121	Ethyl heptanoate	Fruity odor.	220
122	Ethyl hexadecanoate	/	/
123	Ethyl hexanoate	Winey, fruity, sweet odor.	55
124	Ethyl hydrogen succinate	/	/
125	Ethyl isobutyrate	Fruity, sweet, rubber-like odor.	15
126	Ethyl lactate	Buttery, creamy, fruity	128,000
127	Ethyl laurate	Floral, peanut-like odor.	500
128	Ethyl linoleate	Waxy, creamy, fatty, coconut odor.	/
129	Ethyl myristate	Mild, waxy, soapy odor.	180
130	Ethyl nonanoate	/	1,200
131	Ethyl octadecanoate	/	/
132	Ethyl octanoate	Fruity odor.	5
133	Ethyl oleate	/	870
134	Ethyl stearate	/	500
135	Ethyl pentadecanoate	/	/
136	Ethyl pentanoate	Fruity odor.	20
137	Ethyl phenylacetate	Pleasant, sweet odor suggestive of honey and bittersweet flavor.	13
138	Ethyl propionate	Pineapple	1,800
139	Ethyl tetradecanoate	/	/
140	Ethyl undecanoate	/	/
141	Ethyl vanillate	Vanilla-like odor.	990
142	Ethyl-(9Z,12Z)-9,12-octadecadienoate	/	/
143	Ethyl-4-hydroxybenzoate	/	/
144	Ethyl-3-phenylpropanoate	/	30
145	Ethyl 2-furoate	Floral odor.	16,000
146	Ethyl nicotinoate	Smoky-like odor.	/
147	Ethyl 3-methyl-2-butenolate	/	35
148	Ethyl propanoate	Sweet, fruity-like odor.	10
149	Ethyl-2-hydroxypropanoate	/	/
150	Hexyl acetate	/	/
151	Hexadecanoate	/	/
152	Isoamyl acetate	Fruity, fragrant odor; bittersweet taste.	1,100
153	Isoamyl butyrate	/	/
154	Isoamyl hexanoate	/	/
155	Isoamyl octanoate	/	/
156	Isobutyrate	/	/
157	Isopentyl isobutyrate	/	/
158	Methyl butanoate	Ether, fruit, sweet, apple-like odor.	/
159	Methyl dodecanoate	/	/
160	Methyl laurate	Fatty, floral odor.	/
161	Methyl octanoate	/	/
162	Methyl oleate	Pleasant fatty ester odor.	/
163	Methyl salicylate	/	/
164	Octyl formate	/	/
165	Phenethyl acetate	Powerful floral, fruity odor.	/
166	Phenethyl isobutyrate	Fruity odor; bittersweet taste.	/
167	2-Phenylethyl propionate	/	/
168	Propyl acetate	Fruity	/
169	Propyl butyrate	/	/
170	Sotolon	Caramel-like, seasoning-like odor.	9
171	Styrene	/	/
172	Tetraethylene glycol	/	/
173	γ -Butyrolactone	Buttery odor.	16,000
174	γ -Dodecalactone	Peach-like odor.	/
175	γ -Nonalactone	Coconut-like, peach-like odor.	30

Table 1. continued

No.	Compound	Aroma description	Threshold (mg/L)
Esters			
176	γ -Decalactone	Sweet.	/
177	γ -Octanoic lactone	/	/
Amino acids			
178	Alanine	Bittersweet taste.	8
179	Arginine	Flat to bitter taste.	9
180	Aspartic acid	Flat, sour, slightly bitter taste.	2,320
181	Cysteine	Salty taste.	191
182	Citrulline		972
183	Glutamic acid	Meaty odor; salty, fresh taste.	3
184	Glycine	Sweet.	846
185	Histidine	Mineral odor; flat to bitter taste.	129
186	Isoleucine	Flat to bitter taste.	555
187	Leucine	Flat to bitter taste.	1,092
188	Lysine	Mineral odor; bitter taste.	1,738
189	Methionine	Possibly sulphurous, meaty odor; flat to bitter taste.	2,196
190	Phenylalanine	Bitter taste.	3,061
191	Proline	Sweet or salty or sour taste.	300
192	Serine	Flat to sweet taste.	487
193	Threonine	Flat to sweet taste.	2,196
194	Tyrosine	Sour taste.	3061
195	Valine	Bittersweet taste.	300–500
Aldehydes			
196	2-Methyl-2-butenal	Penetrating, powerful, green, ethereal odor.	/
197	2-Methylbutanal	Malty odor.	120
198	2,4-Pentadienal	/	14,100
199	2-Phenyl-2-butenal	Green, floral, woody aroma.	/
200	2-Thiophenecarboxaldehyde	/	253
201	2-Methylbenzaldehyde	/	200
202	2-Tridecenal	/	/
203	3-(Methylthio)propionaldehyde	Boiled potatoes.	50
204	3-Methylbutanal	Malty odor.	120
205	5-Ethylfurfural	/	1,443
206	Furfural	Sweet, almond odor.	14,100
207	Heptanal	/	25,000
208	Hexanal	Green, grass-like odor.	990
209	5-Methyl-2-furfural	Roasted odor.	/
210	Acetaldehyde	Green leaves, fruity odor.	2,500
211	Benzaldehyde	Fruity, floral odor.	160
212	Benzeneacetaldehyde	/	/
213	Capraldehyde	/	/
214	Cinnamaldehyde	Cinnamon-like.	160
215	Decanal	Sweet, floral, citrus, fatty odor.	/
216	Methional	Pungent, potato-like odor.	/
217	Isobutyraldehyde	Sharp, pungent odor.	/
218	Isovaleraldehyde	/	1.1
219	Isobutyraldehyde diethyl acetal	/	/
220	(<i>E</i>)-2-Pentenal		980
221	Octanal	/	/
222	Phenylacetaldehyde	Floral, rose-like odor.	24
223	Propanal	Alcohol, solvent.	/
224	Pentanal	/	/
225	Nonaldehyde	/	/
226	Nonanal	Green, floral, citrus-like odor.	1.3
227	Salicylaldehyde	/	28
228	Vanillin	Sweet, vanilla-like odor.	487
229	Phenols		
230	2-Methoxy-4-vinylphenol	/	/
231	3-Hydroxy-2-methyl-4 <i>H</i> -pyran-4-one	/	/
232	4-Ethylguaiaicol	Clove-like, smoky-like odor; spicy taste.	33

Table 1. continued

No.	Compound	Aroma description	Threshold (mg/L)
Aldehydes			
233	4-Ethylphenol	Smoky-like odor.	30
234	4-Methylguaiaicol	/	9.5
235	4-Methylphenol	Smoky-like, phenolic odor.	68
236	4-Vinylguaiaicol	Clove-like odor; spicy taste.	40
237	4-Ethyl-2-methoxy-phenol	Spicy, smoke odor.	60
238	Guaiaicol	Clove-like odor; spicy taste.	10
239	Butylated hydroxytoluene	/	/
240	Thymol	/	/
241	Phenol	Phenolic, medicinal odor.	30
242	2,4-Di- <i>tert</i> -butylphenol	/	/
Ketones			
243	1-Octen-3-one	Mushroom-like odor.	/
244	1-Phenyl-1-propanone	/	/
245	2-Octanone	Apple-like aroma.	50
246	2-Nonanone	Fruity, foral aroma.	82
247	2-Pentanone	Banana and pineapple.	/
248	2-Heptanone	Fruity, sweet odor.	/
249	2-Undecanone	/	41
250	2,3-Butanedione	Buttery, cream-like odor.	/
251	2,6-Dimethyl-4-heptanone	Mint-like odor.	/
252	3-Hydroxy-2-butanone	Buttery odor.	/
253	3-Octanone	/	65
254	3-Octen-2-one	/	/
255	3-Penten-2-one	/	/
256	6-Methyl-5-hepten-2-one	/	/
257	Acetophenone	Sweet, fruity, floral odor.	65
258	Acetovanillone	Vanilla-like odor.	50.2
259	Butan-2-one	Acetone-like odor.	/
260	β -Damascenone	Floral odor.	82
261	5-Fluoro-2-hydroxy acetophenone	/	/
262	Heptan-2-one	Soap-like odor.	/
263	Pentan-2-one	Ether, fruity-like odor.	/
264	Propan-2-one	Pungent taste.	/
Others			
265	1,2-Dimethoxybenzene	/	/
266	Naphthalene	/	6
267	Ethylbenzene	/	/
268	<i>o</i> -Xylene	/	/
269	1,3-Xylene	/	/
270	2-Methylpyrazine	Peanut.	6.4
271	2,3,5,6-Tetramethylpyrazine	Roasted odor.	20.7
272	<i>p</i> -Xylene	/	/
273	2,6-Dimethylpyrazine	Cooked rice, nutty odor.	13.7
274	2-Acetyl-1-pyrroline	Cooked rice, popcorn-like odor.	/
275	2-Acetyl-5-methylfuran	Plant, leaf-like odor.	/
276	2-Acetylfuran	Caramel-like odor; sweet taste.	/
277	2-Acetylpyrrole	Nutty odor.	/
278	2-Furanacrolein	/	/
279	5-Methylfurfural	Burnt sugar odor.	20,000
280	2,3-Dimethylpyrazine	Nutty, roasted odor.	/
281	<i>n</i> -Propylbenzamide	/	/
282	Benzothiazole	Rubber-like odor.	/
283	2-Pentylfuran	Fruity, green odor.	6
284	1,1-Diethoxyethane	Fruity odor.	/
285	Heterocycles	/	/
286	Dimethyl trisulfide	Cabbage-like odor.	2.4
287	Methyl pyrazine	Nutty.	/
288	Cyclooctane	/	/
289	Butanoate	/	321

Table 1. continued

No.	Compound	Aroma description	Threshold (mg/L)
		Others	
290	Cocal	/	/
292	Ethyl 3-hydroxybutyrate	/	/
293	Four ethylene glycol diethyl ether	/	/
294	Methyl pentadecane ether	/	/
295	Methyl dihydrojasmonate	/	/

^a/: not found in references. Flavor compounds in CRW were obtained from references 14, 22, 23, 33, 54, 62, 86, 87, 98, 119, and 120. Aroma description and threshold were obtained from references 6, 8, 10, 19–21, 49, 50, 74, 75, 103, 105, and 121–123.

to the safety problem of CRW is proposed. This paper aims to provide researchers and brewers with a valuable reference for improving the flavor and quality of CRW through the selection of grains, starters, and processing techniques and reducing the concentration of EC.

2. MAIN FLAVOR COMPOUNDS IN CRW

The flavor compounds in CRW can be categorized into volatile compounds and nonvolatile compounds. Volatile compounds are primarily produced through intricate chemical reactions in the brewing process, including alcohols, esters, phenols, and aldehydes. Among these, alcohols are the largest volatile and semivolatile compounds. Esters are the most abundant volatile compounds in CRW. On the other hand, nonvolatile compounds mainly result from the decomposition of proteins and starches in grains and mainly include acids, amino acids, peptides, and saccharides.

2.1. Volatile Compounds. **2.1.1. Alcohols.** The synthesis pathways of alcohols include amino acid metabolism, lactose metabolism, methyl ketone reduction, and degradation of linoleic acid and linolenic acid.⁹ Leucine degradation produces isoamyl alcohol, which can cause a bitter taste when its concentration is too high.¹⁰ However, microorganisms can effectively reduce the presence of isoamyl alcohol,^{11,12} and the use of appropriate strains can help decrease the number of bitter compounds in CRW. Additionally, 2,3-butanediol, an important byproduct of fermentation, has a special bitter and butter aroma that can alter the overall flavor. Ethanol, which has a sweet flavor, is the alcohol with the highest content in yellow rice wine and is also the most important as it serves as a precursor for synthesizing flavor compounds such as phenyl ethanol and ethyl ester.¹³ 1-Propanol has an aesthetic odor; 2-methyl-1-propanol has an irritating taste; 1-butanol has a floral flavor; and isoamyl alcohol has an astringent and irritating taste. Additionally, CRW contains higher alcohols, which are mixtures of alcohols containing more than six carbon atoms. When the concentration of higher alcohols in CRW exceeds 400 mg/L, it can cause a strong irritation. Concentrations of higher alcohols lower than 300 mg/L can produce fruity flavors.¹⁴ However, low concentrations of 1-hexanol still give CRW a fruity aroma and slightly bitter, plant-like taste.¹⁵ Higher alcohols are mainly formed through an amino acid synthesis pathway, Ehrlich pathway, and sugar metabolism pathway.¹⁰ The most studied higher alcohol is phenyl ethanol, which can be produced by yeast under anaerobic conditions and has a rose honey flavor.¹⁶ Phenyl ethanol is a compound in CRW that contributes to its flavor by increasing caramel-like aroma intensity and decreasing fruity aroma intensity through the masking effect of its sweet smell on alcohol odor.¹⁷ Therefore, alcohols make an essential contribution to the flavor

of CRW. Although most alcohols provide fragrance for CRW, some may also cause significant irritation and should be kept at appropriate concentrations.

2.1.2. Esters. Esters have three sources: (1) raw materials; (2) enzymatic esterification of acids and alcohols (including phenols) during microbial metabolism;²¹ and (3) condensation reaction in the aging process.²² During the brewing process, the concentration of esters gradually increases. Eventually, ethyl hexanoate, ethyl butyrate, and ethyl caprylate exhibit the highest concentration increases. Ethyl hexanoate provides a fruity aroma, and ethyl butyrate and ethyl caprylate impart apple and pineapple flavors.¹⁸ Esters such as ethyl acetate, ethyl lactate, ethyl hexanoate, ethyl phenylacetate, and diethyl succinate give CRW a strong floral and fruity aroma,²³ which can also contribute to different taste sensations. For example, ethyl acetate and ethyl lactate were perceived as bitter in sensory evaluation, while ethyl hexanoate, ethyl phenylacetate, and diethyl succinate were perceived as sweet. Additionally, ethyl lactate, ethyl acetate, ethyl caprylate, diethyl succinate, and ethyl phenylacetate can also be used as reference indexes to distinguish between traditional and industrial CRW.²⁴ In addition, esters can also serve as indicators of both aged and young CRW. Principal compound analysis showed that sotolon, ethyl phenylacetate, ethyl nicotinoate, ethyl 2-methylpropanoate, ethyl 3-methylbutanoate, and γ -decalactone were the main esters in aged CRW.²⁴ Sotolon is produced by the condensation of acetaldehyde and 2-oxobutyric acid during the aging process.²² The longer the aging time, the higher the sotolon concentration in CRW.⁶ Most of the ethyl ester is formed by esterification of fatty acids and ethanol. A high content of ethyl ester makes CRW present a cheese flavor and fruit flavor.²⁵ More than 85% of the total ester content was accounted for by ethyl lactate and ethyl acetate. When ethyl lactate/ethyl acetate and ethyl phenylacetate/ethyl 2-methylbutyrate were presented at specific ratios, the aroma was significantly enhanced.²³ Therefore, esters are the most aromatic chemicals in CRW. They are critical compounds for the aroma and flavor of CRW.

2.1.3. Phenols and Aldehydes. The free radical scavenging and antioxidant capacities of CRW are mainly derived from phenolic compounds. The content of phenolic compounds was proportional to the antioxidant capacity. They have also been considered as important aroma compounds in CRW. Since phenols are mainly produced by lignin degradation in raw materials, the concentration of phenols is lower than that of esters.²² The two main phenols are guaiacol and 4-vinylguaiacol. They are commonly found in traditional CRW. They have typical smoky, herbal, and clove smells, and the flavor compounds constitute medicine and smoke of CRW.⁶ 4-Vinylguaiacol is mainly produced by decarboxylation of ferulic acid. Vanillin has a sweet vanilla flavor, mainly produced

during the aging process. The production of vanillin is more complicated. Studies have shown that 4-vinylguaiacol is oxidized to vanillin. The proportion of benzaldehyde and furfural in aldehydes exceeds 85%.²³ Benzaldehyde is produced by oxidation of phenyl methanol; the production of furfural is related to the Maillard reaction.⁸ In the aging process, benzaldehyde and furfural provide almond and caramel aromas for CRW, but if the concentration is too high, it will cause bitterness.²⁴

2.2. Nonvolatile Compounds. **2.2.1. Acids.** In the past, CRW was referred to as “no sour, no taste”.²⁵ However, the acidity of CRW is actually influenced by sugar content and type of acids present.²⁶ Each acid in CRW has a unique flavor. For example, lactic acid imparts a soft sour flavor to CRW, while citric acid provides a fresh and cool taste; succinic acid is salty and bitter; malic acid is sharp and harshly sour; and tartaric acid is bitter and harshly sour.¹⁸ The presence of acids in CRW can enhance its thick mouthfeel and reduce the sweetness and bitterness to balance other flavor components. These acids are primarily produced through microbial metabolisms like alcoholic fermentation, malolactic fermentation, and ethanol oxidation.²³ In the early stage of fermentation, acids can inhibit the growth of some microorganisms and the production of esters.¹⁸ While CRW with low acid content can taste faint, excessive acidity can negatively impact its flavor, leading to rancidity.²⁷ Therefore, the acid content is regulated within the range of 3.0–7.5 g/L. This ensures its role in buffering and balancing the flavor in CRW, while also gradually producing aromatic esters during the storage process.²⁸ However, attention should be paid to the appropriate content range so as not to affect the taste and quality of CRW. Additionally, the amount of free fatty acids in CRW increases with aging time.²³ Most of these fatty acids come from yeast metabolism, but some come from raw materials. While most fatty acids have pungent smells and tastes, CRW is a complex system of various compounds that interact to form its unique flavor;¹³ however, CRW is a complex system composed of various compounds that collaborate to produce a unique flavor.

2.2.2. Amino Acids and Peptides. CRW contains numerous essential amino acids and peptides with rich nutritional value and health benefits, which is why it is called liquid cake.²⁹ The amino acids and peptides are mainly from the degradation of protein in the raw materials,³⁰ so the content and type of amino acids are affected by the protein in the raw materials. During the brewing process of CRW, amino acids not only enhance its flavor by providing fresh, sweet, bitter, sour, salty, and other tastes but also serve as a source of nutrients for microbial growth and reproduction. The remaining amino acids and other flavor components constitute the unique flavor of CRW.²³ Amino acids are divided into good-taste amino acids and bad-taste amino acids. Good-taste amino acids include umami amino acids and sweet amino acids, and bad-taste amino acids include bitter amino acids, sour amino acids, and salty amino acids. Glutamic acid and aspartic acid are umami amino acids, and glycine, serine, proline, alanine, and threonine are sweet amino acids. Arginine, valine, leucine, histidine, and lysine are bitter amino acids. Tyrosine is a sour amino acid, and methionine and cysteine are salty amino acids.³¹ There are many types of amino acids. In addition to good-taste and bad-taste compounds, other amino acids can also affect the flavor of CRW. For instance, phenylalanine can enhance the malty flavor of CRW, while lysine can improve its

aroma and taste. It is worth noting that arginine, ornithine, and citrulline are precursors for the formation of EC, which is a carcinogen. The healthy limit for EC is 100 $\mu\text{g}/\text{L}$.³² Therefore, it is necessary to control the levels of EC in order to ensure its safety.³³

Polypeptides play a crucial role in CRW as functional compounds, serving purposes such as antioxidation and protection of the cardiovascular system.^{34,35} Studies have shown that over 90 peptides in CRW have health benefits. Among these, more than 30 peptides possess a bitter taste. However, only Arg-Pro-Gly, Gly-Val-Val, Ile-Val, Leu-Arg-Leu, Leu-Glu, and Phe-Leu-Leu are solely bitter in taste, while other peptides also have functional properties.³⁶ Glu-Tyr, Thr-Glu, Val-Glu, Val-Asp, Val-Val, and Pro-Glu were among the dipeptides, which provided umami taste for CRW.³⁷ Phe-Pro, Val-Phe, and Gly-Leu have bitter taste.³⁶ It is evident that bitter peptides are plentiful in CRW, and bitterness is one of the flavors present. Excessive bitterness can disrupt the overall balance of CRW, but hydrolyzing the peptides into smaller amino acids and peptides reduces the bitterness. Some studies suggest that treatment of grains with protease can decrease the presence of bitter peptides in CRW.³⁸ Nevertheless, further research is needed to determine whether this reduction affects the nutritional value and health benefits.

2.2.3. Saccharides. The saccharides in CRW are mainly derived from the enzymatic hydrolysis of starch in grains.⁷ These saccharides are essential in creating the distinctive flavor of CRW and also offer health benefits. The saccharide composition in CRW mainly consists of monosaccharides like glucose, Arabic sugar, galactose, and mannose and oligosaccharides like maltose, isomaltose, isomaltotriose, and panose.²⁷ The content of saccharide in CRW varies with different brewing processes and grains, but glucose is the predominant sugar.³⁶ Oligosaccharides and active polysaccharides found in CRW contribute to the intestinal microecological environment, thereby regulating the gastrointestinal function. Several studies have shown that regular consumption of functional oligosaccharides can significantly improve human gastrointestinal function.³⁹ Therefore, consumption of CRW in moderation can promote good health.

3. BREWING OF CRW

3.1. Relationship between Raw Materials and the Formation of CRW Flavor.

3.1.1. Grains. The main grains used to make CRW are rice and glutinous rice. There were 54 flavor compounds in cooked glutinous rice and 69 in rice.⁴⁰ It mainly includes aldehydes, ketones, acids, esters, alcohols, phenols, and heterocyclic compounds.⁴⁰ Some of the flavor compounds in rice and glutinous rice are converted into other compounds during the brewing process, and some of them are involved in the flavor components of CRW, which can bring a harmonious and delicate pleasant aroma to CRW.⁴¹ Grains are the basis for producing flavor compounds in CRW.⁴² The contents of amylopectin, amylose, fat, and protein in grains are different. In the CRW brewing process, starch and protein undergo degradation to produce sugars and amino acids that impact microorganism growth and ultimately influence the CRW's flavor. The ratio of amylose to amylopectin is an important factor affecting starch properties.⁴³ Amylopectin is readily decomposed into monosaccharides and dextrin by amylase, which is conducive to the transformation of sugars into alcohols during fermentation processes. They are primarily regulated by plant cell starch biosynthesis, with

different grains having varying proportions. Therefore, CRW produced with high amylopectin content offers a more pleasant flavor and is conducive to improved liquor yield.⁴³ By selecting different grain varieties to control the starch proportion, one can alter both the taste and liquor yield. The physicochemical properties of red millet, Zhang hybrid millet, glutinous rice, golden millet, broomcorn millet, and japonica rice were compared. It was found that the content of amylopectin in glutinous rice was the highest, while amylose was the lowest.⁴⁴ Therefore, glutinous rice is often used as a raw material for high-quality CRW. Grains such as black rice, corn, and Tartary buckwheat are used as novel brewing materials to cater to different consumers,^{44–47} but there are no more specific studies. In addition, the content of alcohols and esters in CRW added with glutenin, respectively, increased by 18% and 99%. Sensory analysis showed that the content of glutenin in CRW increased its alcohol content, fruity and honey-like aroma characteristics, and fresh, sour, and bitter taste.⁹ To date, there has been a lack of comprehensive research on the impact of glutenin on both flavor compounds and sensory quality in CRW. Red millet has higher protein and fat content than other grains, and this may produce CRW with higher amino acid and free fatty acid content.⁴⁴

3.1.2. Fermentation Starters. Starters are sources of microbes and crude enzymes that provide flavor compounds to CRW as part of the raw materials.⁴⁸ These lead to different flavor characteristics of different starters. The effect of starters on flavor mainly has two aspects: (1) the starters bring their own flavor compounds into CRW, like hexanal, ethyl hexanoate, 1-octen-3-ol, and phenylacetaldehyde⁴⁹ and (2) the microbial diversity in the starters affected the formation of flavor compounds in the fermentation process of CRW.⁵⁰ Among them, the latter is an important influencing factor. Differential compounds of CRW mixed with Wheat Qu, Hong Qu, and Xiao Qu and without Hong Qu were isoamyl alcohol, phenyl ethanol, 2-methylpropyl acetate, 2-methyl-1-propanol, ethyl cetylolate, and ethyl caprate, but the contents of peptides and amino acids in CRW with only Hong Qu were higher.³⁶ Differences in the species and number of microorganisms present in various starters can impact the formation of CRW flavor, as they may break down different raw material components and produce varying metabolites.

The main starters commonly used in traditional CRW are Wheat Qu, Hong Qu, Xiao Qu, and yeast. Wheat Qu is a culture mixture containing yeast, mold, and bacteria, which contains amylase, glucoamylase, protease, and flavor compounds.⁵¹ Microorganisms in Wheat Qu changed with the changes in environment and brewing process.⁵¹ Hong Qu is produced from steamed red yeast rice by solid-state fermentation with *Monascus*.⁵² Xiao Qu, also known as Jiu Yao, is fermented in the open air in the natural environment with rice flour or rice bran as raw materials, adding a small amount of Chinese herbal medicine and appropriate water.⁵³ Xiao Qu is rich in natural microbial resources, so it is often used to screen the dominant strains. Wheat Qu, Hong Qu, and Xiao Qu are produced in a nonsterile environment, so many kinds of microorganisms in these Qu cannot be controlled.⁵⁴ The difference analysis of Qu from Hubei, Sichuan, and Guangxi provinces in China was carried out, and the Shannon index, the average number of reads, the number of OTUs, and the rank abundance curve were analyzed. The results showed that the microbial diversity of Hubei Qu and Guangxi Qu was higher than that of Sichuan Qu.⁵⁵ The higher the microbial

diversity, the more difficult it is to control the flavor of CRW. We collected the dominant bacterial genera of Qu in a variety of different regions and types in Table 2.

In order to improve the flavor quality of traditional CRW, researchers have carried out a lot of strain screening and transformation work. BR14 is considered a probiotic *Saccharomyces cerevisiae* that can significantly increase alcohol content and total acidity. Additionally, cofermentation of BR14 can significantly reduce the urea content of EC formation.⁵⁶ Yuan et al. isolated a yeast strain (YB-12), for which the gene sequence was highly similar to that of *Meyerozyma*, and found that YB-12 could significantly reduce the content of fusel alcohols in CRW.⁵⁷ The aroma-producing *Mucor indicum* strain (ZT018) in CRW starter can cooperate with *Rhizopus oryzae* to produce aroma and improve the aroma quality of CRW by increasing the content of flavor compounds such as phenethyl acetate, isoamyl alcohol, and so on.⁵⁸ Hybrid F23 had better metabolism ability of 2-phenylethanol, short-chain fatty acid ethyl ester, and long-chain fatty acid ethyl ester than diploid yeast, which gave wine a better fruity and mellow taste.⁵⁹ The metagenomics approach analysis of the microorganisms of yeast strains (N85 and XZ11) showed that significant differences were found in the microbial composition and flavor components of CRW after fermentation by the two strains.⁶⁰ For example, compared with *Nakazawaea ishiwadae* (SITCY001), *Wickerhamomyces anomalus* (SITCY601), and *Candida glabrata* (SITCY597), the CRW fermented by *Wickerhamomyces anomalus* (SITCY125) had a stronger ester and alcohol fragrance. SITCY125 fermented CRW taste: fruity, floral, sweet score higher, bitter score lower. It has potential industrial applications in CRW brewing.⁶¹ Although the content of flavor compounds in CRW fermented with artificial starters was lower than in traditional starters, mainly flavor compounds in the two wines were consistent.⁶²

3.1.3. Microbial Diversity. CRW was fermented in an open environment with a variety of microorganisms.⁶³ There may be symbiotic, alternate, parasitic, and antagonistic relationships among them,⁶⁴ and a complex microbial community is gradually formed during the preparation of starters,⁶⁵ which determines the final yield and flavor quality of CRW.⁶⁶ Cai et al. studied the effect of the microbial composition of starters on the flavor of CRW despite regional differences in China. They found that the classification and metabolism of microorganisms in starters greatly influenced the flavor of CRW.⁶⁷ *Lactobacillus* plays a significant role in reducing the acidity of CRW,^{68,69} and the correlation between them and various core flavor compounds was strongest in the late stages of fermentation (5–23 days).⁷⁰ In the early stage of the fermentation processes, *Lactobacillus* and *Saccharomyces* grew rapidly, and lactic acid and alcohol were produced to change the living environment of the microorganisms.⁷¹ Hence, the growth of *Saccharomyces*, *Leuconostoc*, *Pediococcus*, and *Lactobacillus* can help prevent the deterioration of CRW by inhibiting the growth of a variety of harmful bacterial and fungi, including *Klebsiella*, *Staphylococcus*, *Candida*, and *Acetobacter*.⁷¹ In addition, different microorganisms also promote and inhibit each other. There was a negative correlation between the abundances of *Staphylococcus*, *Leuconostoc*, *Pediococcus*, *Pantoea*, *Acetobacter*, *Klebsiella*, and *Saccharomyces* and a positive correlation between the abundance of *Weissella*, *Lactobacillus*, and *Saccharomyces*. Correspondingly, the abundance of *Leuconostoc*, *Pediococcus*, and *Lactobacillus* was negatively correlated with the abundance of *Monascus* and *Candida*.⁷¹

Table 2. Dominant Bacterial and Fungal Genera of Qu in Different Regions and Species in China^a

Microbes	Genus' level	Unknown		Unknown		Hong Qu		Hong Qu		Hong Qu		Wheat Qu		Wheat Qu		Wheat Qu		Xiao Qu		
		Chongming (CMQ), Shanghai, China ¹²⁰	Ningbo (NBQ), Zhejiang, China ¹²⁰	Yichang (YCQ), Hubei, China ²⁰	Jian'ou, (JOHQ), Fujian, China ⁵⁴	Fuzhou (FZHQ), Fujian, China ⁵⁴	Fuqing (FOHQ), Fujian, China ⁵⁴	Gutian (GTHQ), Fujian, China ⁵⁴	Yongchun (YCHQ), Fujian, China ⁵⁴	black Wuyi (BHQ), Fujian, China ⁷⁶	red Wuyi (RHQ), Fujian, China ⁷⁶	Shaoxing city (SXWQ), Zhejiang, China ⁵	Jimo city, Shandong (JCWQ), China ⁵	Nanping city (NPWQ), Fujian, China ²⁴	Xinhua county (XHQQW), Hunan, China ²³					
Bacteria (genus level)		/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	
	<i>Klebsiella</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Acetobacter</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Acinetobacter</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Agrobacterium</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Bacillus</i>	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	<i>Burkholderia</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Erwinia</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Klebsiella</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Lactobacillus</i>	/	+	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Lactococcus</i>	/	/	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Leuconostoc</i>	/	/	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Ochrobactrum</i>	/	/	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Pantoea</i>	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Pedococcus</i>	/	+	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Saccharopolyspora</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Shewanella</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Staphylococcus</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Streptomyces</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Weissella</i>	/	+	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Enterobacter</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Gluconacetobacter</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Alternaria</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
Fungi (genus level)		/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Aspergillus</i>	/	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Aspergillus</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Aspergillus flavus</i>	/	/	/	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	<i>Aspergillus niger</i>	/	/	/	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	<i>Candida</i>	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Eurotiumcyetes sp</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Fusarium pseudisiforme</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Gibberella</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Monascus</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Monascus purpuratus</i>	/	/	/	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	<i>Mucor</i>	/	/	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Pichia</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Rhizopus</i>	+	+	+	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/

Table 2. continued

Microbes	Genus' level	Unknown	Unknown	Unknown	Hong Qu	Hong Qu	Hong Qu	Hong Qu	Hong Qu	Hong Qu	Hong Qu	Hong Qu	Wheat Qu	Wheat Qu	Wheat Qu	Wheat Qu	Xiao Qu
		Chongming (CMQ), Shanghai, China ¹²⁰	Ningbo (NBQ), Zhejiang, China ¹²⁰	Yichang (YCQ), Hubei, China ²⁰	Jian'ou (JOHQ), Fujian, China ⁵⁴	Fuzhou (FZHQ), Fujian, China ⁵⁴	Fuqing (FOHQ), Fujian, China ⁵⁴	Gutian (GTHQ), Fujian, China ⁵⁴	Yongchun (YCHQ), Fujian, China ⁵⁴	black Wuyi (BHQ), Fujian, China ⁷⁶	red Wuyi (RHQ), Fujian, China ⁷⁶	Shaoxing city (SXWQ), Zhejiang, China ⁵	Jimo city, Shandong (JCWQ), China ⁵	Nanping city (NPWQ), Fujian, China ²⁴	Xinhua county (XHXQW), Hunan, China ²³		
	<i>Rhizopus oryzae</i>	/	/	/	/	/	+	/	/	/	/	/	/	/	/	/	/
	<i>Saccharomyces</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Saccharomyces sp</i>	/	/	/	+	+	+	+	+	+	+	+	+	+	+	+	+
	<i>Saccharopolyspora</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Thermoascus</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Trichosporon</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
	<i>Wickerhamomyces</i>	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/

^a+: This dominant bacterial or fungal genus exists in this Qu. /: This dominant bacterial or fungal genus does not exist in this Qu.

Saccharomyces, *Aspergillus*, *Saccharopolyspora*, *Staphylococcus*, *Lactobacillus*, and *Lactococcus* were closely related to the production of amino acids, alcohols, acids, phenols, and esters.⁷² *Saccharopolyspora* has a great influence on amino acid synthesis, fatty acid synthesis, and triglyceride hydrolysis but has little effect on ethanol, higher alcohols, and phenols.⁷² *Saccharopolyspora*, *Staphylococcus*, *Aspergillus*, and *Saccharomyces* are all involved in the formation of esters, acids, and alcohols, which significantly affects the synthesis of esters.⁷² The production of aldehydes and ketones is mainly influenced by molds. Molds promoted glucoamylase, amylase, and protease activities in the brewing of CRW. The highest levels of glucoamylase and protease activity were found in *Aspergillus flavus*.⁷³ Because the enzymes produced by different strains are usually different, various bacteria and fungi interact with each other during the brewing process to create the unique flavor of CRW.⁷⁴ Therefore, we collected the correlation information on various bacteria, fungi, and flavor compounds in CRW (Figure 1), and the information in Table 2 and Figure 1 is drawn as Figure 2. According to the existing data analysis, it was found that *Enterobacter*, *Rhizopus*, *Corynebacterium*, *Burkholderia*, *Saccharomyces*, *Lactiplantibacillus*, *Wickerhamomyces*, *Aspergillus*, *Bacillus*, and *Penicillium* were highly correlated with more flavor compounds. Therefore, we should focus on these 10 microorganisms when regulating the flavor of CRW.

3.2. Innovation in the Brewing Processes. Traditional brewing processes of CRW involve soaking, steaming, fermentation, sterilization, and aging.^{77,78} Traditional brewing processes cannot meet the industrial development of modern CRW, so each brewing stage has different innovative technology (Figure 3). These innovative processes have different advantages and have an effect on the flavor components and quality of CRW (Table 3).

3.2.1. Pretreatment of Grains. Traditional pretreatment of grains mainly includes soaking and cooking. Soaking grains is one of the crucial stages in the production processes of CRW, and the soaking time is as long as 2–3 days.⁷⁹ Soaked grains have a lower gelatinization temperature to cook easily.⁸⁰ However, there is no uniform regulation of soaking time; the standard is the experience of long-term practice. During the soaking stages, the nutrients of the grains are used by microorganisms to proliferate and produce a variety of acids that are helpful for alcohol fermentation to proceed smoothly. This diversity will have an impact on the final flavor and sensory qualities of CRW. Due to the long soaking time in the traditional process, Zhu et al. proposed a vacuum soaking technology, which can complete the soaking process in 1 h.⁸¹ Compared with the traditional soaking method, the content of amino acid nitrogen and acids of the vacuum soaking group was significantly higher than that of the traditional brewing group. However, the main types of volatile flavor compounds in CRW brewed by two different soaking methods were similar.⁸¹ In addition, Wei et al. proposed an innovative process, canceling the rice soaking process, which adds lactic acid bacteria to supplement the total acid. The CRW brewed in this way has an increased content of esters, resulting in a more soft taste and a relatively shorter aging time.⁸² Both of these processes can compensate for the disadvantages of a traditional soaking time that is too long, and different soaking processes can be selected according to different phases in production.

In steaming rice, the gelatinization of starch by heat and water absorption is beneficial to the role of amylase and

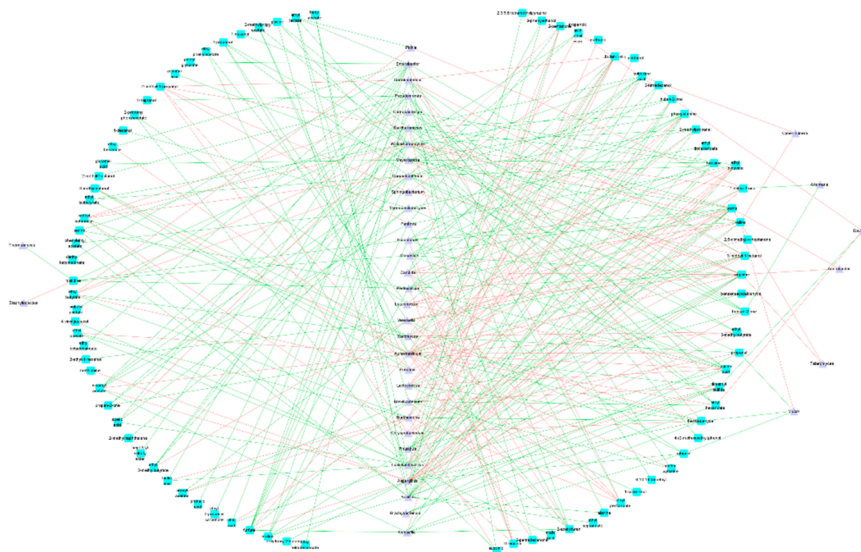


Figure 1. Correlations exist between microorganisms and flavor compounds. Green line: significant positive correlations in references; red line: significant negative correlations in references. Blue square nodes represent flavor compounds, and arrow nodes represent pathways. All the information comes from refs 18, 71, 32, 66, 67, and 72.

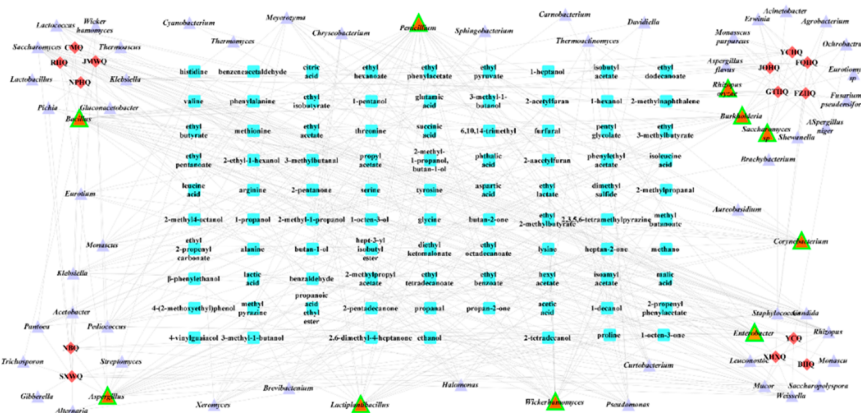


Figure 2. Network diagram of Qu, microorganisms, and flavor compounds. Red diamond nodes represent Qu; blue square nodes represent flavor compounds; and arrow nodes represent pathways. The color from violet to orange depends on the degree value.

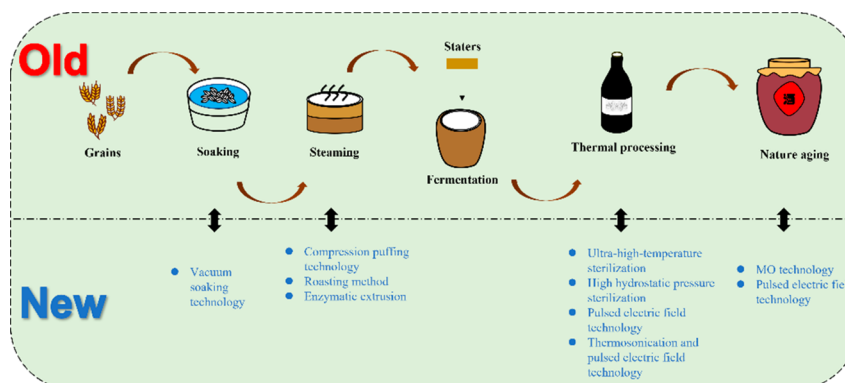


Figure 3. Traditional and innovative brewing processes of CRW.

glucoamylase and can also sterilize the grains.⁶⁷ With the innovation of the brewing processes, there are also compression puffing technology, the roasting method, and enzymatic extrusion in modern production to replace cooking.⁸³ The content of protein and free fat in glutinous rice after puffing technology is reduced, and the content of amino acids

is increased; it can provide ample flavor precursors. However, compared with cooking, the saccharification speed of the roasting method is faster.⁸⁴ The roasting method is to heat the superheated steam to about 200 °C for 20 to 40 s, and the gelatinization degree of rice is the same as cooking. Excessive denaturation of protein at high temperatures will reduce its

Table 3. Compared with Traditional Brewing Processes, the Influence of Innovative Technology on Flavor Compounds and Its Advantages and Disadvantages

Processes	Processes or technologies	Stage of substitution	Advantages or disadvantages	Changes of flavor compounds	References	
Pretreatment of grains	Vacuum soaking technology	Soaking	This technology could shorten the soaking time.	The contents of amino acid nitrogen and organic acid were increased.	81	
	Excluding the rice soaking process	Soaking	This technology could save water resources and improve the security of the CRW.	The content of ester increased.	82	
	Compression puffing technology	Cooking	Increasing raw material utilization and starch gelatinization degree.	Lowered content of amino acids and alcohols.	85	
	Roasting method	Cooking	This technology could save energy and reduce environmental pollution.	Esters, alcohols, and amino acids were increased.	84	
	Enzymatic extrusion	Cooking	It had a higher fermentation rate.	The contents of sugar and amino acid were reduced.	87	
	Uncooked material method	Soaking and cooking	This technology could save energy and reduce costs; however, this method can lead to a thin aroma of CRW, and the taste is not mellow.	No report.	89	
	Liquefaction method	Soaking and cooking	This technology could save energy, reduce emissions, and transport conveniently.	No report.	90	
	Fermentation processes	Simultaneous cofermentation	Sequential cofermentation	The flavor and quality of CRW produced by this technique are lower than those by traditional sequential cofermentation.	The contents of ethyl acetate, ethyl hexanoate, and phenylethyl acetate were increased.	21
		UHT sterilization	Thermal processing	This technology could shorten processing time with minimum quality loss.	Free amino acids and esters were increased.	99
	Sterilization	HHP sterilization	Thermal processing	This technology could shorten the aging time of CRW.	The content of total free amino acids was increased.	99
PEF technology sterilization		Thermal processing	This technology could ensure good product quality and energy use efficiency.	The contents of phenolic compounds and anthocyanins were increased.	100	
TS and PEF technology		Thermal processing	This technology could enhance the bactericidal effect.	No report.	102	
Micro-oxygen technology		Natural aging	This technology could shorten the aging time.	No report.	106	
Pulsed electric field technology		Natural aging	It can shorten the aging time.	No report.	106	

decomposition ability, so the content of amino acid nitrogen will decrease. However, compared with cooking, the CRW made from puffed glutinous rice has lower content of amino acids and alcohols.⁸⁵ Compared with traditional CRW, baked rice wine contains more alcohol, esters, protein, ascorbic acid, amino acids, and total phenols.⁸⁶ Enzymatic extrusion-processed CRW contains more reducing sugars and amino acids.⁸⁷ Compression bulking techniques may reduce the nutrients in CRW, but baking and enzymatic decompression methods can increase the protein and amino acid content. Principal component analysis showed obvious differences in the flavor compounds between enzymatically extruded wine and CRW,⁸⁷ but the flavor differences between these methods have not been fully reported.

In addition, the brewing method of uncooked material and the liquefaction method are brewing methods that eliminate the process of soaking and cooking.⁸⁸ Winemaking with uncooked materials can save energy and reduce costs;⁸⁶ however, this method can lead to a thin aroma of CRW, and the taste is not mellow.⁸⁹ The liquefaction method is a new brewing process that involves adding amylase to crushed raw materials and decomposing amylase into sugars through a liquefaction reaction. This method replaces the traditional process of soaking and steaming rice. The suitable liquefaction conditions are as follows: the crushing degree of raw materials is 27 mesh; the amount of high-temperature enzyme is 5U, and it is liquefied at 100 °C for 6 min.⁹⁰ However, due to limited research, the effect of these two methods on the flavor of CRW was not reported.

3.2.2. Fermentation Process. Saccharification and fermentation are the two phases of the CRW fermentation process.⁹¹ Saccharification is the process of converting starch and protein in grain into sugars and amino acids under the action of starters. Fermentation is the process by which microorganisms convert sugars and amino acids into flavor compounds like alcohols, acids, and esters. Fermentation can be divided into simultaneous cofermentation and sequential cofermentation according to different sequences. Saccharification and fermentation are simultaneous with cofermentation; after saccharification for 1–3 days and then fermentation into sequential cofermentation, different sequences greatly impact the flavor characteristics of CRW.²¹ Although simultaneous cofermentation can save time and cost, it may produce some pungent odors and harmful alcohols that could affect the quality and taste of CRW. Compared with simultaneous cofermentation, CRW brewed by sequential cofermentation contains 3-methyl-1-butanol with a pungent odor, but there are no 3-methyl-1-pentanol and thiols.²¹ The contents of phenyl ethanol and 1-propanol were also higher in sequential cofermentation. This means that it is free of unpleasant odors and harmful alcohols and has a fruity and floral scent. In addition, sequential cofermentation has a strong ability to synthesize higher alcohols. In addition, the content of octanoic acid in sequential cofermentation is low, so the aroma of the wine is more harmonious and balanced.²¹ So sequential cofermentation is superior to simultaneous cofermentation because it prevents the production of pungent odors and harmful alcohols, resulting in improved quality and flavor.

Temperature is also an important factor in determining flavor compounds during the CRW fermentation process. The temperature plays a role mainly by affecting the growth and diversity of the microorganisms, for example: affecting yeast growth to change the fermentation rate⁹² and affecting the

microbial diversity to change the types of acids, amino acids, and other flavor compounds in CRW.⁷¹ The genetic algorithm determined that the temperature for maximizing the ethanol production for CRW fermentation was 26 °C up to 30 h.⁹³ In addition, Liu et al. conducted experiments on the fermentation process of CRW at different temperatures (16, 18, 23, 28, and 33 °C). It was found that the ethanol concentration was the highest at 23 °C and the lowest at 33 °C. Too low of a temperature will slow down the reaction rate, but high temperatures will accelerate the aging of yeast to reduce the production of ethanol. At 33 °C, the concentrations of acetic acid, tartaric acid, and lactic acid increased significantly mainly because high temperatures can accelerate the growth of lactic acid bacteria and lead to the generation and accumulation of lactic acid.⁹⁴ In addition, the temperature and bitter amino acid content were proportional. Under the condition of 30 °C, the concentration of bitter and astringent amino acids was nearly twice as much as those at 20 °C.⁷¹ As a result, the fermentation temperature must be kept in a suitable range.⁹⁵ When the ambient temperature is low, the content of higher alcohols, such as 2-methylpropanol and 3-methylbutanol, decreases, and the content of most ester compounds increases. CRW aroma is also more harmonious.⁹⁶ During fermentation, it is recommended to control the fermentation temperature to about 26 °C.

3.2.3. Sterilization Process. Sterilization ensures the safety and prolongs the shelf life of CRW.³³ The traditional sterilization method for CRW is thermal processing,⁹⁷ and aldehydes can be volatilized in thermal processing to accelerate the conversion of alcohol and nutrients.⁹⁸ However, high temperatures can result in the accumulation of EC, a compound that is known to cause cancer. Li et al. discovered that at temperatures below 85 °C the concentration of EC in CRW was lower than the safe limit of 100 µg/L.³² The content of the carcinogen EC can be reduced by properly lowering the sterilization temperature. Therefore, appropriate low temperatures should be chosen for the traditional heat treatment. Compared to untreated wine, the ethanol and amino acid contents of CRW were reduced after thermal processing, and the loss of flavor components was 4.68–8.61%.⁹⁹ Thermal processing limited the development of CRW due to high nutritive loss and poor wine taste. Therefore, researchers have developed a series of new sterilization technologies to address this problem, which can reduce nutrient and flavor component loss while maintaining safety and extending shelf life. The following sections introduce several common new sterilization technologies.

Ultra-high temperature (UHT) is another new sterilization method that can deal with food that needs to be sterilized in a short time. The flavor characteristics of UHT-treated wine are similar to those of traditionally thermal processed wine,⁹⁹ both of which result in nutrient loss. The advantage of UHT over traditional thermal processing is that it processes faster and has less nutrient loss to the food. However, UHT processing also has some disadvantages, such as complex operation and a high equipment cost. The content of free amino acids and esters in CRW treated with high hydrostatic pressure (HHP) sterilization increased. The optimal HHP treatment (400 or 600 MPa for 20 min) had little effect on the flavor characteristics of CRW, maintained the amino acid content in CRW, and mainly presented a fruity aroma.⁹⁹ After HHP treatment, the content of flavor components in yellow rice wine increased by 7.35%, which effectively improved the

quality of the wine and made it a feasible sterilization method.⁹⁹ Pulsed electric field (PEF) technology is also a nonthermal process, which is often used to inactivate microorganisms in liquid food.¹⁰⁰ PEF treatment with CRW can effectively inactivate spoilage yeast in CRW, and it has a positive effect on the quality indices of CRW.¹⁰¹ In addition, although thermosonication (TS) (35 °C, 750 W, 120 min) could not completely inactivate *Saccharomyces* in CRW, the combination of TS and PEF had additive effects on *Saccharomyces* inactivation.¹⁰² However, future studies should focus on differences in the flavor components and sensory evaluation of CRW treated by PEF and TS, as there is currently a lack of research in this area.

3.2.4. Aging Process. Aging is the process of sterilizing young CRW in sealed pots at room temperature for more than one year.¹⁰³ Air can slowly infiltrate into the ceramic, accelerating the oxidation of alcohols and aldehydes to organic acids, followed by the hydrolysis and esterification of organic acids. With the increase of time, alcohol and amino acid nitrogen levels gradually decreased, and more nutrients increased.³³ In addition, the overall flavor characteristics of aged CRW are more complex.¹⁰⁴ The difference in flavor between young and aged Guyue Longshan CRW was studied by using a comparative dilution method of aroma extraction. It was found that the contents of 10 flavor compounds in aged CRW were 5–56 times higher than those in young CRW.¹⁰³ Only the contents of 3 flavor compounds (including 4-vinylguaiacol, methional, and 2,3-butanedione) were lower than those of young CRW.¹⁰³ As the aging time increases, the difference between furan caramel flavor and the fruity, floral, and honey aromas from esters and benzene becomes greater, and the fragrance is intensified by the rising tendency of acids, aldehydes, and ketones.¹⁰⁵ The high cost of CRW natural aging equipment, difficult management, and long-term defects need to be compensated for. So, the aging process of CRW can be accelerated by using microoxygen (MO) and pulsed electric fields (PEF). In a short time, the sweetness and MSG of CRW treated with MO and PEF (0.35 mg L/day or 0.5 mg L/day for 60 days) reached natural aging levels and reduced the content of bitter amino acids.¹⁰⁶ These two techniques have a significant positive effect on the sensory and flavor characteristics of CRW. In addition, the content of 2-methyl-1-propanol, 3-methyl-1-butanol, phenyl ethanol, and aromatic compounds can also reach natural aging levels, but MO combined with PEF can enhance the flavor intensity of CRW.¹⁰⁶ These two techniques can simulate the effects of natural aging in a short time, which can be applied to CRW aging.

4. SAFETY AND IMPROVEMENT METHODS OF CRW

4.1. Safety of CRW. CRW can increase the activity of antioxidant enzymes and decrease the content of malondialdehyde in the brain and liver of aging mice by using D-galactose.¹⁰⁷ It is also possible to reverse the cognitive dysfunction of mice and reduce the apoptosis of nerve cells by regulating the expression of Bax/Bcl2 and caspase-3 genes.¹⁰⁸ However, the presence of an EC in CRW has raised widespread concerns about food safety. EC is mainly produced by the reaction of urea and citrulline with ethanol under high-temperature conditions (sterilization process) in CRW. Urea and citrulline are intermediate products of arginine degradation, which is a main amino acid in CRW. In addition, urea and citrulline can also be produced by *Saccharomyces cerevisiae* and other metabolic processes.¹⁰⁹ Therefore, we can consider

reducing the EC content in CRW by eliminating the precursor substances of EC and changing the starter culture.

4.2. Improvement Methods of CRW. The elimination of EC precursors is enzymolysis, and these two enzymes are acid urease and ethyl carbamate. Acid urease reduces the formation of EC by eliminating urea in CRW. Acid urease exhibiting high tolerance toward ethanol and acid is an optimal enzyme for such applications.¹¹⁰ Yang et al. cloned acid urease gene cluster *ureABCEFGD* from *Lactobacillus reuteri* CICC6124, which produced acid urease that could eliminate about 95.8% urea in CRW.¹¹¹ Although many acidic ureases have been commercialized, their use in food is still limited due to security issues related to the need for Ni²⁺ binding for urea elimination. However, Liu et al. discovered that the urease of *Bacillus parachliteniformis* ATCC 9945a binds with Fe³⁺ instead of Ni²⁺, making it a viable option for use in the food industry.¹¹² Carbamate esterase is the direct decomposition of EC into ammonia, ethanol, and carbon dioxide. It has a higher safety when used in fermented food. *Lysinibacillus fusiformis* SCO2 urethanase exhibited a promising potential in degrading EC present in soy sauce and CRW.¹¹³ In addition, *Oenococcus oeni*, *Lactobacillus brevis*, and *Lactobacillus plantarum* can produce carbamate esterase during coculture with *Saccharomyces cerevisiae*.¹¹⁴ Therefore, acid urease can be used in the brewing process of CRW to eliminate urea and to reduce EC content accumulation before sterilization. After sterilization, carbamate esterase is used to degrade EC in CRW.

Besides, different starters are also responsible for the different EC contents in CRW. *Lactobacillus brevis* 2–34 exhibits a strong citrulline reuptake ability, and the EC content in CRW can be controlled by reducing the citrulline content. Inoculating CRW with this strain resulted in reduced concentrations of citrulline, EC, and several harmful higher alcohols.¹¹⁵ In addition, genetic modification of the strains is also an option. Wu et al. used the improved CRISPR/Cas9 system to genetically modify the strain. After modification, it was shown that the modified strain reduced urea concentration by 92.0% and EC concentration in CRW by 58.5%, respectively. Furthermore, in repeated brewing experiments, the genetically modified strain demonstrated good genetic stability.¹¹⁶ The EC synthesis was inhibited by adding gallic acid and protocatechuic acid in the fermentation process of CRW, and the growth of *Saccharomyces* was not affected. The addition of gallic acid and protocatechuic acid reduced EC by up to 91.9%.¹¹⁷ However, under cofermentation conditions, adding gallic acid could not reduce the content of ethyl carbamate.¹¹⁸

5. PROSPECTS AND CONCLUSIONS

CRW is brewed from grain and Qu as the raw materials. CRW's unique raw materials and complex brewing processes endow it with a rich taste and aroma. The precursors of the CRW flavor compounds come from grains and starters. The flavor compounds of CRW are affected by microbial diversity and the brewing processes. In the future, CRW flavor can be precisely regulated from the following aspects: (1) the selection of grain and starters; (2) screening and modification of excellent strains; (3) the correlation between microorganisms and flavor compounds; (4) the pretreatment of grains and fermentation process selection; and (5) sterilization and aging technology selection. However, the formation of CRW flavor compounds is a complex process. The interaction of various flavor compounds also determines their final flavor

characteristics; therefore, the quantitative study of the intensity of flavor compounds in CRW is also needed for CRW flavor regulation and to comprehensively understand the flavor compounds and aroma description of CRW and the influence of various factors on the flavor compounds of CRW. Various methods can be employed to reduce the high EC content in CRW and improve its quality, but studies on the elimination of EC in CRW lack information regarding its effect on flavor. Future research could investigate whether reducing the EC content also affects flavor. It is of great significance to the development of CRW, the evaluation of CRW flavor, and the improvement of its quality. The study of flavor compounds is conducive to further promote the diversified development of CRW and satisfy different consumer groups.

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Notes

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