fermentation

MDPI

Article

The Implications of Composite Dark Purple Rice Malt on Phenolic Acid Profiles, 4-Vinyl Guaiacol Reduction and Enhancing the Antioxidation of Beer

Hellie Gonu [1,2] and Ulaiwan Withayagiat [1,2,*]

[1] Department of Biotechnology, Faculty of Agro-Industry, Kasetsart University, Chatuchak, Bangkok 10900, Thailand
[2] Fermentation Technology Research Center, Faculty of Agro-Industry, Kasetsart University, Chatuchak, Bangkok 10900, Thailand
* Correspondence: fagiulw@ku.ac.th; Tel.: +66-844-306-742

Abstract: This study highlights the dynamics of phenolic acids, antioxidation, and 4-vinylguaiacol in beer produced with dark purple rice malt, also known as Riceberry rice malt, as an adjunct and base malt. Riceberry rice malt substituted barley malt at 40% (*w*/*w*), 60% (*w*/*w*), 80% (*w*/*w*), and 100% (*w*/*w*) with 100% (*w*/*w*) barley malt as the control. Two types of ale beer were produced with two yeasts, designated as POF^- and POF^+. The wort produced with the Riceberry rice malt had higher anthocyanin and vanillic acids relative to all barley malt wort. Fermentation and beer maturation reduced phenolic acids and antioxidant activity in all treatment. Nevertheless, beer produced from 40% (*w*/*w*)–80% (*w*/*w*) Riceberry rice malt maintained higher p-coumaric acid, vanillic acid, anthocyanin, and antioxidant activity in beers with lower 4-vinylguaiacol relative to all barley malt beer, which also had higher ferulic acid and sinapic acid contents. The beers made from POF^+ yeast contained more 4-vinylguaiacol contents than those found in beers made from POF^- yeasts. This study suggests that Riceberry rice malt or POF^- yeast are suitable raw materials for phenolic acid off-flavour reduction and the stabilisation of antioxidant activity in beer.

Keywords: Riceberry rice; *p*-coumaric acid; ferulic acid; 4-vinyl guaiacol; antioxidant activity

Citation: Gonu, H.; Withayagiat, U. The Implications of Composite Dark Purple Rice Malt on Phenolic Acid Profiles, 4-Vinyl Guaiacol Reduction and Enhancing the Antioxidation of Beer. *Fermentation* **2022**, *8*, 392. https://doi.org/10.3390/fermentation8080392

Academic Editors: Vinko Krstanović, Krešimir Mastanjević and Kristina Mastanjević

Received: 30 June 2022
Accepted: 3 August 2022
Published: 15 August 2022

1. Introduction

Phenolic acids are often referred to as aromatic compounds with carboxylic functional groups, which form an integral part of cereal chemical structure and play an important role in antioxidation activity. Phenolic acids consist of two groups: hydroxycinnamic acids and hydroxybenzoic acids. The hydroxycinnamic acids comprise ferulic acids, *p*-coumaric acids, and sinapic acids [1], which are the precursors for volatile phenolic compounds in non-alcoholic beverages (fruit juices and coffee) and alcoholic beverages (wine and beer) [2]. In beer, the most profound volatile phenolic compound is 4-vinylguaiacol (4-VG), which is derived from the decarboxylation of ferulic acids [3]. In the beer brewing process, hydroxycinnamic acids solubilise in wort during mashing and are decarboxylated during the boiling of wort. High amounts of decarboxylated hydroxycinnamic acids occur during wort fermentation by yeast activity [2,3]. Yeast capable of decarboxylating phenolic acid is designated as a phenolic off-flavour producer (POF^+). This yeast possesses the phenylacrylic acid decarboxylase (PAD1) and ferulic acid decarboxylase (FDC1) gene [4]. In addition, the non-decarboxylating phenolic acid yeast is classified as a non-phenolic off-flavour producer (POF^-) [5]. Recently, the PAD1 has been renamed as flavin prenyl-easterase, which produces a modified flavin cofactor prenylated FMNH2 for the ferulic acids decarboxylase (FDC1) enzyme activity [6]. Ogata et al. [7] suggested that the mutation of PAD1 and/or FDC1 caused *Saccharomyces cerevisiae* to lack the ability to produce 4-vinylguaiacol from ferulic acid, which is preferred by German and English beer styles, whereas in wheat beer

and Saison beer, 4-vinylguaiacol is a key aroma for its organoleptic character. However, the amount of hydroxycinnamic acid in beer is dependent on the type of raw material, brewing process, and the amount of hydroxycinnamic acid decarboxylated by yeast. Therefore, the phenolic acid profile in the composite wort and their transformation by brewing yeast and the brewing process were investigated in this study.

The current surge of craft brewing has had a significant influence on the choice of raw material to obtain diverse organoleptic properties of beer. Moreover, the use of other malted cereals such as rye, oat, rice, sorghum, and buckwheat in substituting barley malt affects phenolic acid profiles, the amount of volatile phenolic aroma compounds produced, and antioxidation [2]. Among these cereals, rice is used globally as an adjunct for beer brewing except in countries such as Germany, which restricts the use of other cereals apart from barley.

In Asia, rice is an integral part of many beers produced. The prospects of rice as an adjunct in beer brewing and as a base malt for the production of gluten-free beer have been researched exhaustively with reports on malt attributes, organoleptic properties [8,9] coupled with the study of specialty rice malt, and functional properties such as antioxidation and total phenolic acid in pigmented and non-pigmented rice cultivars [10,11]. Moirangthem et al. [12] reported higher oxidation stability in black rice malt beer than in barley malt beer. In addition, the supplementation of barley malt with pigmented rice influenced the colour and antioxidant activity of the wort via anthocyanin [10,11]. Pigmented rice such as the Riceberry rice (*Oryza sativa*) is a dark purple rice variety that is widely cultivated in Thailand. This rice cultivar is produced via cross-breeding between the Khao Hom Nin rice variety, which contains high antioxidants, and Khao Hom Mali 105, a fragrant rice cultivar. It is cultivated for its nutritional value as a rich source of anthocyanin and antioxidant activity [13].

The attributes associated with rice malt as an adjunct or base material on phenolic acid dynamics, antioxidation, and volatile phenolic aromatic compounds in beer brewing have not been investigated. The composition of malt grist, the brewing process, and the type of yeast for fermentation affect the amount of hydroxycinnamic acid solubilisation and 4-vinylguaiacol produced in wort after mashing and wort fermentation. Hence, this study takes an in-depth look at hydroxycinnamic acid, antioxidation, and 4-vinylguaiacol dynamics in wort made by substituting barley malt with varying Riceberry rice malt fractions (40% (*w*/*w*), 60% (*w*/*w*), 80% (*w*/*w*), and 100% (*w*/*w*)), and in the respective beer produced using *Saccharomyces cerevisiae* and *Saccharomyces cerevisiae* var *diastaticus*.

2. Materials and Methods

2.1. Malts and Yeast Strain

Malted Riceberry rice malt was produced according to the method described by Gonu and Withayagiat [11] using Weyermann pilsner barley malt, German; centennial pellet hop (T90) with alpha acid of 9.5–11.5%, 2020 cropping year, USA; and two top fermenting yeast strains, LalBrew Belle Saison™ (*Saccharomyces cerevisiae* var. *diastaticus*) and LalBrew BRY 97™ (*Saccharomyces cerevisiae*), produced by Lallemand Inc., Montréal, QC, Canada, which were labelled as phenolic off-flavour producers (POF^+) and phenolic off-flavour negative (POF^-), respectively.

2.2. Wort Production and Characterisation

The previous study showed that the phenolic acid profile and antioxidant activity of wort containing 20% (*w*/*w*) of Riceberry rice malt in the composite malt was not significantly different from 100%*w*/*w* barley malt [11]. Thus, in this study, the wort was prepared by substituting barley malt (100% (*w*/*w*)) with Riceberry rice malt at 40% (*w*/*w*), 60% (*w*/*w*), 80% (*w*/*w*), and 100% (*w*/*w*) as shown in Table 1. The hundred percentage of barley malt was designated the control of this study.

Table 1. Physico-chemical properties of composite malt extract [11].

Samples	Composite Malt Ratio		Wort Characteristics	
	Barley Malt (B) (%*w*/*w*)	Riceberry Rice Malt (R) (%*w*/*w*)	Free Amino Nitrogen (mg/L)	pH
B100	100	0	282.00 ± 1.92 f	5.36± 0.01 c
R40B60	60	40	195.90 ± 0.72 d	5.36 ± 0.01 c
R60B40	40	60	171.70 ± 1.32 c	5.28 ± 0.01 b
R80B20	20	80	145.70 ± 0.39 b	5.24 ± 0.00 c
R100	0	100	127.70 ± 1.00 a	5.21 ± 0.01 a

Mean ± standard error. The different alphabet shown in the same column indicates significant differences at $p < 0.05$.

The Riceberry rice malt–barley malt composite wort was produced in a mashing steel bucket (10 L) with an overhead stirrer (250 rpm) using 1 kg malt to 5 L water. Wort was produced using a temperature–time mashing schedule, as follows: an initial mashing-in temperature of 45 °C for 20 min; 62 °C for 30 min; 72 °C for 30 min; and mashing-out at 78 °C for 20 min. The temperature increase was performed at a rate of 1 °C/min [11]. The original gravity of each wort was adjusted to a specific gravity of 1.040, and free amino nitrogen and the pH were analysed according to the EBC method (European Brewery Convention, 2000) [14] reported in previous studies [11].

2.3. Bioreactor Configuration for Fermentation

A complete Winpact one fermenter system for 5 L double jacket vessel (Winpact Scientific; PS-06-220 model, Saratoga, CA, USA) containing 4.2 L of oxygen saturated wort was used for fermentation. The fermentation vessel was monitored via a control unit, which consisted of a dissolved oxygen probe, a pH probe, a foam detection probe, and a temperature probe with a coolant system connected to the heat exchanger unit (Winpact one system FS-06 Series). The impeller was set at 100 rpm for gentle mixing, better consistency of fermentation, and shorter time to attenuation. Dry yeast was activated according to the Lallemand manufacturer guidelines using 1 g/L of wort. Fermentation was carried out for 72 h at 20 °C. After 72 h fermentation, the impeller was stopped for yeast cells to flocculate for 6 days at 4 °C. About 50 mL of the sample was taken every 12 h during fermentation and at the end of yeast rest for sugar and ethanol analysis, phenolic acid, 4-vinyl guaiacol analysis by HPLC, colour of beer by spectrophotometry [14] and antioxidant activity (AOA) analyses.

2.4. Bottle Maturation of Beer

A sterilised swing-top amber bottle was used for beer packaging. Fermented wort obtained after flocculation was conditioned to 20 ± 0.5 °C (temperature for beer maturation) before being filled into the sterilised 0.33 L bottle. Glucose, which was used for carbonation to obtain dissolved CO_2 within 4.7–5 g/L, was calculated using Equation (1) [15]. The filled capped bottles were kept for 4 weeks at 20 °C. Beer samples were analysed weekly for phenolic acids, 4-vinylguaiacol, monomeric anthocyanin, and AOA.

$$\mathrm{Cb} = \mathrm{Cf} + \frac{0.5(0.91\mathrm{D})}{\mathrm{Vb}} \quad (1)$$

where Cf is the level of CO_2 at end of fermentation, approximately 1.5 g/L; D is the amount of glucose (g); Vb is the volume of beer per bottle (L); and C_B is the carbonation level in the finished beer (g/L).

2.5. Analysis of Fermented Wort and Matured Beer

2.5.1. Extraction of Phenolic Acids in Wort and Beer via the Non-Hydrolysed Method

A wort/beer sample of 20 mL was adjusted to a pH of 2.5 with HCl (4 M), and 500 mg of NaCl was added. The mixture was vortexed until NaCl was completely dissolved. Ethyl acetate of 10 mL was added to the mixture to extract phenolic acids. The mixture was vortexed and centrifuged at 10,000 rpm for 5 min to obtain the ethyl acetate fraction. The extraction of phenolic acid was repeated thrice. The ethyl acetate was pooled together and evaporated at 38 °C with a vacuum rotatory evaporator. The dry phenolic extract was solubilised with 5 mL of 50% methanol and was kept at −20 °C prior to sample analysis [2].

2.5.2. Analysis of Phenolic Acids in Composite Malt Wort and Beer

The analyses of ferulic acid, *p*-coumaric acid, sinapic acid, and vanillic acid were analysed according to Gonu et al. [11]. A gradient condition was set with a Phenomenex (USA), Luna® C18 column (250 × 4.6 mm, 5 µm), conditioned at 35 °C. The mobile phase constituted acetic acid (2%) (solvent A) and absolute acetonitrile (solvent B). The mobile phase elution was performed at a flow rate of 0.8 mL/min with an elution gradient as follows: 0–5 min, 4% (*v*/*v*) B; 5–15 min, 5% (*v*/*v*) B; 15–30 min, 10% (*v*/*v*) B; 30–50 min, 15% (*v*/*v*) B; 50–60 min, 18% (*v*/*v*) B; 60–70 min, 20% (*v*/*v*) B; 70–72 min, 25% (*v*/*v*) B; 72–74 min, 55% (*v*/*v*) B; 74–75 min, 70% (*v*/*v*) B; 75–78 min, 70% (*v*/*v*) B; 78–80 min, 4% (*v*/*v*) B; and 80–83 min, 4% (*v*/*v*) B. The peaks of ferulic acid, *p*-coumaric acid, and sinapic acid were detected at 320 nm, and the peak of vanillic acid was detected at 280 nm.

2.5.3. Quantification of 4-Vinylguaiacol in Wort and Beer

The 4-vinylguaiacol was quantified via HPLC analysis on a Luna C18 column (250 mm × 4.6 mm, 5 µm) with a mobile phase of 2% aqueous acetic acid (solvent A) and 100% acetonitrile (solvent B), with flow rates set at 0.35 mL/min and 0.45 mL/min, respectively. Approximately 20 µL of filtered (0.45 µm nylon filter) phenolic extract was used for the HPLC analysis. The 4-vinylguaiacol yield via fermentation was determined as the ratio of 4-vinylguaiacol production (mg/L) to ferulic acid consumption (mg/L), and the rate of 4-vinylguaiacol production (mg/L/h) was the ratio of 4-vinylguaiacol yield (mg/L) to fermentation time (h).

2.5.4. Total Anthocyanin by the pH Differential Method

Wort/beer of 0.6 mL was added to 2.4 mL potassium chloride (KCl) buffer of pH 1, and the same volume of sample was added to 2.4 mL of sodium acetate (CH_3COONa) with pH 4.5. The samples were incubated at room temperature for 15 min, and the spectrophotometer absorbance was set at 510 nm and 700 nm for each sample with water used as a reference [12].

The absorbance (A) of the samples was calculated as

$$A = (A_{\lambda\ vis\ max} - A_{700})_{pH\ 1.0} - (A_{\lambda\ vis\ max} - A_{700})_{pH\ 4.5} \quad (2)$$

The total anthocyanin pigment (mg/L) was calculated as

$$\text{Total anthocyanin pigment} = \frac{A \times MW \times DF \times 1000}{(\varepsilon \times 1)} \quad (3)$$

where MW is the molecular weight of anthocyanin (449.2 g/mol), DF is the dilution factor, and ε is molar absorbance of cyn-3-glu (26,900 L/mol/cm).

2.5.5. Antioxidant Activity via 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and 2-2′-Azino-bis-3-ethylbenzothiazoline-6-sulfonic Acid (ABTS) Assays

The antioxidant activity of wort and beer were assayed by colorimetric methods. The 2,2-di-phenyl-1-picrylhydrazyl (DPPH) is a stable free-radical compound used for a radical scavenging capacity assay of hydrophobic compounds based on electron transfer activity,

whereas ABTS is another form of free-radical compound used for an assay of hydrogen atom transfer by antioxidant compounds soluble in an aqueous and organic solvent.

A DPPH solution of 80 µM was prepared in absolute methanol. A DPPH solution of 2.9 mL was mixed with a 100 µL sample (wort/fermented wort and beer), deionised water (blank), or standard Trolox. The mixture was then vortexed and incubated at room temperature for 60 min and kept in a dark cabinet. The absorbance values of the sample, a blank, and the standard reaction were taken at a wavelength of 517 nm using a Shimadzu UV–1800 spectrophotometer. A standard calibration curve of Trolox (0.02–0.08 mM) was plotted. The antioxidant activity of each sample was expressed as micromole of Trolox equivalent (TE) per litre (µmoL TE L^{-1}) [16].

The ABTS assay was conducted according to Suriano et al. [16] and Gonu et al. [11]. About 10 mL ABTS solution (7 mM) was prepared containing 2.45 mM potassium persulphate. The mixture was kept at room temperature for 16 h in a dark cabinet prior to use. The 10 mL stock ABTS solution was mixed with reverse osmosis (RO) water to have a final absorbance of 0.700 ± 0.02 at 734 nm using RO water as a reference. Antioxidant activity was determined using a similar DPPH reaction volume with an incubation time of 20 min. A Trolox (0.01–0.08 mM) calibration curve was constructed

2.5.6. Statistical Analysis

Each experimental treatment was carried out in triplicate. The data gathered were subjected to a two-way analysis of variance (ANOVA) using GenStat Edition 12 (VSN International, Hemel Hempstead, UK).

3. Results and Discussion

3.1. Glucose and Maltose Dynamics during Fermentation and Beer Maturation

Glucose, maltose, and ethanol in fermented wort were analysed by HPLC. Glucose content was significantly different ($p < 0.05$) in the composite wort prior to fermentation (Figure 1A,B). In the worts, glucose content was the highest in R100 (18.3 mg/L) and the least in B100 (10.21 mg/L). Substituting barley with 40% (*w*/*w*), 60% (*w*/*w*), and 80% (*w*/*w*) Riceberry rice malt was directly proportional to the glucose content in the worts. This is because rice has more starch content, with amylopectin being the major component (65–85%) compared to the starch amylopectin of barley (42–71.2%) [17]. The amylopectin is hydrolysed during rice malting by the enzymes limiting dextrinase and alpha glucosidase, which are in abundance in malted rice [18]. POF^- and POF^+ utilised glucose rapidly in the worts of R80B20 and R100 during 36 h fermentation (Figure 1A,B), but it was faster in POF^+. Yeasts of *diastaticus* variants are characterised as rapid fermenters and have optimum activity at higher temperatures of 20–35 °C [19]. Maltose content in composite worts reduced with increasing percentages of Riceberry rice malt between 60% (*w*/*w*) and 100% (*w*/*w*). The maltose content varied significantly ($p < 0.05$) among the composite worts produced. The wort B100 had the maximum maltose content (Figure 1C). The high maltose content in B100 after mashing was because barley malt possesses a higher amount of beta amylase, which cleaves the non-reducing ends of the polysaccharide chain to produce maltose [20]. This enzyme activity is limited in rice malt, resulting in low maltose production during mashing [8]. Hence, barley malt additions complemented maltose content in the composite wort. The dynamics of maltose during the fermentation of each composite wort by POF^- and POF^+ is presented in Figure 1C,D. Both POF^- and POF^+ were able to utilise maltose sufficiently within 72 h, and the peak of the maltose metabolism was within 60–72 h of fermentation. Comparing POF^- and POF^+, the latter had a stronger affinity to metabolise maltose during active fermentation (72 h) in each treatment (Figure 1C,D) and hence is referred to as a super-attenuating yeast [19].

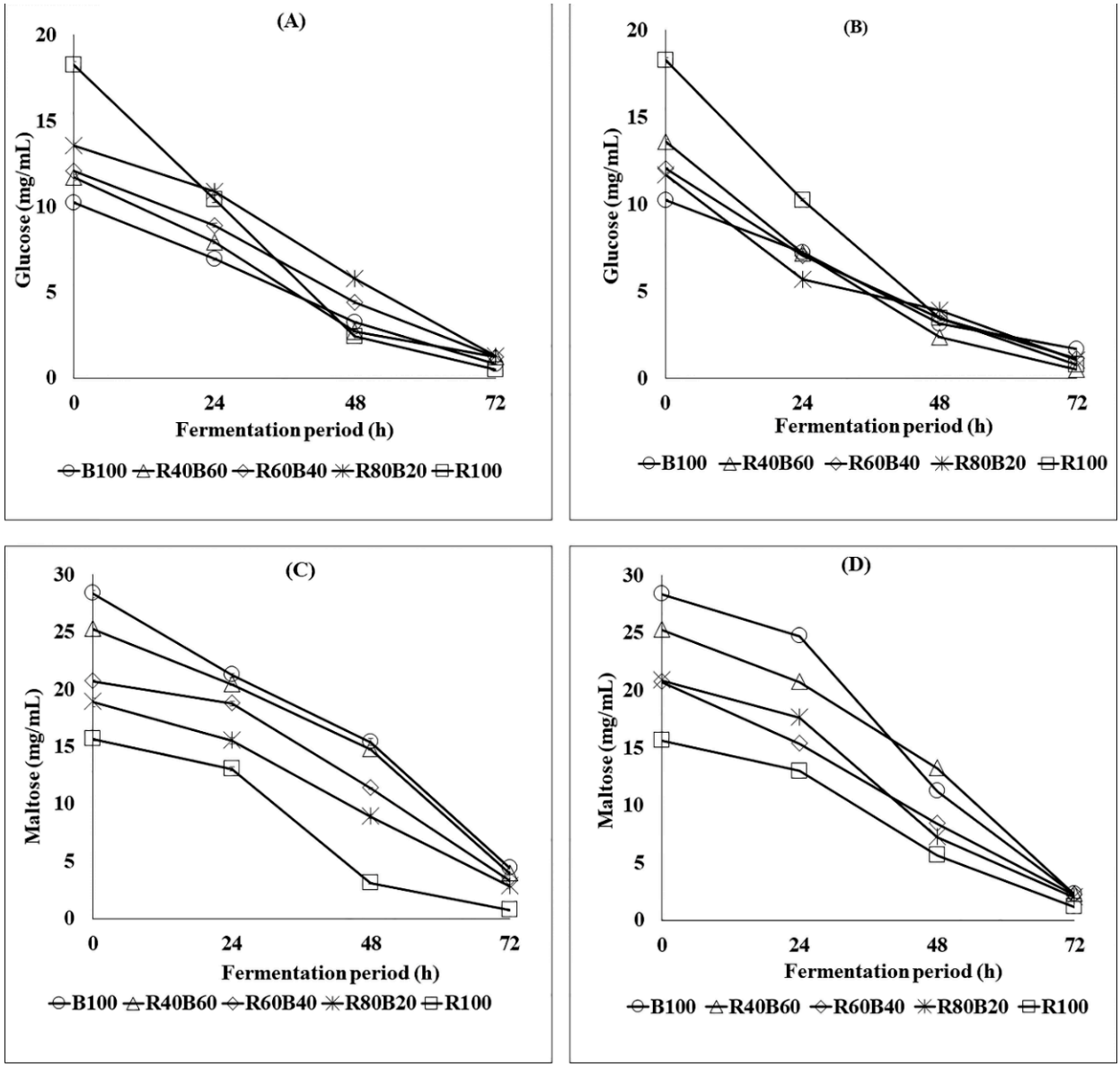

Figure 1. Dynamic of glucose and maltose during 72 h fermentation of composite wort: glucose metabolism by POF^- (**A**) and POF^+ (**B**); maltose metabolism by POF^- (**C**) and POF^+ (**D**). BRY 97 (*S. cerevisiae*), POF^-; and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+.

3.2. Ethanol Production

The ethanol production by POF^- and POF^+ was relatively similar but pronounced in POF^+ within 24 h. After 72 h of active fermentation with POF^-, the ethanol yield was 4.0% (*v*/*v*), 3.9% (*v*/*v*), 3.8% (*v*/*v*), 3.6% (*v*/*v*), and 3.5% (*v*/*v*) in B100, R40B80, R60B40, R80B20, and R100, respectively. For POF^+, values of 4.1% (*v*/*v*), 3.7% (*v*/*v*), 3.7% (*v*/*v*), 3.6% (*v*/*v*), and 3.5% (*v*/*v*) were recorded in B100, R40B80, R60B40, R80B20, and R100, respectively. Substituting barley malt with Riceberry rice malt resulted in a low ethanol yield, unlike the use of unmalted rice as an adjunct, which increases ethanol yield. After 4 weeks of beer maturation, the alcohol content in B100, R40B60, and R60B40 was within 4.0–4.2% (*v*/*v*), and that in R80B20 and R100 was 3.6–3.8% (*v*/*v*), respectively, for both POF^- and POF^+. In this study, beer of R100 had an ethanol content above 3.19–3.28% (*v*/*v*), which was reported in black rice beer by Moirangthem et al. [12] but was below the values (4.59–5.12% (*v*/*v*)) reported by Mayer et al. [9] for malted rice beer.

3.3. Colour Unit of Cast Wort and Matured Beer

The EBC colour unit was anaylsed [14] and varied among each treatment cast wort, as shown in Table 2. This was observed in beer after 4 weeks of maturation. Increasing the percentage of Riceberry rice malt in malt compositions resulted in an increasing colour unit. This was because Riceberry rice malt contains anthocyanin. It was observed that beer produced with POF^- had a higher EBC colour unit than beer produced with POF^+.

Table 2. EBC colour value of composite wort and beer after 4 weeks of maturation produced with BRY 97 (*S. cerevisiae*), POF^-; and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+.

	B100	**R40B60**	**R60B40**	**R80B20**	**R100**
Cast wort	10.36 ± 0.11	11.89± 0.29	16.43 ± 0.24	26.27 ± 0.18	29.49 ± 0.15
POF^-	8.36 ± 0.12	7.36 ± 0.23	13.28 ± 0.11	22.02 ± 0.16	24.46 ± 0.20
POF^+	7.36 ± 0.10	6.97 ± 0.18	12.36 ± 0.05	20.36 ± 0.09	22.15 ± 0.1

3.4. Ferulic Acid Changes and 4-Vinylguaiacol Production during the Brewing Process as an Impact of Composite Riceberry Rice Malt–Barley Malt and Yeast Type

Wort produced with Riceberry rice malt as a base raw material and as an adjunct was characterised prior to fermentation. The Riceberry rice malt wort had ferulic acid (3.8 mg/L) lower than that found in barley malt wort (4.5 mg/L) (Figure 2). The ferulic acid in each treatment was within a range of 1.5–6.6 mg/L, as reported by McMurrough et al. [21] and Coghe et al. [3] in wort. In this research, the dynamics of ferulic acid during the fermentation process by POF^- and POF^+ yeast is illustrated in Figure 2. Wort fermented by POF^+ yeast had its ferulic acid concentration increase within 36 h (Figure 2). This was observed in wort produced with 40% (*w*/*w*), 60% (*w*/*w*), and 100% (*w*/*w*) barley malt but was absent in wort produced with 80% (*w*/*w*) and 100% (*w*/*w*) Riceberry rice malt. The increase in ferulic acid during fermentation may be due to the presence of a ferulic acid complex, such as ferulic-arabinoxylan in barley malt wort [22], which can be liberated by the POF^+ yeast. This group of yeast possesses the phenylacrylic acid decarboxylase (PAD1) and ferulic acids decarboxylase (FDC1) gene [2] to express feruloyl esterase to liberate ferulic acid bonded to a solubilised arabinoxylan structure [23]. Beyond 36 h of active fermentation with POF^+, ferulic acid began to decrease. This phenomenon was not observed in wort fermented with POF^- yeast (Figure 2). There was an indication that POF^- yeast lacked the expression of the FDC1 gene during fermentation, but ferulic acid declined in each wort fermented by POF^- (Figure 2), which was associated with phenolic acid absorption, a phenomenon associated with brewing yeast [24]. At yeast rest (flocculation), the ferulic acid content decreased in each treatment (Figure 2) and proceeded to be relatively stable in beer produced with POF^- during the 4 weeks of maturation, and ferulic acid declined in each beer produced with POF^+ within 2 weeks of maturation and maintained stability until the fourth week (Figure 2). The stability of ferulic acid was associated with two factors, including an increase in carbon dioxide production in the bottled beer because of the priming sugar, hence creating an anaerobic condition which limited yeast metabolism [15]. The second factor was the decline in yeast nutrients during beer maturation. The POF^- produced beer after 4 weeks of maturation, and B100, R40B60, R60B40, R80B20, and R100 recorded ferulic acid of 4.00 mg/L, 3.59 mg/L, 3.54 mg/L, 3.55 mg/L, and 3.34 mg/L, respectively. In beer produced with POF^+, the ferulic acid was 3.66 mg/L, 3.48 mg/L, 3.33 mg/L, 2.95 mg/L, and 3.32 mg/L in B100, R40B60, R60B40, R80B20, and R100 beer, respectively. These concentrations were within the ferulic acid content (2.8–6.6 mg/L) reported by McMurrough et al. [21] in ale commercial beers.

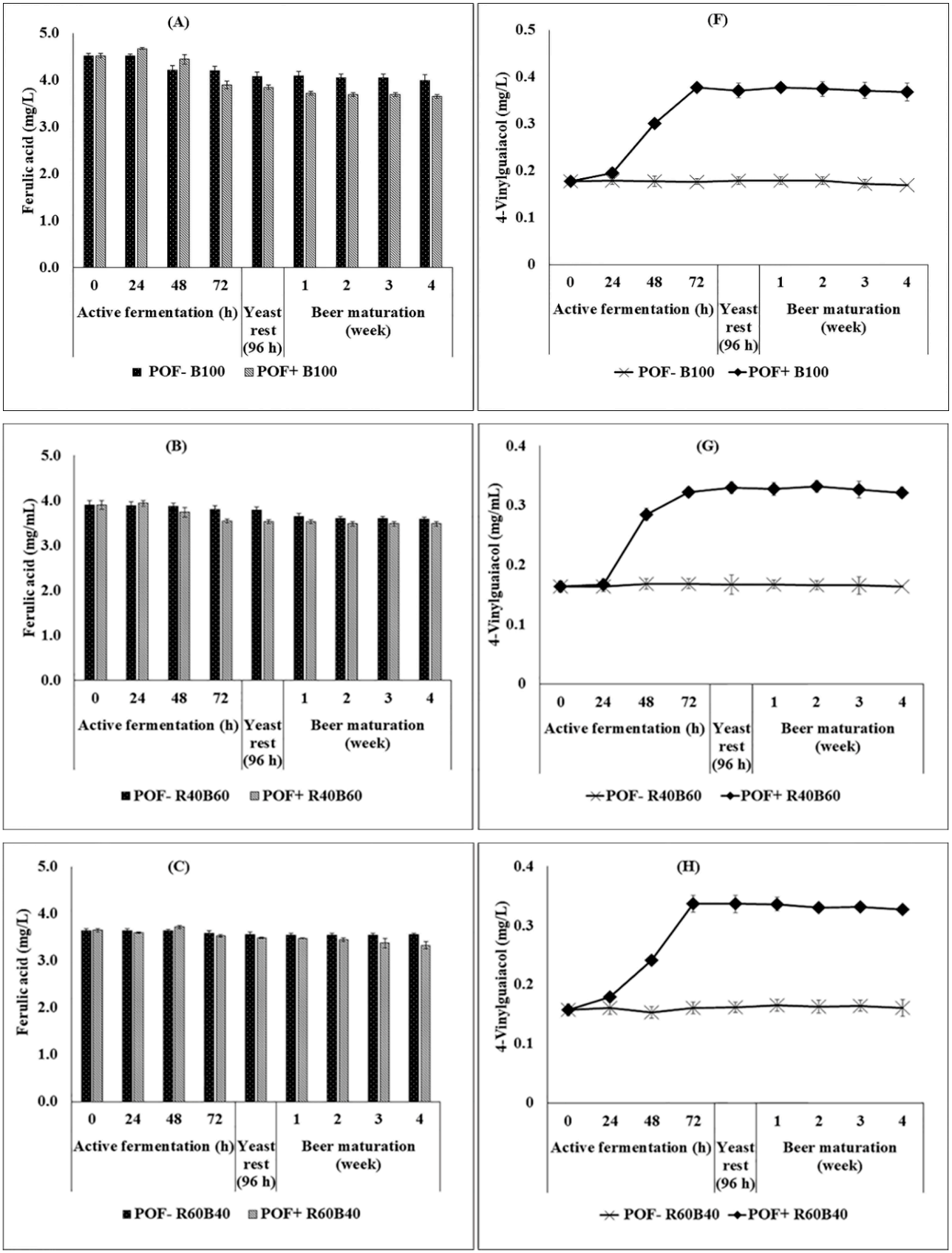

Figure 2. *Cont.*

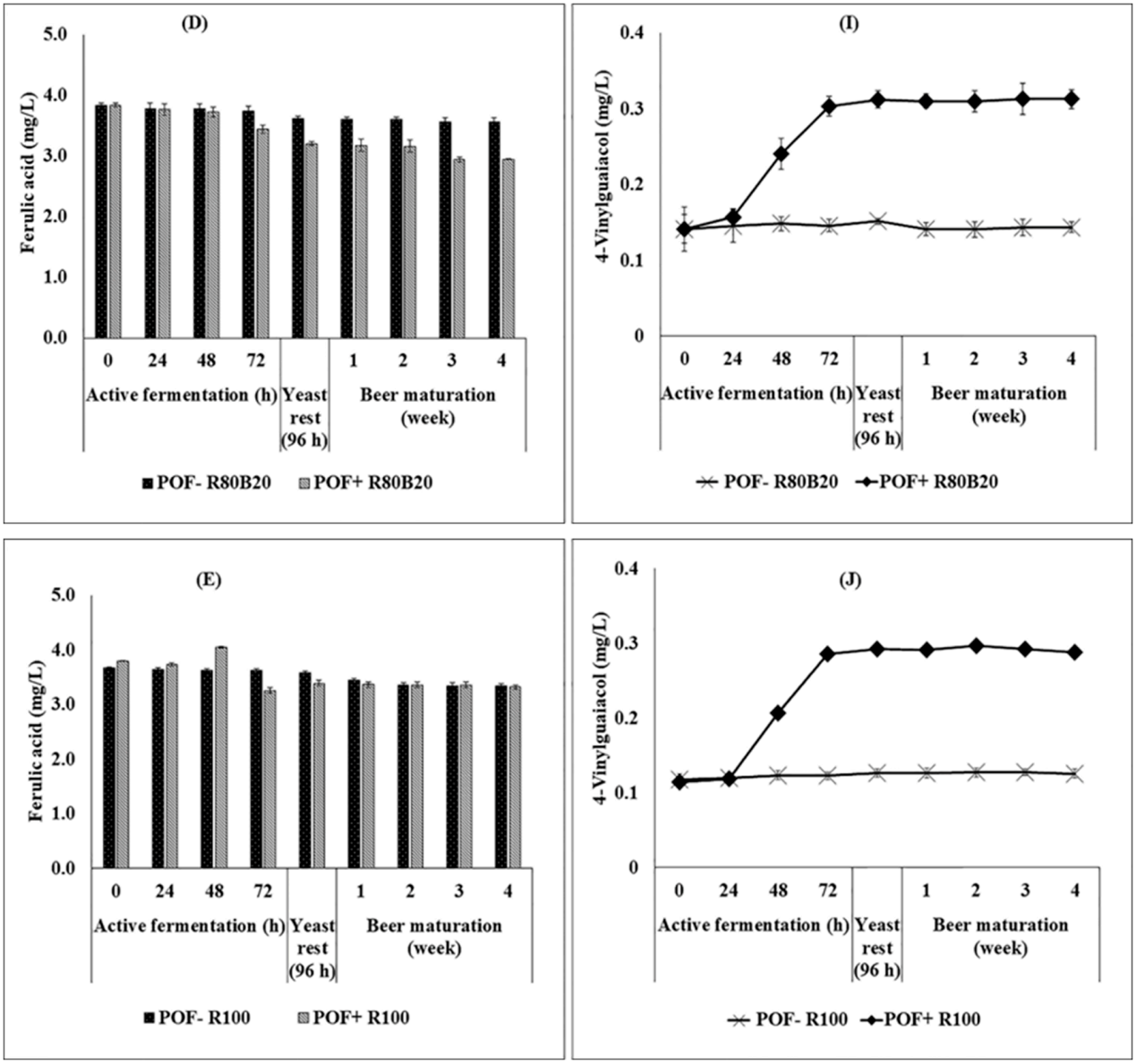

Figure 2. Ferulic acid and 4-vinylguaiacol concentration in the fermented composite wort (B100, R40B60, R60B40, R80B20, and R100) by two yeast strains BRY 97 (*S. cerevisiae*), POF^{-} (**A**–**E**); and Belle Saison (*S. cerevisiae var. diastaticus*), POF^{+} (**F**–**J**).

The 4-vinglyguaiacol is responsible for the spice/clove characteristic in beverages and some types of beer. Prior to fermentation, each composite cast wort contained 4-vinylguaiacol, as shown in Figure 2. Its level in boiled wort and cast wort ranged between 0.15–0.20 mg/L; 0.13–0.17 mg/L; 0.15–0.17 mg/L; 0.14–0.15 mg/L; and 0.11 mg/L in B100, R40B60, R60B40, R80B20, and R100, respectively. These were within the range of 0.05–0.28 mg/L reported in wort after mashing and boiling by Coghe et al. [3] and McMurrough et al. [21]. According to Figure 2, adding Riceberry rice malt to produce wort led to low 4-vinylguaiacol production via boiling. In addition, that barley malt was the sole contributor of ferulic acid for 4-vinylguaiacol production. The amount of 4-vinylguaiacol produced by yeast activity was significantly influenced by the composite wort. Figure 2A–C show 4-vinylguaiacol production by *S. cerevisiae var. diastaticus* (POF^{+}), which occurred simultaneously with ferulic acid release within 36 h. This led to an average 4-vinylguaiacol content of 0.40 mg/L, 0.32 mg/L, 0.34 mg/L, 0.31 mg/L, and 0.29 mg/L after 72 h of fermentation in B100, R40B80, R60B40, R80B20, and R100, respectively. Hence, POF^{+} yeast

was neither influenced by wort composition nor the amount of ferulic acid present in wort to produce 4-vinylguaiacol, and the presence of 4-vinylguaiacol before fermentation did not inhibit its synthesis by POF^+ yeast. The 4-vinylguaiacol yield by POF^+ yeast was 0.05, 0.04, 0.05, 0.05, and 0.05 in B100, R40B80, R60B40, R80B20, and R100, respectively, with production rates between 0.002 and 0.003 mg/L/h after 72 h of fermentation. The amount of 4-vinylguaiacol produced in each fermented wort was within the threshold of 4-vinylguaiacol in 18 types of beer (0.01–0.57 mg/L) reported by McMurrrough et al. [21]. Unlike POF^+, the POF^- yeast did not possess the ability to decarboxylate ferulic acids to 4-vinylguaiacol in each composite wort (Figure 2) because it lacked the expression of the PAD1 and FDC1 gene. However, 4-vinylguaiacol produced via boiling prior to fermentation persisted during fermentation and declined gradually. Bottled beers produced with POF^+ after 4 weeks of maturation had 4-vinylguaiacol content between 0.29–0.37 mg/L, and that of matured beer produced with POF^- ranged between 0.13 mg/L and 0.17 mg/L.

3.5. p-Coumaric Acid Kinetic in Fermented Wort and Beer

The *p*-coumaric acid in each composite wort prior to fermentation was 2.26 mg/L, 2.65 mg/L, 3.18 mg/L, 3.72 mg/L, and 4.20 mg/L in B100, R40B60, R60B40, R80B20, and R100, respectively (Figure 3). The difference in malt composition influenced *p*-coumaric acid in worts significantly ($p < 0.05$). The concentration of *p*-coumaric acid was dependent on the malted rice fraction. Gonu et al. [13] reported that malted paddy rice contained high *p*-coumaric acid because of the rice husk containing more *p*-coumaric acid [1]. The *p*-coumaric acid varied significantly ($p < 0.05$) with fermentation period and among the yeast strains. In POF^+ fermented wort, *p*-coumaric acid declined with fermentation period (Figure 3A) because of the decarboxylation of *p*-coumaric acid to 4-vinylphenol by POF^+ metabolism [5]. This resulted in a reduction in *p*-coumaric acid by 1.9%, 19.2%, 9.9%, 13.4%, and 8.7% in B100, R40B60, R60B40, R80B20, and R100, respectively, after 72 h of fermentation. The *p*-coumaric acid content was stable in POF^- fermented wort (Figure 3A). For the bottled maturation of beer produced with POF^+, *p*-coumaric acid declined after a week of maturation, and it maintained a relatively stable concentration into week 4 of beer maturation (Figure 3A). POF^+ beer recorded a final *p*-coumaric acid concentration of 2.10 mg/L in B100, R40B60 (1.98 mg/L), R60B40 (2.74 mg/L), R80B20 (3.07 mg/L), and R100 (3.66 mg/L). The beers produced with POF^- had *p*-coumaric acid of 2.08 mg/L in B100, R40B60 (2.42 mg/L), R60B40 (2.96 mg/L), R80B20 (3.50 mg/L), and R100 (3.96 mg/L). The observed effect of brewing with R100 on the hydroxycinnamic acid profile contradicts with that of *p*-coumaric acid reported by Moirangthem et al. [12] in rice beer, which was below the ferulic acid concentration. The variation in results reported can be associated with the mashing schedule, boiling period, the type of rice cultivar, and the phenolic acid extraction process. In this study, prior to phenolic extraction, the acidification was carried out, which was absent in the work by Moirangthem et al. [12]. Nevertheless, worts produced with 50% of rice and 50% barley malt contained higher *p*-coumaric acid than ferulic acid [2], which corroborates with this study. However, inadequate data exist on rice malt as an adjunct on hydroxycinnamic acid in beer. This study suggests that paddy rice malt additions changed the dynamics of hydroxycinnamic acid and the potential of more decarboxylated *p*-coumaric acid during boiling and fermentation with POF^+, unlike conventional beer brewed with barley malt [2] or wheat malt [3], which have higher concentrations of ferulic acid and 4-vinylguaiacol.

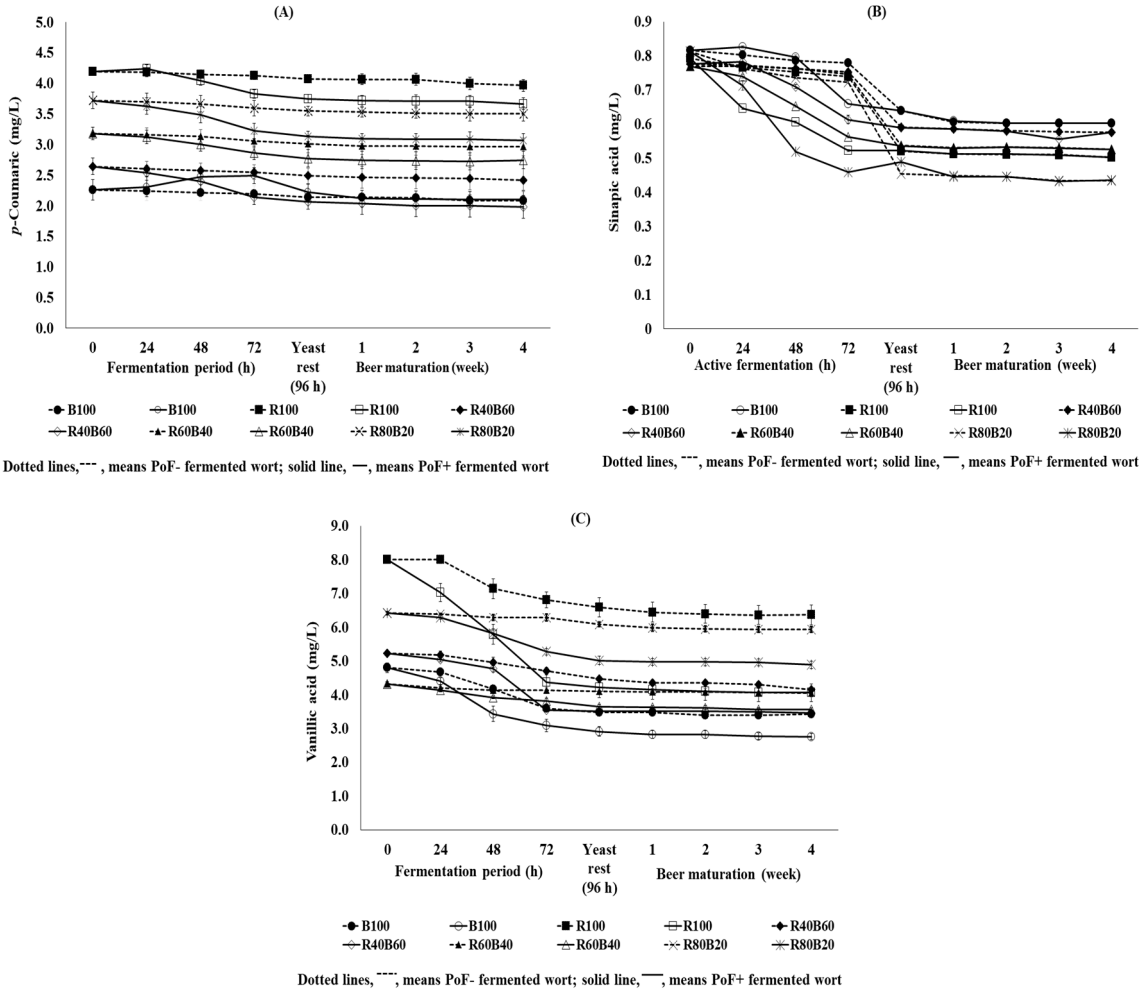

Figure 3. Changes in *p*-coumaric acid (**A**), sinapic acid (**B**), and vanillic acid (**C**) in fermented composite wort (B100, R40B60, R60B40, R80B20, and R100) by the two yeast strains BRY 97 (*S. cerevisiae*), POF^-; and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+.

3.6. Sinapic Acid (SA) Kinetics in Fermented Wort and Beer

The sinapic acid kinetics in composite worts during 72 h of fermentation by POF^- and POF^+ yeast is illustrated in Figure 3B. In cast worts of B100, R40B60, R60B40, R80B20, and R100, the SA content was 0.82 mg/L, 0.78 mg/L, 0.77 mg/L, 0.81 mg/L, and 0.79 mg/L, respectively, and was present in each fermented wort and bottled beer (Figure 3B). The amount of SA in worts before fermentation and during fermentation was the least among the phenolic acids analysed. The fermented wort after 72 h by POF^- recorded a decline in sinapic acid by 4.89%, 3.8%, 2.6%, 10.0%, and 6.3% in B100, R40B60, R60B40, R80B20, and R100, respectively. Sinapic acid reduction is linked to two activities: the adsorption of sinapic acid by the yeast cell wall, and sinapic acid polyphenol complex formation, which precipitates during fermentation and yeast rest [25]. In worts fermented with POF^+, sinapic acid is significantly reduced in B100 (19.5%), R40B60 (21.8%), R60B40 (27.3%) R80B20 42.5%), and R100 (34.2%).

3.7. Vanillic Acid Dynamics in Fermented Wort and Matured Beer

Vanillic acid is classified among hydroxybenzoic acids. The concentration of vanillic acid varied significantly in each sample (Figure 3C). Riceberry rice malt as an adjunct elevated vanillic acid content in worts produced because Riceberry rice malt contains high vanillic acid [13]. The dynamics of vanillic acid after 72 h of fermentation (Figure 3C) among the two yeast strains were similar to those of sinapic acid. For the reduction in

vanillic acid from worts to beer, there was a similar pattern observed in all black rice malt beer (a decline from 3.37 mg/L in wort to 3.06 mg/L in beer), but in white kernel rice malt beer, vanillic acid increased from 0.29 mg/L to 0.52 mg/L despite using the same yeast strain Nottingham ale dried yeast [12]. These comparisons suggest that vanillic acid can be metabolised by the brewing yeasts or reduce via precipitation in the pigmented fermented wort, as observed in this study. Vanillic acid concentrations after 4 weeks in beer were 3.43 mg/L, 4.05 mg/L, 4.15 mg/L, 5.94 mg/L, and 6.37 mg/L in B100, R40B60, R60B40, R80B20, and R100, respectively, with POF^-. In addition, in POF^+ matured beer, vanillic acid content was 2.76 mg/L, 3.47 mg/L, 3.56 mg/L, 4.89 mg/L, and 5.06 mg/L in B100, R40B60, R60B40, R80B20, and R100, respectively, which were above the vanillic acid concentratiosn reported by Moirangthem et al. [12] in rice beer produced from pigmented rice (3.06 mg/L) and non-pigmented (0.52 mg/L) rice. However, vanillic acid concentrations in this study and those reported by Moirangthem et al. [12] were above the ferulic, *p*-coumaric, and sinapic acid reported in beer samples; hence, the use of pigmented rice malt as an adjunct in the brewing process improves vanillic acid concentrations in beers.

3.8. Dynamics of Monomeric Anthocyanin in Fermented Wort and Beer

Figure 4 illustrates monomeric anthocyanin kinetics during fermentation for each treatment. Before fermentation, monomeric anthocyanin was present in worts containing Riceberry rice malt, except for B100. The monomeric anthocyanin declined as fermentation progressed and experienced a further reduction during the yeast flocculation period in both POF^- and POF^+ fermented wort (Figure 4) samples. The reduction in monomeric anthocyanin was because of the precipitation of anthocyanin due to the decline in pH and the yeast cell membrane adsorption of anthocyanin during fermentation [26]. Furthermore, anthocyanin can precipitate via anthocyanin and hydroxycinnamic acid interactions, but this requires a prolonged period [27], such as during beer maturation. According to de Freitas et al. [27], declines in anthocyanin during fermentation are also contributed to by yeast metabolites such as acetaldehyde, which reacts with anthocyanin to form pyranoanthocyanins or polymeric anthocyanin. The transformed compound reacts with proteins and precipitates at low temperatures during yeast flocculation [28]. Hence, there is a low anthocyanin content in composite beer containing high barley malt proportions (60–100% (*w*/*w*)) because barley is a rich source of protein. These factors, in addition to the instability of monomeric anthocyanin, collectively led to the decline of the EBC colour value of beer after 4 weeks of maturation in R80B20 and R100 significantly (Table 1).

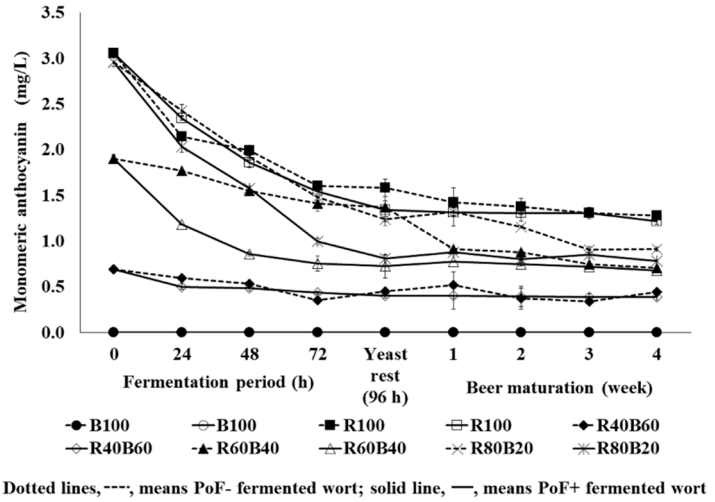

Figure 4. Monomeric anthocyanin kinetics in fermented composite wort (B100, R40B60, R60B40, R80B20, and R100) by the two yeast strains BRY 97 (*S. cerevisiae*), POF^-; and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+.

3.9. Antioxidant Activity (AOA) during Wort Fermentation and in Beer via DPPH and ABTS Assay

The antioxidant activity via DPPH assay during fermentation among the five composite worts by each yeast strain are shown in Figure 5A,B. The AOA of the fermentation in each composite wort was significantly different ($p < 0.05$) and was more noticeable in the R80B20 and R100 treatments. The antioxidant activity of R80B20 and R100 in each fermentation treatment was similar (Figure 5). Likewise, the fermented worts of B100, R40B60, and R60B80 shared similarity of antioxidant activity. The fermentation of worts by POF^+ was associated with unstable antioxidant activity, as shown in Figure 5B, because of the POF^+ ability to metabolise phenolic acids, including ferulic acid to 4-vinylguaiacol and *p*-coumaric and sinapic acid to 4-vinylphenol and 4-vinylsyringol, respectively [5]. These phenolic derivatives possess less antioxidation. Each fermented wort containing Riceberry rice malt between 40% (*w*/*w*) and 100% (*w*/*w*) had higher or equal antioxidant activity relative to B100. Although some antioxidants are associated with oxidation stability, others can serve as being pro-oxidant, thus influencing the total antioxidation stability of the fermented wort or the end product of beer. Barley malt beer has been reported to have less oxidation stability compared to black rice malt beer [12]. The high antioxidant activity in R40B60, R60B40, R80B20, and R100 was attributed to the trace amount of monomeric anthocyanin present in the beer, the high vanillic acid content, and the potential of low pro-oxidant species possessed in rice malt relative to barley malt beer, as proposed by Moirangthem et al. [12]. In the course of bottled beer maturation with POF^- for 4 weeks at 20 °C, the DPPH assay revealed a steady decline in antioxidant activity between 1–2 weeks, and it maintained stable antioxidant activity between 3 and 4 weeks. Hence, antioxidant activity via DPPH assay had ranges of 748.8–782.6 µmoL TE/L, 745.6–756.1 µmoL TE/L, 750.1–761.7 µmoL TE/L, 901.2–924.9 µmoL TE/L, and 961.0–970.8 µmoL TE/L in B100, R40B80. R60B40, R80B20, and R100, respectively, in the beer produced with POF^-. In POF^+ beer, AOA was 756.8–785.7 µmoL TE/L, 778.3–795.2 µmoL TE/L, 820.6–835.0 µmoL TE/L, 900.9–918.8 µmoL TE/L, and 878.5–895.1 µmoL TE/L in B100, R40B80, R60B40, R80B20, and R100, respectively. Few data are available on the AOA of rice beer and composite beer with rice. However, Ceccaroni et al. [11] reported AOA in rice beer via a DPPH assay with a range of 437–587 µmoL TE/L, which is below the values recorded in this study because of rice cultivar variation. Nevertheless, the range of AOA via DPPH was within the range of values recorded for some commercial Polish beer, such as wheat beer (>500 µmoL TE/L), lager (>1200 µmoL TE/L), bock beer (>2000 µmoL TE/L), and porter beer (>2200 µmoL TE/L) [29].

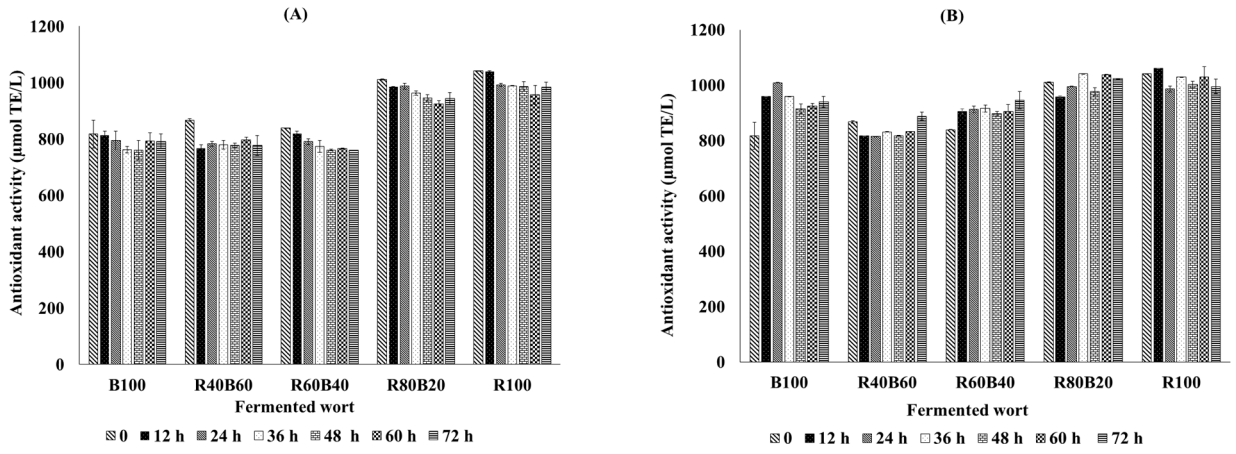

Figure 5. Changes in antioxidant activity via DPPH in fermented wort (B100, R40B60, R60B40, R80B20, and R100) by the two yeast strains BRY 97 (*S. cerevisiae*), POF^- (**A**); and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+ (**B**).

The AOA of fermenting composite worts via ABTS assay is illustrated in Figure 6. The mean AOA via ABTS assay was significantly different among the treatment carried out for each yeast strain. The ABTS assay maintained a similar pattern, as reported for AOA via DPPH. POF^- beer had AOA of 1615.6–1625.3 µmoL TE/L, 1461.4–1491.4 µmoL TE/L, 1470.2–1492.9 µmoL TE/L, 1706.3–1745.4 µmoL TE/L, and 1702.8–1723.6 µmoL TE/L in B100, R40B80. R60B40, R80B20, and R100, respectively. However, POF^+ beer had AOA in B100, R40B80. R60B40, R80B20, and R100 as 1589.3–1650 µmoL TE/L, 1525.5–1558.6 µmoL TE/L, 1608–1636.6 µmoL TE/L, 1765.8– 800.8 µmoL TE/L, and 1721.9–1754.4 µmoL TE/L, respectively. The AOA values were above the antioxidant activity via ABTS recorded for 12 non-hydrolysed dark beer samples, which ranged between 390 and 720 µmoL TE/L [30] but were below the AOA in beer produced with specialty rice malt with 2860 ± 440 µmoL TE/L [31]. This was because the AOA was assayed using the native beer, hence the potential interference of other macromolecules, such as proteins.

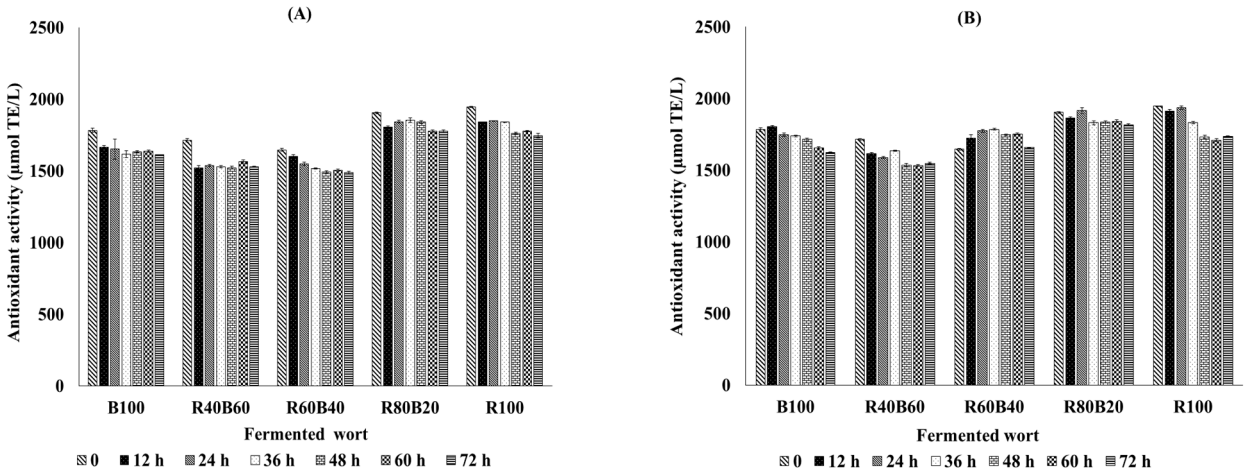

Figure 6. Changes in antioxidant activity via ABTS in fermented wort (B100, R40B60, R60B40, R80B20, and R100) by the two yeast strains BRY 97 (*S. cerevisiae*), POF^- (**A**); and Belle Saison (*S. cerevisiae var. diastaticus*), POF^+ (**B**).

4. Conclusions

Using Riceberry rice malt as a raw material for brewing purposes is feasible. Using Riceberry rice malt as an adjunct for beer production influenced phenolic acid by reducing ferulic acid and elevating vanillic acid in wort at 40% (*w*/*w*), 60% (*w*/*w*), 80% (*w*/*w*), and 100% (*w*/*w*). The reduced ferulic acid led to low production of 4-vinylguaiacol in wort and in beer by POF^- and POF^+ yeast strains. Substituting barley malt with Riceberry rice malt increased monomeric anthocyanin content in wort produced, and it was present in the final beer. Hence, there was higher antioxidant activity in cast wort and beer than all barley wort and beer produced. The study revealed that the amount of phenolic acids and monomeric anthocyanin in wort prior to fermentation are a determinant of the antioxidant activity in beer. However, phenolic acid metabolising yeast such as *S. cerevisiae* var *diastaticus* (POF^+), which can convert hydroxycinnamic acids to volatile phenolic acids (4-vinylguaiacol), can influence the final antioxidant activity in beer relative to beer produced with *S. cerevisiae* (POF^-), which maintains a relatively stable antioxidant activity. This study suggests that using pigmented rice such as Riceberry rice malt as an adjunct can increase vanillic acid and monomeric anthocyanin content in worts and can improve the antioxidation activity of beer.

Author Contributions: H.G.: Formal analysis (lead); Investigation (lead); Methodology (equal); Writing–original draft preparation (lead). U.W.: Conceptualisation (lead); Formal analysis (supporting); Funding acquisition (lead); Investigation (supporting); Methodology (supporting); Project administration (supporting); Supervision (lead); Writing—review and editing (equal). All authors have read and agreed to the published version of the manuscript.

Funding: The National Research Council of Thailand, project number RUN 2561-10.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

Acknowledgments: The authors are grateful to the National Research Council of Thailand, project number RUN 2561-10, for the financial support of this research.

Conflicts of Interest: The authors declare no financial or personal conflict of interest.

References

1. Butsat, S.; Weerapreeyakul, N.; Siriamornpun, S. Changes in phenolic acids and antioxidant activity in thai rice husk at five growth stages during grain development. *J. Agric. Food Chem.* **2009**, *57*, 4566–4571. [CrossRef] [PubMed]
2. Vanbeneden, N.; Van Roey, T.; Willems, F.; Delvaux, F.; Delvaux, F.R. Release of phenolic flavour precursors during wort production: Influence of process parameters and grist composition on ferulic acid release during brewing. *Food Chem.* **2008**, *111*, 83–91. [CrossRef]
3. Coghe, S.; Benoot, K.; Delvaux, F.; Vanderhaegen, A.B.; Delvaux, F.R. Ferulic acid release and 4-vinylguaiacol formation during brewing and fermentation: Indications for feruloyl esterase activity in *Saccharomyces cerevisiae*. *J. Agric. Food Chem.* **2004**, *52*, 602–608. [CrossRef]
4. Mukai, N.; Masaki, K.; Fujii, T.; Kawamukai, M.; Iefuji, H. PAD1 and FDC1 are essential for the decarboxylation of phenylacrylic acids in Saccharomyces cerevisiae. *J. Biosci. Bioeng.* **2010**, *109*, 564–569. [CrossRef]
5. Vanbeneden, N.; Gils, F.; Delvaux, F.; Delvaux, F.R. Formation of 4-vinyl and 4-ethyl derivatives from hydroxycinnamic acids: Occurrence of volatile phenolic flavour compounds in beer and distribution of Pad1-activity among brewing yeasts. *Food Chem.* **2008**, *107*, 221–230. [CrossRef]
6. Maeda, M.; Motosoko, M.; Tokashiki, T.; Tokashiki, J.; Mizutani, O.; Uechi, K.; Goto, M.; Taira, T. Phenolic acid decarboxylase of *Aspergillus luchuensis* plays a crucial role in 4-vinylguaiacol production during awamori brewing. *J. Biosci. Bioeng.* **2020**, *130*, 352–359. [CrossRef] [PubMed]
7. Ogata, T.; Yamada, R.; Ayuzawa, R.; Nakamura, K. Mutation and deletion of pad1 and/or fdc1 and absence of phenolic off-flavor production in top and bottom-fermenting yeasts. *J. Am. Soc. Brew. Chem.* **2020**, *78*, 74–79. [CrossRef]
8. Usansa, U.; Sompong, N.; Wanapu, C.; Boonkerd, N.; Teaumroong, N. The influences of steeping duration and temperature on the α- and β-amylase activities of six thai rice malt cultivars (*Oryza sativa* L. Indica). *J. Inst. Brew.* **2009**, *115*, 140–147. [CrossRef]
9. Mayer, H.; Ceccaroni, D.; Marconi, O.; Sileoni, V.; Perretti, G.; Fantozzi, P. Development of an all rice malt beer: A gluten free alternative. *LWT Food Sci. Technol.* **2016**, *67*, 67–73. [CrossRef]
10. Dos Santos, J.P.; Acunha, T.d.S.; Prestes, D.N.; Rombaldi, C.V.; El Halal, S.L.M.; Vanier, N.L. From brown, red, and black rice to beer: Changes in phenolics, γ-aminobutyric acid, and physicochemical attributes. *Cereal Chem.* **2020**, *97*, 1148–1157. [CrossRef]
11. Gonu, H.; Withayagiat, U. Composite malt with dark-purple rice malt improves the phenolic profile and antioxidant activity of malt extract. *Int. J. Food Sci. Technol.* **2021**, *56*, 5833–5842. [CrossRef]
12. Moirangthem, K.; Jenkins, D.; Ramakrishna, P.; Rajkumari, R.; Cook, D. Indian black rice: A brewing raw material with novel functionality. *J. Inst. Brew.* **2020**, *126*, 35–45. [CrossRef]
13. Gonu, H.; Wanapu, C.; Withayagiat, U. The Correlation of Free and Bound Phenolic Acid with Antioxidant Activity Accelerated by Germination Period and Temperature. *J. Am. Soc. Brew. Chem.* **2022**, *80*, 305–315. [CrossRef]
14. European Brewery Convention. *Analytica-EBC*; Verlag Hans Carl: Nürnberg, Germany, 2000.
15. Štulíková, K.; Novák, J.; Vlček, J.; Šavel, J.; Košin, P.; Dostálek, P. Bottle Conditioning: Technology and Mechanisms Applied in Refermented Beers. *Beverages* **2020**, *6*, 56. [CrossRef]
16. Suriano, S.; Iannucci, A.; Codianni, P.; Fares, C.; Russo, M.; Pecchioni, N.; Marciello, U.; Savino, M. Phenolic acids profile, nutritional and phytochemical compounds, antioxidant properties in colored barley grown in southern Italy. *Food Res. Int.* **2018**, *113*, 221–233. [CrossRef] [PubMed]
17. Bao, J. Rice starch. In *Rice Chemistry and Technology*; Bao, J., Ed.; Woodhead Publishing and AACC International Press: Duxford, UK, 2018; pp. 55–108. [CrossRef]
18. Usansa, U.; Burberg, F.; Geiger, E.; Back, W.; Wanapu, C.; Arendt, E.K.; Kreisz, S.; Boonkerd, N.; Teaumroong, N.; Zarnkow, M. Optimization of malting conditions for two black rice varieties, black non-waxy rice and black waxy rice (*Oryza sativa* L. *indica*). *J. Inst. Brew.* **2011**, *117*, 39–46. [CrossRef]
19. Meier-Dörnberg, T.; Kory, O.I.; Jacob, F.; Michel, M.; Hutzler, M. Saccharomyces cerevisiae variety diastaticus friend or foe?—spoilage potential and brewing ability of different Saccharomyces cerevisiae variety diastaticus yeast isolates by genetic, phenotypic and physiological characterization. *FEMS Yeast Res.* **2018**, *18*, 1–27. [CrossRef]
20. Henson, C.A.; Duke, S.H.; Vinje, M.A. Comparison of Barley Malt Amylolytic Enzyme Thermostabilities and Wort Sugars Produced During Mashing. *J. Am. Soc. Brew. Chem.* **2014**, *72*, 51–65. [CrossRef]

21. McMurrough, I.; Madigan, D.; Donnelly, D.; Hurley, J.; Doyle, A.-M.; Hennigan, G.; McNulty, N.; Smyth, M.R. Control of Ferulic Acid and 4-Vinyl Guaiacol in Brewing. *J. Inst. Brew.* **1996**, *102*, 327–332. [CrossRef]
22. Wu, D.; Zhou, T.; Li, X.; Cai, G.; Lu, J. POD promoted oxidative gelation of water-extractable arabinoxylan through ferulic acid dimers. Evidence for its negative effect on malt filterability. *Food Chem.* **2016**, *197*, 422–426. [CrossRef]
23. Szwajgier, D.; Pielecki, J.; Targoński, Z. The Release of Ferulic Acid and Feruloylated Oligosaccharides During Wort and Beer Production. *J. Inst. Brew.* **2005**, *111*, 372–379. [CrossRef]
24. Pulvirenti, A.; De Vero, L.; Blaiotta, G.; Sidari, R.; Iosca, G.; Gullo, M.; Caridi, A. Selection of Wine *Saccharomyces cerevisiae* Strains and Their Screening for the Adsorption Activity of Pigments, Phenolics and Ochratoxin A. *Fermentation* **2020**, *6*, 80. [CrossRef]
25. Jiao, Y.; Zhang, Y.; He, Z.; Zhai, W.; Gong, H.; Yang, Z. Effect of Ferulic Acid on the Formation of Pyranoanthocyanins from Purple Corn (*Zea mays* L.) Cob in a Model System and Their Effects on Color. *Int. J. Food Prop.* **2015**, *19*, 847–858. [CrossRef]
26. Echeverrigaray, S.; Scariot, F.J.; Menegotto, M.; Delamare, A.P.L. Anthocyanin adsorption by Saccharomyces cerevisiae during wine fermentation is associated to the loss of yeast cell wall/membrane integrity. *Int. J. Food Microbiol.* **2020**, *314*, 108383. [CrossRef]
27. de Freitas, V.; Mateus, N. Formation of pyranoanthocyanins in red wines: A new and diverse class of anthocyanin derivatives. *Anal. Bioanal. Chem.* **2011**, *401*, 1463–1473. [CrossRef]
28. Outtrup, H. Proanthocyanidins, The Brewing Process, and The Quality of Beer. In *Basic Life Science; Plant Polyphenols: Synthesis, Properties, Significance*; Hemingway, R.W., LAKS, P.E., Eds.; Springer: Boston, MA, USA, 1992; Volume 59, pp. 849–858. [CrossRef]
29. Ditrych, M.; Kordialik-Bogacka, E.; Czyżowska, A. Antiradical and reducing potential of commercial beers. *Czech J. Food Sci.* **2016**, *33*, 261–266. [CrossRef]
30. Socha, R.; Pająk, P.; Fortuna, T.; Buksa, K. Antioxidant activity and the most abundant phenolics in commercial dark beers. *Int. J. Food Prop.* **2017**, *20* (Suppl. 1), S595–S609. [CrossRef]
31. Ceccaroni, D.; Sileoni, V.; Marconi, O.; De Francesco, G.; Lee, E.G.; Perretti, G. Specialty rice malt optimization and improvement of rice malt beer aspect and aroma. *LWT* **2019**, *99*, 299–305. [CrossRef]