

桑葚酒生物活性成分及香气品质研究进展

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桑葚酒生物活性成分及香气品质研究进展

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摘要: 桑葚是一种传统的药食两用水果, 具有良好的抗氧化、抗炎和抗癌等功能特性。用桑葚果实发酵制成的桑葚酒因其丰富的活性物质和独特的风味表现, 近年来受到消费者的密切关注。本文结合当前国内外桑葚酒的研究现状, 从生物活性成分的角度综述了桑葚酒的健康功效, 总结并探讨了桑葚酒的香气物质组成以及影响桑葚酒香气品质的关键因素, 并对未来桑葚酒的研究方向进行展望, 以期桑葚酒的理论探究和产品开发提供新思路, 推动桑葚酒的标准化和产业高值化发展。

关键词: 桑葚酒, 生物活性成分, 香气物质, 影响因素

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Research Progress on Bioactive Components and Aroma Quality of Mulberry Wine

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Abstract: Mulberry has a long history of use as an edible fruit and medicine. Studies have shown that mulberry possesses several functional characteristics including antioxidation, anti-inflammation and anticancer. In recent years, mulberry wine has gained much attention from consumers due to its abundant bioactive components and distinctive sensory attributes. Based on the latest research, this review summarizes the health effects of mulberry wine from the perspective of bioactive components, clarifies the aroma composition of mulberry wine, discusses the main factors affecting the aroma quality of mulberry wine and prospected the future research trends. It is expected to provide new ideas for theoretical study and product development of mulberry wine, which will be helpful to promote the quality standardization of mulberry wine and enhance the economic value of mulberry wine industry.

Key words: mulberry wine; bioactive components; aroma substances; influence factors

桑葚 (*Fructus mori*) 是桑科桑属植物的成熟果实, 主要种植于全球温带和亚热带地区^[1]。研究表明, 桑葚富含氨基酸、维生素、矿物质等营养成分和花色苷、多糖、黄酮等活性物质, 具有抗氧化、降血糖、提高免疫力、改善睡眠等多种功效, 被称作“民间圣果”, 具有极高的营养和药用价值^[2-6]。桑葚酒是桑葚果实经破碎压榨后发酵制成的低度酒饮 (酒精度 7% vol~15% vol), 可以有效保留桑葚果实中的营养

物质, 呈现出的抗氧化活性显著高于葡萄酒^[7], 符合现代消费者对于健康养生的需求, 是一种极具发展潜力的果酒品类。香气是评价桑葚酒品质最重要的指标之一^[8], 香气的浓郁程度、协调性及典型性等均会直接影响到消费者的购买行为。

目前围绕桑葚酒的研究主要集中在香气化合物组成、酿造工艺和抗氧化性评价上, 如通过筛选酿酒酵母和非酿酒酵母, 以获得适合桑葚酒发酵的优质菌

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种^[9];通过响应面试验优化发酵温度、发酵时间、初始含糖量等参数以获得桑葚酒的最佳工艺条件^[10];通过鉴定发酵过程中香气成分变化,以揭示桑葚酒的风味组成和变化规律^[11]等。本文从生物活性成分的角度阐述了桑葚酒的功能特性,总结了桑葚酒的香气物质组成并探讨了影响桑葚酒香气品质的关键因素,旨在为桑葚酒的品质调控和产品研发提供科学依据及理论参考。

1 桑葚酒生物活性成分

研究表明桑葚酒的白藜芦醇含量、抗氧化活性等显著高于红葡萄酒^[12],因此桑葚酒被视为绝佳的健康饮品。现将桑葚酒中主要生物活性成分归纳于表 1。

1.1 类黄酮及其衍生物

类黄酮是广泛存在于植物中的一种酚类化合物,以 1 个吡喃环连接 2 个芳香环为骨架特征,由于取代基不同可被分为花色苷类、原花青素类、黄烷醇类、黄酮醇类、黄酮类、查尔酮类^[13]。其中,桑葚酒、葡萄酒等果酒中的主要类黄酮类化合物是花色苷类、黄酮醇类和黄烷醇类^[14]。

花青素是一种水溶性类黄酮化合物,在植物体内起协同呈色、抗紫外线、抗氧化和抗真菌作用。桑葚酒中的花青素多以糖基化形式的单体花色苷存在,主要是矢车菊素-3-O-葡萄糖苷(C3G)和矢车菊素-3-O-芸香糖苷(C3R)两种花色苷,二者对桑葚酒的抗氧化性贡献极大^[14]。在桑葚酒发酵过程中,上述 2 种单体花色苷含量一般呈现先上升后下降的变化趋势^[15],特别是发酵后矢车菊素-3-O-葡萄糖苷的含量低于发酵前,这可能是发酵菌种将矢车菊素-3-O-葡萄糖苷转变为吡喃花色苷和聚合花色苷^[16],或是间接代谢形成原儿茶素,Gao 等^[17]提供的原儿茶素代谢途径印证了这一解释。

除花色苷外,桑葚酒中还检出槲皮素、二氢槲皮素、芦丁等黄酮醇物质,总含量高于发酵前,表明发

酵过程中黄酮醇物质不断溶出,且溶出速率高于其分解速率^[35]。其中,芦丁(又名“槲皮素-3-O-芸香糖苷”)是桑葚酒中一种重要的黄酮醇,通过抑制脂肪酶活性来降低血糖,还有抗氧化、抗高脂血症等功效^[36]。

1.2 酚酸及其衍生物

桑葚酒中的酚酸按其碳骨架结构分为对羟基苯甲酸衍生物和对羟基肉桂酸衍生物两大类,主要以糖苷和酯的共轭形式存在^[37]。苯甲酸型以没食子酸、原儿茶酸和香草酸为主,其中原儿茶酸被证实有积极的抗炎活性,可通过调节体内的巨噬细胞来减轻高胆固醇饮食诱导小鼠的动脉粥样硬化^[24],还可增强多种抗氧化酶活性并直接捕捉自由基以降低机体氧化损伤^[25];肉桂酸型包括咖啡酸、*p*-香豆素、阿魏酸、绿原酸等^[38],在抗炎、抗菌、抗癌、抗氧化等健康效益的贡献不容忽视^[17]。在桑葚酒发酵过程中,总酚酸含量始终维持在 60~75 mg/L 的水平,两类酚酸化合物在酵母生长及代谢产物的影响下相互转化,其中绿原酸被大量降解,直至发酵结束时,羟基苯甲酸衍生物的含量为羟基肉桂酸衍生物的两倍以上^[37]。

1.3 其他健康功能成分

桑葚多糖是一类功能性多糖,主要由鼠李糖、阿拉伯糖、半乳糖、葡萄糖等单糖脱水缩合而成,其含量和种类因桑葚品种不同具有一定差异性。郑若欣等^[39]通过醇沉-苯酚硫酸法测得桑葚酒中多糖含量为 127.58 mg/L。研究表明桑葚多糖通过抑制蛋白酪氨酸磷酸酶 1B 表达,激活 PI3K-Akt 通路,减轻由高脂肪和链脲佐菌素诱导的二型糖尿病大鼠肝脏中的氧化应激,有效地调节葡萄糖代谢并使胰岛素信号传导正常化^[31]。此外,桑葚多糖还有降低炎症因子表达、改善胰腺功能等作用^[40]。

经过发酵的桑葚酒可以溶解释放果实中 50% 以上的白藜芦醇^[41]。白藜芦醇是一种抗菌性含芪类结构的多酚化合物,近年来被证实在抗心血管疾病、抗

表 1 桑葚酒中主要生物活性成分

Table 1 Main bioactive components in mulberry wine

分类	名称	生物活性	参考文献
类黄酮	矢车菊素-3-O-葡萄糖苷	协同呈色、抗紫外线、抗氧化、抗真菌等	[13-18]
	矢车菊素-3-O-芸香糖苷		
	芦丁		
酚酸	槲皮素	抗氧化、抗糖尿病、抗高脂血症、抗肥胖等	[19]
	二氢槲皮素	抗氧化、抗菌、抗病毒、抗炎、抗癌等	[20-21]
	没食子酸	抗氧化、抗肿瘤、阻止细胞凋亡、抗纤维化、抗过敏等	[22]
	原儿茶酸	抗乙型肝炎、保护肝脏免受损伤等	[23]
	香草酸	抗炎、减轻动脉粥样硬化、降低氧化损伤等	[24-25]
	咖啡酸	抗炎、抗糖尿病、保肝、抗高血压等	[26]
	对香豆酸	抗炎、抗菌、抗氧化、延缓衰老等	[27]
	阿魏酸	清除自由基、抑制脂质过氧化、免疫调节、抗血小板聚集等	[28]
	绿原酸	抗菌、消炎、治疗记忆障碍等	[29]
	多糖	桑葚多糖	抗炎、降压、降血糖、改善母乳品质等
其他	白藜芦醇	调节葡萄糖代谢、抑制炎症反应、改善胰腺功能等	[31]
	褪黑素	抗菌、抗肿瘤、抗炎等	[32-33]
		清除自由基、调节昼夜节律、增强免疫等	[34]

炎症和抗氧化等领域具有发展潜力^[28]。Cheuk 等^[33]研究发现白藜芦醇的使用可显著提高乳腺癌细胞增殖抑制率,同时降低肿瘤微环境中促炎细胞因子白细胞介素-6 水平,提高乳腺癌细胞对顺铂药物的敏感度。

研究还发现桑葚酒中富含褪黑素,即 N-乙酰-5-甲氧基色胺^[34]。褪黑素是一种通过色氨酸途径合成的色胺类激素,具有调节昼夜节律、延缓衰老、清除自由基、增强免疫等功能^[30]。Wang 等^[42]发现在桑葚鲜果和桑葚酒发酵过程中均检出较高水平的褪黑素,并且褪黑素含量变化与乙醇产率显著相关,说明褪黑色素可能参与了发酵过程中酿酒酵母的生理调节。

2 桑葚酒香气品质

2.1 桑葚酒香气物质组成

香气化合物是桑葚酒产品典型性的重要表征,影响着桑葚酒产品的可接受度和受欢迎度。桑葚酒的香气物质主要来源于桑葚果实、发酵和陈酿过程,Chen 等^[43]指出桑葚果实中约有 55%~65% 的风味物质以无味的结合态糖苷形式存在,酒精发酵有助于结合态化合物转化为游离态,充分释放桑葚果实的香气潜力,同时通过微生物代谢生成新的香气物质,从而获得更丰富的风味表现。此前的研究证实了这一观点^[44]——桑葚鲜果可被检出的挥发性风味化合物约有 30~40 种,而桑葚酒的香气成分大大增加,约有 120~130 种,大致可分为酯类、醇类、酸类、酚类、醛酮类和萜烯类等,如图 1 所示。通常在桑葚酒中酯类化合物种类最丰富,约占总香气成分的 40%~60%,其次是醇类化合物,约占 5%~20%,酸类化合物约占 1%~3%,醛酮类化合物约占 1%~20%;酚类和萜烯类化合物含量较少,约占 1%^[45-47]。

种类多且含量高的酯类和醇类化合物赋予了桑

葚酒浓郁的花香和果香。桑葚酒中常被检出的乳酸乙酯具有凤梨香、己酸乙酯具有菠萝香、乙酸异戊酯具有香蕉香、辛酸乙酯具有杏子香、癸酸乙酯具有葡萄香,异戊醇呈淡淡的青草香、苯乙醇呈紫罗兰香^[48]。适量的酸类物质会给桑葚酒带来入口时的清爽特性并保持酒体平衡,桑葚酒中检测到的酸类化合物以乙酸、辛酸、己酸为主^[46]。在其他酒类研究中,醛酮类化合物被认为对果酒香气起到不可忽视的协调作用^[49]。桑葚酒中存在的醛酮类物质有苯甲醛(坚果香)、癸醛(柑橘香)、2,3-丁二酮(乳香)等^[47]。萜烯类物质主要由桑葚果实中存在的香气前体物经酵母代谢或水解作用转化而成,往往感官阈值较低,因而会对桑葚酒风味作出重要贡献。桑葚酒中常见的萜烯类化合物有 4-萜品醇、里那醇、香茅醇等,会赋予桑葚酒更多的甜香、清香^[45]。

近期关于桑葚酒的香气研究除了借助气质联用手段定性并定量桑葚酒的香气化合物组成外,还会通过主成分分析(PCA)、气味活性值(OAV)、香气贡献率(ROC)等揭示各香气物质对桑葚酒主体香气的贡献程度。如孙佳懿等^[50]通过主成分分析鉴定出桑葚酒关键香气物质为苯乙醇、十六酸乙酯、辛酸、癸酸乙酯、苯甲酸乙酯、辛酸乙酯、壬醛。Chen 等^[51]以 OAV 值大于 1 为指标筛选桑葚酒中的主要呈香成分,结果表明 2,3-丁二醇、大马士酮、3-甲基-1-丁醇、壬酸乙酯、乙偶姻等构成了桑葚酒的特征香气。需要指出的是,不同的研究中对于桑葚酒关键香气成分的存在较大差别,目前尚没有对桑葚酒的整体香气特征形成统一的描述,这可能与香气萃取方法、检测手段不同等因素有关,因此未来应考虑继续开展桑葚酒特征香气物质及典型性风味研究。

2.2 影响桑葚酒香气品质的关键因素

2.2.1 发酵菌种 发酵菌种作为酿造过程中最活跃的影响因素^[52],通过各种代谢途径影响发酵进程和发酵产物,对于桑葚酒的香气塑造至关重要。

自然环境来源的酵母是优质发酵菌种的潜在资源。曹倩雯等^[53]从桑葚自然发酵汁中多级筛选得到 1 株发酵性能良好的酿酒酵母(*Saccharomyces cerevisiae*),用该菌发酵的桑葚酒中辛酸乙酯、 β -大马士酮、乙醛二乙缩醛含量远高于阈值,花果香浓郁,所有香气物质的总 OAV 值高于其他试验菌株。刘宜睿等^[54]从桑葚鲜果表皮中筛选到库德毕赤酵母(*Pichia kudriavzevii*)和有孢汉逊酵母(*Hanseniaspora uvarum*),在发酵桑葚酒产香方面性能差异显著。边名鸿等^[55]利用从桑园土壤中筛选得到的 1 株异常威克酵母(*Wickerhamomyces*)与商业酿酒酵母混合发酵可以提升桑葚酒中的总酯含量(3.21 g/L)。此外也可考虑从其他发酵食品中筛选适应桑葚酒发酵的菌种。谭霄等^[56]用从四川泡菜中分离得到的一株产 γ -氨基丁酸酿酒酵母酿制桑葚酒,产生的挥发性物质种类明显优于商业活性干酵母,其中己酸甲酯

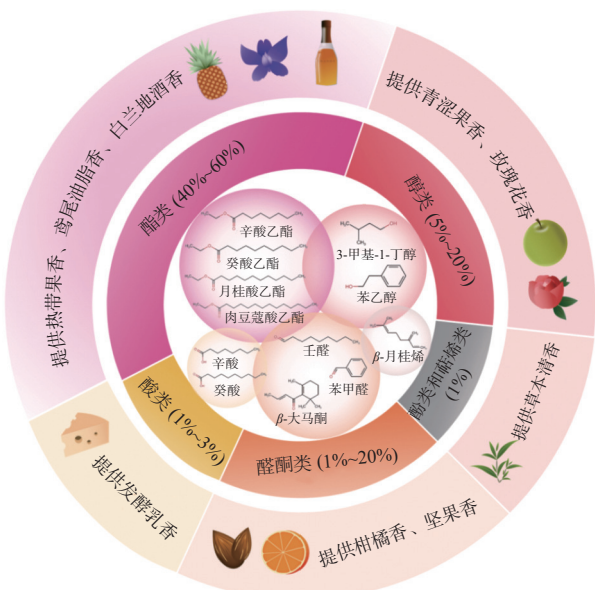


图 1 桑葚酒的挥发性香气化合物组成^[45-47]

Fig.1 Volatile aroma compounds composition of mulberry wine^[45-47]

(菠萝香)、甲酸异戊酯(甜水果香)为该酵母酿造桑葚酒所独有。由此可见,深入挖掘并筛选野生酵母是培育桑葚酒专用酵母、构建优良菌种库的必要手段。

对比评价目前市售的商业酵母是另一种菌株筛选途径。商业酵母纯度高、活性好,适应大多数果酒的发酵环境,通常能迅速生长成为主导发酵过程的优势菌群,在果酒发酵的稳定性、安全性和高效性方面表现较好^[57]。不同商业酵母对桑葚酒香气品质有不同贡献。孔燕等^[58]发现来自法国 LAFFORT 的酿酒酵母 F15 发酵桑葚酒中挥发酸含量低于国标限量(0.45 g/L),感官综合评分较高。Ouyang 等^[59]发现意大利 Enartis 系列商业酵母 RV 增强了桑葚酒的果香。

另外,目前已有研究者通过基因编辑技术对酿酒酵母进行分子改造,在保留高效发酵能力的同时改善产香代谢能力^[60]。徐佳等^[61]对自然发酵桑汁中筛选的酿酒酵母进行 *BAT2* 等位基因敲除,该基因编码酿酒酵母高级醇代谢途径中氨基酸转氨酶的表达,最终获得的重组酿酒酵母有效降低了桑葚酒的高级醇含量。但是,基因编辑菌株目前还没有被列入国家新食品原料目录,其市场化应用也处在技术累积阶段。

2.2.2 接种方式 传统的自然发酵由于受制于环境微生物,发酵常常面临着质量波动大、发酵进程难以预估的风险。接种单一菌种发酵虽然稳定度高,但会导致酒体缺乏风格,风味寡淡^[62]。因此随着对发酵菌种的研究不断深入,多菌种混合接种成为最常见的发酵方式。

采用酿酒酵母和非酿酒酵母混合发酵能极大改善酒体风味缺陷,增强桑葚酒的香气复杂性^[63]。叶片等^[64]使用酿酒酵母、贝型酵母(*Saccharomyces bayanus*)与戴尔有孢圆酵母(*Torulasporea delbrueckii*)共发酵,发现花果香调显著提升且香气化合物种类增多。许强等^[65]的研究也表现出相似的结果,使用异常威克汉逊酵母与酿酒酵母进行共发酵后,桑葚酒的挥发性组分增加了 6 种并赋予其更多的坚果和水果风味。

2.2.3 发酵温度 发酵温度显著影响酵母的生长代谢,直接影响到果酒的口感和品质。若温度过高,酵母的生长活力受到抑制,因而会过早地衰亡导致发酵结束^[66]。低温发酵可以有效控制高级醇^[67]、挥发酸^[68]的产生,且能够影响菌种代谢通路产生复杂的香气化合物,是大多数果酒提升香气的最佳发酵方式,如酿酒酵母和库尔德酵母混合在 12 °C 下能通过除 Ehrlich 途径以外的其他补偿途径(如磷酸戊糖途径)应对寒冷胁迫并产生丰富的风味酯类化合物^[69];此外,温度还影响发酵微生物种群结构并调节菌群演替进程,对果酒风味的塑造产生重要作用^[70]。因此,低温发酵是目前果酒酿造中普遍采用的一种策略。但过低的温度条件会降低酵母的生长能力,不利于酒精的积累^[66]。现有研究表明桑葚酒在 22~28 °C 这一温度区间内进行发酵时香气品质最佳^[71-74]。

2.2.4 外源添加物 在发酵过程中添加合适的辅料同样对提升桑葚酒香气品质发挥着作用。酵母可同化氮是酵母菌可以直接利用的氮源,包括氨基酸、铵离子、小分子肽等,当环境中氮源不足时添加氮源有利于酵母菌的生长繁殖。谢克英等^[75]研究发现在桑葚酒发酵体系中加入 150 mg N/kg 的硫酸铵可以降低异戊醇、苯乙醇等高级醇的生成量。高级醇是桑葚酒中常见的一类香气化合物,可以与酸类物质生成酯,但果酒中高级醇含量一般不超过 400 mg/L,含量过高则会造成辛辣刺激的口感,因此高级醇对于桑葚酒风味的塑造具有显著影响^[67]。此外,在桑葚酒发酵起始阶段添加单一氨基酸会显著提高桑葚酒中挥发性化合物的总含量,并且不同氨基酸的增香效果也有差异,如添加苯丙氨酸会增加酒中苯乙醇、乙酸苯乙酯的含量,这两种物质呈玫瑰花香,而添加缬氨酸会增加酒中乙酸异丁酯(花果香)的含量^[76]。因此,在桑葚酒发酵阶段适量添加辅料也是提升桑葚酒香气品质的重要手段。

3 总结与展望

近年来我国果酒产业呈现多样化的发展态势,加上“健康经济”、“颜值经济”的兴起,香气浓郁、营养丰富的桑葚酒正受到越来越多的关注。在未来桑葚酒开发领域,仍需加强以下几方面的研究:a.深入探究桑葚酒对人体健康和生理功能的作用机制;b.进一步完善桑葚酒香气分析方法,在明确桑葚酒香气物质组成的基础上进行香气重组和缺失验证实验;c.筛选并驯化性能相兼容的产地野生菌种实现产业规模的混菌发酵,以期酿造出具有各产地风土特色的优质桑葚酒;d.建立桑葚酒品质评价体系,推动桑葚酒的标准生产。

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