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Production and Characterization of Kombucha Tea from Different Sources of Tea and Its Kinetic Modeling

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Abstract: This study aimed to investigate the fermentation performance, sugar consumption, pH changes, total phenolic compounds, and antioxidant activity produced using different tea extracts and sugar concentrations and the kinetic characteristics of Kombucha fermentation. Three independent sugar concentrations (10 g/L, 40 g/L, and 70 g/L) were used in the fermentation process. The results showed that the Kombucha culture consumed all sugar in the fermentation medium when the sugar concentration was below a certain threshold, but when the sugar concentration was high, not all substrate was consumed. Sugar consumption values ranged from 48.39 to 55.40 g/L and affected biomass formation, with higher sugar consumption resulting in increased biomass production. The pH decreased during fermentation due to the production of organic acids and microbial by-products, while total acidity increased. Total phenolic compounds increased during fermentation, with the highest concentrations observed in herbal Kombucha teas. Antioxidant activity varied, with some samples showing a decrease in DPPH scavenging ability. Kinetic characterization revealed the relationship between substrate depletion, sugar consumption, total acidity, and phenolic compound production. The results showed that sugar concentration influenced the fermentation kinetics and end-product characteristics of Kombucha tea. Overall, this study provides valuable insights into the fermentation process of Kombucha tea and its impact on various parameters, contributing to the understanding of the factors affecting its quality and health benefits.

Keywords: Kombucha fermentation; bioactive component; proximate composition; kinetic parameters; kinetic modeling

Citation: Tarhan Kuzu, K.; Aykut, G.; Tek, S.; Yatmaz, E.; Germec, M.; Yavuz, I.; Turhan, I. Production and Characterization of Kombucha Tea from Different Sources of Tea and Its Kinetic Modeling. *Processes* **2023**, *11*, 2100. https://doi.org/10.3390/pr11072100

Academic Editor: Chi-Fai Chau

Received: 11 June 2023
Revised: 8 July 2023
Accepted: 11 July 2023
Published: 14 July 2023

1. Introduction

Kombucha has been consumed all over the world but historically in China, Russia, and Eastern European countries. Kombucha is a fermented sugared black tea by yeasts and *Acetobacter* species [1,2]. The various yeast species of Kombucha tea are *Brettanomyces bruxellensis* [3], *Candida stellata* [3], *Schizosaccharomyces pombe* [3], *Torulaspora delbrueckii* [3], *Zygosaccharomyces bailii* [3], *Saccharomycodes ludwigii* [4], *Kloeckera apiculata* [5], *Saccharomyces cerevisiae* [4], *Brettanomyces lambicus* [6], *Brettanomyces custersii* [6], *Candida krusei* [5], and *Pichia* species [7]. This means that Kombucha culture differs from place to place and could easily be understood from different research about Kombucha cultures [2]. Namely, the kombucha culture is a symbiotic culture of bacteria and yeast (SCOBY) that is essential for the fermentation process of Kombucha [2]. The role of yeasts in the Kombucha fermentation is to hydrolyze sucrose from the cultivation medium to glucose and fructose and metabolize these monosaccharides to ethanol, which is further oxidized to acetic acid by acetic acid bacteria (AAB). AAB cannot uptake sucrose alone because of the lack of

enzymes for the extracellular hydrolysis of sucrose or its transport into the cell. AAB uses yeast-derived glucose to synthesize gluconic acid and bacterial cellulose in the form of a pellicle, which is commonly described as the "fungus" [8–10]. Microbial community type and composition play an important role in the biochemistry dynamics of Kombucha. These associations help to decrease pH and reduce microbial growth of other microorganisms with antimicrobial metabolites [11]. The time of Kombucha fermentation is between 7 and 60 days. During this time, biological activities increase. On the other hand, it was reported that the best results were yielded in an average of 10 days [12]. According to the Food and Drug Administration Model Food Code for Kombucha Brewing, more than 10 days of fermentation are not suggested if produced for human consumption [13]. Therefore, 8–10 days can be enough to obtain the best beverage specifications, and microorganisms use sugar to produce value-added acids and antimicrobial metabolites [14]. The kombucha tea yielded after fermentation consists of sugars (glucose, fructose), gluconic, glucuronic, L-lactic, acetic, malic, tartaric, malonic, citric, and oxalic acids, as well as ethanol, 14 amino acids, water-soluble vitamins, antibiotically active matters, and some hydrolytic enzymes [15].

Kombucha tea has beneficial features on human health, such as improving the immune system, detoxifying harmful substances, lowering blood pressure, treating gastritis and cholesterol, and exhibiting antioxidant, antibacterial, anticancer, and antidiabetic activities [2]. The research about the antimicrobial activity of Kombucha tea showed that the antimicrobial agent was acetic acid content, and it inhibited *Agrobacterium tumefaciens*, *Bacillus cereus*, *Salmonella choleraesuisserotypetyphimurium*, *Staphylococcus aureus*, and *Escherichia coli*. However, due to the fermented samples including 33 g/L total acid (7 g/L acetic acid), these values indicated the yielded beverage samples were not suitable for drinkable levels, but Kombucha had antimicrobial activity against pathogenic bacteria [16]. The other research demonstrated that Kombucha tea had an antimicrobial effect against a range of pathogenic bacteria, several clinical *Candida* species, fermented *L. citriodora*, and *F. vulgare* [17]. Kombucha could also be used against enteropathogenic bacterial infections due to its polyphenolic content [18]. Various Kombucha cultures also showed different antioxidant activity under the same fermentation conditions (10% starter addition to the fresh medium prepared, 30 °C, and 15 days fermentation time), mostly indicating time-dependent properties [19]. The conformable research showed the difference between antioxidant activity values from different starter cultures and tea extracts [15]. The Kombucha fermentation with different initial sucrose concentrations (ISCs) (70 g/L, 50 g/L, and 35 g/L of sucrose) was studied, and the highest sugar concentration value was found to be an optimal concentration of carbon source, providing high pH, low acetic acid, and high L-lactic acid content and highest sucrose consumption [20].

In the literature, there are some similar studies regarding the production of Kombucha tea from different types of herbal and fruit teas. For instance, Zubaidah et al. [21] examined the physical, chemical, and microbiological features of Kombucha from different varieties of apples (Anna, Manalagi, Fuji, Granny Smith, Red Delicious, Rome Beauty, and Royal Gala). Based on the results, it was reported that the best treatment was yielded on Fuji varieties of Kombucha apple (total acid 1.33%, pH 2.95, total phenol 268.57 µg/mL GAE, total sugar 6.74%, antibacterial activity against *Staphylococcus aureus* 21.30 mm, antibacterial activity *Escherichia coli* 21.20 mm, antioxidant activity 35.62%, organoleptic aroma 3.55, taste 3.3, and color 3.4 (on a scale of 1–5)) [21]. In another study, where the different carbon sources (glucose, fructose, xylose, lactose, sucrose (70 g/L)), types of teas (black tea, green tea, sage tea, pomegranate (hibiscus) tea, blueberries tea, and rosehip tea), and coffee were used as resources to produce Kombucha [22], the pH, acidity, antioxidant activity, phenolic substance, biomass development, color change, organic acid profile, ethanol, and sensory analysis were examined. The results indicated that the value of pH decreased during fermentation, and the Kombucha from fruit teas were greater acidity than herbal teas and coffee extract. The phenolic substance content and antioxidant activity of the Kombucha produced have been found to have the potential to be an important product. Regarding biomass growth, it was determined most in glucose and sucrose (tea samples) and lactose

(coffee extract) and at the least in fructose (tea samples) and lactose (coffee extract). When color changes were examined, it was detected that the L, a, and b values of herbal tea changed in a fermentation medium supplemented with glucose, xylose, or fructose. During the fermentation, most of the organic acids, including oxalic acid, tartaric acid, malic acid, lactic acid, citric acid, succinic acid, and fumaric acid, were measured. On the other hand, it was reported that no ethanol production was observed at the end of the fermentation. Based on the sensory analysis, the most and least preferred Kombucha teas were produced from pomegranate and sage teas, respectively [22]. In a different study, Tamer et al. [23] evaluated the bio-accessibility and functional features of Kombucha teas fortified with different medicinal plant extracts (linden, lemon balm, sage, *Echinacea*, mint, and cinnamon). Based on the results, the antioxidant capacity (AC), ferric-reducing antioxidant power, and cupric-reducing AC were 13.96%, 48.90%, and 55.54%, respectively. It was also found that during 9-day storage, the bio-accessibility of total phenolic and AC dramatically increased after gastric and intestinal digestion [23]. Additionally, the changes in the content of organic acids and polyphenols during the Kombucha fermentation from green tea, black tea, and tea manufacturing waste [24] and the antibacterial and antifungal activities of black and green Kombucha teas [25] were also examined. Moreover, the kinetics of sucrose fermentation by Kombucha culture was also studied by using Boltzmann's functions [26]. The fermentation conditions were performed on 1.5 g/L of black tea, with 67 g/L of sucrose, and using 10% or 15% of Kombucha culture (v/v). The model was described as a sigmoid function at two different temperatures (22 °C and 30 °C). Based on the results, it was determined that the rate of fermentation was maximum on days 4–5, and after reaching the maximal rate, it dramatically decreased. It was reported that as the temperature and inoculum concentration increased, the rate of the fermentation increased, the optimal fermentation time was 3.5–5 days under the implemented circumstances, and the saturation curves indicated the sigmoid kinetics at the selected sucrose concentration [26]. When considering this information, this study has novelty in terms of the use of some different types of teas in the production of Kombucha teas, kinetic characterization of Kombucha fermentations performed at different substrate concentrations, and kinetic modeling of Kombucha fermentations in terms of substrate consumption and total acidity. Therefore, this study is filled the significant gap in the literature.

Kombucha tea is generally produced from black and green tea, but commercial firms' market started to produce new Kombucha teas with lemon, apples, peach, blackberries, and rosehip. Therefore, the objective of this study is to investigate the production of Kombucha teas with diverse chemical compositions by utilizing various substrates. Additionally, the study seeks to analyze the kinetic properties of Kombucha fermentation and develop a kinetic model for fermentations involving different tea sources.

2. Materials and Methods

2.1. Kombucha Culture and Media

Kombucha culture was obtained from the commercial firm "Comboutea" (Tema Pharmaceutical Vitamin Cosmetics Limited Company, Samsun, Turkey). The media components for stock and pre-culture were 10 g/L yeast extract, 20 g/L glucose, and 20 g/L peptone [27]. After the medium composition was prepared, the medium pH was adjusted to 4 using 10 N HCl. The prepared medium was sterilized at 121.1 °C for 15 min. Subsequently, the medium was cooled to room temperature. It was inoculated with 10% (v/v) of Kombucha culture. The stock and pre-cultures were incubated at 24 °C for 10 days, and the stock cultures were stored at 4 °C. Stock cultures were renewed for one month to have viability and productivity.

2.2. Experimental Design

In this study, different fruit (bilberry, rosehip, apple, and pomegranate tea) and herbal (green, sage, linden, and black tea) teas were used for tea extraction. On the other hand, the limited sugar value to produce Kombucha from different types of tea was determined

from a previous study [15]. After determining the maximum sugar limit, sucrose as the sole carbon source was added to the fermentation medium by decreasing it to 30 g/L to instigate the effect of the initial sucrose concentration. Thus, the ISCs were 10 g/L, 40 g/L, and 70 g/L in the present study. Each of the extracts was prepared with three different ISCs (10 g/L, 40 g/L, and 70 g/L), and a coded system for the samples is given in Table 1. All production and analyses were replicated two times. Kinetic parameters of Kombucha fermentation were also calculated. Fermentations were kinetically modeled using the logistic model (LM) and the Luedeking–Piret model (LPM) [28]. The LM was used to predict the experimental substrate consumption values, and LPM was employed to estimate the experimental total acidity values of fermentation.

Table 1. Sample code system.

Tea Origin	Tea	Initial Sugar Concentration (g/L)		
		10	40	70
Herbal tea	Sage tea (ST)	ST-10	ST-40	ST-70
	Linden tea (LT)	LT-10	LT-40	LT-70
	Green tea (GT)	GT-10	GT-40	GT-70
	Black tea (BT)	BT-10	BT-40	BT-70
Fruit tea	Apple tea (AT)	AT-10	AT-40	AT-70
	Rosehip tea (RT)	RT-10	RT-40	RT-70
	Pomegranate tea (PT)	PT-10	PT-40	PT-70
	Bilberry tea (BBT)	BBT-10	BBT-40	BBT-70

2.3. Preparation of Tea Extracts, Inoculation, and Fermentation

Four different herbal teas (green (GT), sage (ST), linden (LT), and black tea (BT)) and four various fruit teas (bilberry (BBT), rosehip (RT), apple (AT), and pomegranate tea (PT)) were used for Kombucha production. All the tea samples were provided by the Unilever Company in Konya, Turkey.

The extraction process was realized by mixing 1 L of boiled pure water with 10 g tea and waiting for 15 min to obtain tea extracts [29]. The mix was filtered by using roughing filter paper (cellulosic filter paper) to separate the insoluble materials. After filtration, different amounts of sucrose (10 g/L, 40 g/L, or 70 g/L) were immediately added. After the sugar was completely dissolved, the mixture was transferred into 250 mL flasks (100 mL working volume) and cooled to room temperature. It was stored in appropriate conditions until inoculation.

After pre-culture and fermentation media were prepared, the flasks were inoculated with 10 mL of pre-culture. Inoculated sugared tea mixtures were incubated at 24 °C for 10 days with no agitation, and samples were taken daily under aseptic conditions and stored at 4 °C [30].

2.4. Analysis

The total acidity was determined by adding 0.1 N NaOH to samples until the pH was 8.2 [31]. The pH values of fermented teas were measured with an electronic pH meter (Thermo Scientific Orion 4 Star, Singapore). The total biomass of fermented samples was gravimetrically determined. The collected samples during fermentation were filtered by using pre-weighed filter paper (Whatman No.: 1), and the fermented broth was removed. The filter cake (biomass) was then dried at 60 °C in the oven until constant weight [32]. The residual sugar concentration was spectrophotometrically determined using the 3,5-dinitrosalicylic acid method [33]. The Folin–Ciocalteu method was used to determine the total phenolic substance concentration in samples. The results were given as milligrams of gallic acid equivalents per liter (mg GAE/L) of Kombucha [34]. The antioxidant analysis was determined with the α, α-diphenyl-β-picrylhydrazyl (DPPH) free radical scavenging method, and the antioxidant capacity was determined as % inhibition [19].

2.5. Kinetic Parameters

Kinetic parameters including substrate consumption (ΔS, g/L), maximum substrate consumption rate (Q_S, g/L/d), substrate utilization yield (η, %), biomass production (ΔX, g/L), maximum biomass production rate (Q_X, g/L/d), biomass yield ($Y_{X/S}$, g biomass/g substrate), total acidity (TA, %), maximum total acidity production rate (Q_{TA}, %/d), phenolics production (ΔPH, mg/L), maximum phenolics production rate (Q_{PH}, mg/L/d), and phenolics yield ($Y_{PH/S}$, mg phenolics/g substrate) were calculated. The details regarding how kinetic parameters are calculated can be found in previous similar studies [28].

2.6. Kinetic Modeling

The LM (Equation (1)) and LPM (Equation (2)) were utilized to estimate the experimental substrate consumption and total acidity data of Kombucha fermentation. Microsoft Office Excel 2013 was used. Traditionally, LM is used to describe cell growth. However, in this work, the model was modified to define sugar consumption and independently employed cell growth data because there is no sigmoid growth of biomass because of high acidity or low pH values.

$$\frac{-dS}{dt} = \mu_{m,S}\left[1 - \frac{S}{S_m}\right]S \quad (1)$$

where −dS/dt is the substrate consumption rate (g/L/d), $\mu_{m,S}$ is the specific sugar consumption rate (1/d), S is the residual substrate concentration at the time "t" (g/L), and S_m is the maximum substrate concentration (g/L).

The LPM is utilized to define the metabolite production rate (dP/dt) related to cell growth. However, dP/dt is also thought to be contingent on both momentary S and dS/dt linearly [35].

$$\frac{dP}{dt} = \alpha\frac{dS}{dt} + \beta S \quad (2)$$

where dP/dt is the total acidity rate (%/d) and α and β are the empirical constants that vary based on fermentation conditions and are determined with the best appropriate real data. Moreover, the determination of coefficient (R^2) was utilized to comprehend whether modeling is accomplished or not [28].

2.7. Statistical Analysis

The SAS version 7 program (Statistical Analysis System, TS P1, Cary, NC, USA) was used for the statistical evaluation of the data obtained from the study, and variance analysis was performed. Significant differences were evaluated by the Duncan Multiple Comparison Test at a confidence level of 95%.

3. Results and Discussion

3.1. Sugar Consumption and Biomass Production

Three ISCs were used to determine the fermentation performance of the commercial Kombucha culture. It was determined that all the sugar in the fermentation medium is consumed by the Kombucha culture in fermentations performed with AT-10, RT-10, PT-10, and BT-10. However, when the fermentation medium contains high sugar concentration, all the substrate in the medium is not consumed by the Kombucha culture at the end of the fermentation (Figure 1A and Supplementary Materials). The highest sugar consumption value is 55.40 g/L for LT-70 herbal Kombucha tea, whereas its highest value is 48.39 g/L for AT-70 fruit Kombucha tea (Figure 1A and Table 2). These sugar consumption values also affect biomass formation. In Figure 1B and the Supplementary Materials, biomass production curves from different medium compositions are given. The difference in the medium used, amount of sugar, the composition of culture, fermentation conditions, and applied period are effective in the chemical composition of Kombucha [36]. Moreover, ISC affects total sugar consumption. High sugar consumption is observed in fermentations with high ISCs (Table 2). It is determined that residual sugar concentration decreases

during fermentation. Furthermore, the highest biomass productions for herbal and fruit Kombucha teas are determined as 6.46 g/L for GT-70 and 8.77 g/L for PT-70 (Figure 1B). These results show that biomass formation increases with an increase in sugar consumption (Table 2). In work conducted by Muhialdin et al. [37], the amount and yield of biomass were associated with the sugar source.

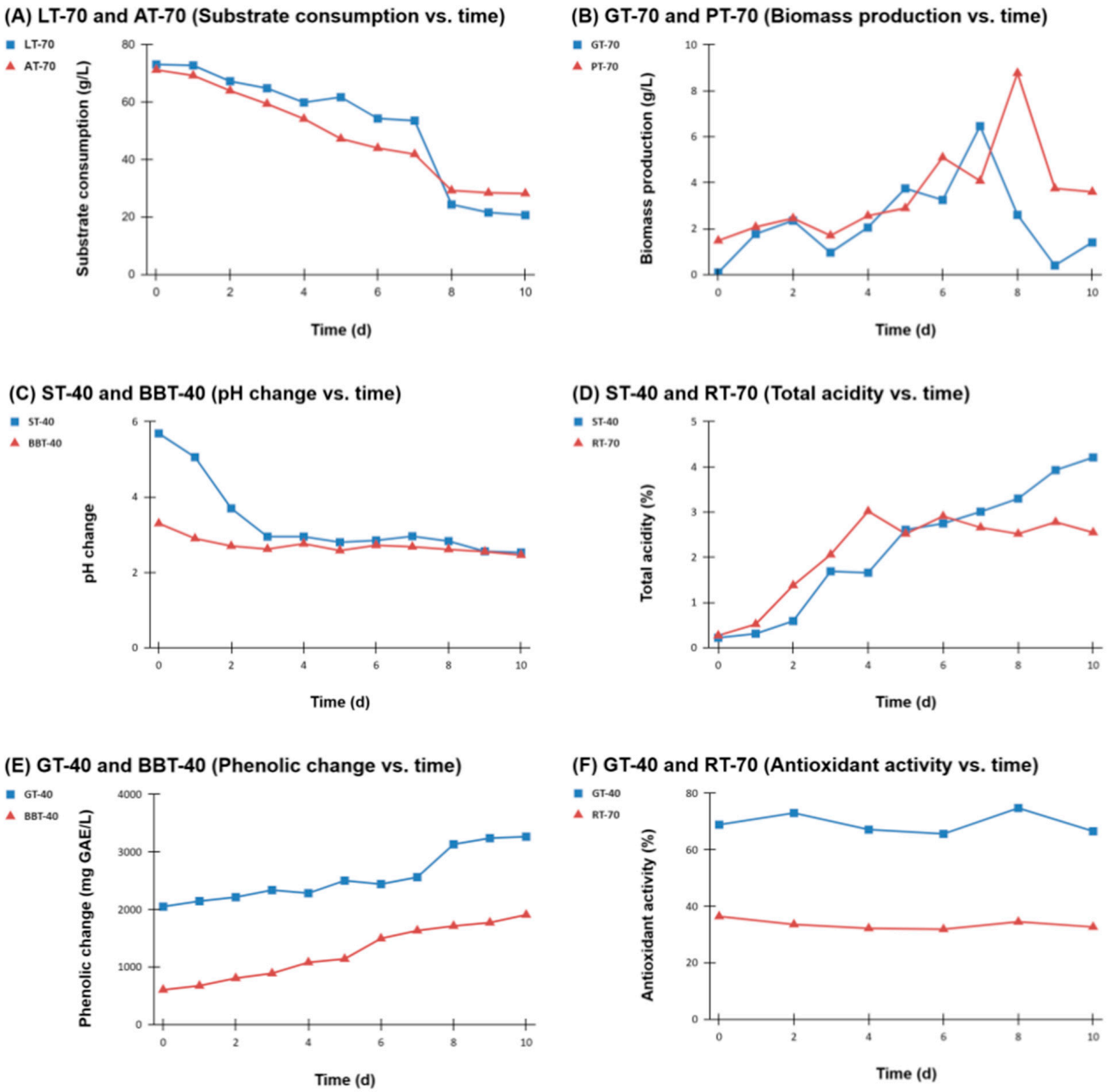

Figure 1. Change of substrate concentration, biomass production, pH, total acidity, phenolic, and antioxidant activity during Kombucha fermentation performed on media containing different initial sugar concentrations. (**A**) Substrate consumption vs. time with sage tea (ST-70) and linden tea (LT-70). (**B**) Biomass production vs. time with green tea (GT-70) and pomegranate tea (PT-70). (**C**) pH change vs. time with sage tea (ST-40) and blueberry tea (BBT–40). (**D**) Total acidity vs. time with sage tea (ST-40) and rosehip tea (RT-70). (**E**) Phenolic change vs. time with green tea (GT-40) and blueberry tea (BBT-40). (**F**) Antioxidant activity vs. time with green tea (GT-40) and rosehip tea (RT-70).

Table 2. Kinetic parameters of Kombucha fermentation performed with extracts from different types of tea with various initial sugar concentrations.

Tea	[Substrate]	Kinetic Parameters										
		ΔS (g/L)	**Q_S (g/L/d)**	**η (%)**	**ΔX (g/L)**	**Q_X (g/L/d)**	**$Y_{X/S}$ (g/g)**	**ΔTA (%)**	**Q_{TA} (%/d)**	**ΔPH (mg/L)**	**Q_{PH} (mg/L/d)**	**$Y_{PH/S}$ (mg/g)**
ST	10 g/L	8.52	1.29	79.48	0.72	0.12	0.08	0.17	0.08	445.96	97.18	52.34
	40 g/L	13.10	3.71	29.39	1.74	0.51	0.13	4.10	0.60	883.17	169.75	67.42
	70 g/L	26.62	4.05	32.71	3.96	0.94	0.15	1.51	0.28	900.31	189.07	33.82
LT	10 g/L	11.66	2.72	97.25	1.02	0.22	0.09	0.59	0.17	242.12	26.80	20.77
	40 g/L	19.85	4.00	47.40	2.72	1.20	0.14	1.04	0.27	399.65	49.54	20.13
	70 g/L	55.40	11.28	72.78	3.80	0.14	0.07	1.63	0.31	227.06	48.14	4.10
GT	10 g/L	12.13	2.40	88.80	0.74	0.26	0.06	0.35	0.15	1217.60	410.68	100.38
	40 g/L	32.43	4.90	56.65	2.52	0.78	0.08	0.87	0.33	1215.90	345.69	37.49
	70 g/L	44.07	6.34	53.72	6.46	1.22	0.15	1.42	0.37	1268.40	264.16	28.78
AT	10 g/L	15.75	2.82	100.00	1.09	0.34	0.07	0.34	0.15	321.98	64.14	20.44
	40 g/L	18.61	3.07	39.89	3.76	1.39	0.20	1.18	0.27	785.45	159.90	42.21
	70 g/L	48.39	5.28	63.15	4.28	1.37	0.09	1.64	0.41	466.38	74.71	9.64
RT	10 g/L	15.43	3.65	100.00	2.01	0.23	0.13	1.07	0.36	130.66	21.39	8.47
	40 g/L	28.74	6.37	54.33	4.71	2.17	0.16	1.61	0.49	295.36	76.28	10.28
	70 g/L	45.66	8.32	51.38	5.60	1.31	0.12	2.77	0.82	255.86	56.83	5.60
PT	10 g/L	10.33	2.47	100.00	2.20	0.91	0.21	0.39	0.20	549.89	95.36	53.23
	40 g/L	21.04	3.67	44.69	3.25	1.43	0.15	1.30	0.30	800.04	101.66	38.02
	70 g/L	42.92	6.36	50.61	8.77	1.66	0.20	2.18	0.34	930.94	166.20	21.69
BBT	10 g/L	12.67	2.54	95.62	0.81	0.31	0.06	0.44	0.18	655.20	95.51	51.71
	40 g/L	16.05	3.07	32.56	2.07	0.70	0.13	1.52	0.39	1311.92	187.92	81.74
	70 g/L	21.41	3.24	26.02	3.55	1.40	0.17	1.73	0.41	782.90	178.13	36.57

ΔS, substrate consumption (g/L); Q_S, maximum substrate consumption rate (g/L/d); η, substrate utilization yield (%); ΔX, biomass production (g/L); Q_X, maximum biomass production rate (g/L/d); $Y_{X/S}$, biomass yield (g biomass/g substrate); ΔTA, total acidity (%); Q_{TA}, maximum total acidity production rate (%/d); ΔPH, phenolic production (mg/L); Q_{PH}, maximum phenolic production rate (mg/L/d); and $Y_{PH/S}$, phenolic yield (mg phenolic/g substrate).

3.2. pH and Total Acidity

Kombucha fermentation was performed with herbal and fruit tea extracts for 10 days, and pH changes are shown in Figure 1C and the Supplementary Materials. Herbal and fruit tea extracts' initial pH values range from 4.86 to 5.90 and from 3.12 to 3.53, respectively. Differences between initial and final values of pH are higher for herbal tea samples than for fruit tea samples (Figure 1C and Supplementary Materials). The lowest final pH values of herbal and fruit teas are measured to be 2.53 for ST-40 and 2.46 for BBT-40 (Figure 1C), and their highest values are 3.73 for ST-10 and 3.57 for AT-10 (Supplementary Materials). It is seen that a slight pH increase at the end of Kombucha fermentation in AT supplemented with 10 g/L sucrose (Supplementary Materials). This may be due to the breakdown of dead cells in the fermentation medium [38]. All pH values, except for that of AT-10, decrease during fermentation because microorganisms metabolize sugar into different metabolites, such as organic acids and by-products (Figure 1C and Supplementary Materials). For all samples, a significant decrease in pH is noticed between days 0 and 5, and changes in pH are statistically insignificant after day 5 ($p < 0.05$). Total acidity values in all herbal and fruit tea samples are below 0.30% acetic acid at the beginning of fermentation. Initial total acidity values differ from 0.04% to 0.22% for herbal tea extracts and 0.15% to 0.27% for fruit tea extracts (Figure 1D and Supplementary Materials). The highest and lowest total acidity values for herbal and fruit teas are determined to be 4.21% (ST-40) (Figure 1D), 0.15% (GT-10) (Supplementary Materials), and 2.55% (RT-70) (Figure 1D), 0.23% (AT-10) (Supplementary Materials). In general, when examining Figure 1D and the Supplementary Materials, an increase in total acidity values is observed as a result of organic acid production and dead cell fragmentation during fermentation [26]. These results show that organic acid production and microbial by-products, which occur in

parallel with sugar consumption during fermentation, decrease pH and increase total acidity (Figure 1D and Supplementary Materials).

3.3. Total Phenolic Compounds and Antioxidant Activity

The changes in total phenolic compounds for herbal and fruit Kombucha teas are given in Figure 1E and the Supplementary Materials. The phenolic concentration increases in almost all herbal (except for those of BT-40 and BT-70) and fruit Kombucha tea experiments at the end of fermentation. The highest total phenolic compound values are determined to be 1522.90 mg GAE/L in ST-40, 1061.55 mg GAE/L in LT-10, and 3266.05 mg GAE/L in GT-40 (Figure 1E), and 1025.67 mg GAE/L in BT-10 for herbal Kombucha teas after 10 days fermentation (Supplementary Materials). For fruit Kombucha teas, the highest total phenolic compound values are calculated to be 1041.21 mg GAE/L in AT-40, 1016.99 mg GAE/L in RT-40, 1420.44 mg GAE/L in PT-40 (Supplementary Materials), and 1907.53 mg GAE/L in BBT-40 (Figure 1E) after 10 days fermentation. When considering all fermentations, the highest increase in total phenolic substance concentration is yielded as 215.44% with BBT-40. An increase in phenolic content with fermentation may be related to the enzymes of mixed Kombucha culture, the acidic environment of Kombucha tea, the synergistic effect of different components in tea, and the breakdown of complex phenolic compounds [39]. Moreover, the fact that phenolic components are more stable at acidic pH may cause differences in the total amount of phenolic substances during fermentation [40]. The decline in total phenolic concentration in black Kombucha tea samples (BT-40 and BT-70) might be due to the characteristics of black tea of that season. The total amount of phenolic substances in green tea is higher than in black tea [40]. Moreover, the percentage increase in total phenolic concentration in fruit Kombucha teas (average 112.49%) is higher than in herbal Kombucha teas (average 49.14%) (Figure 1). In summary, the total amount of phenolic compounds increased.

The antioxidant activity results are given in Figure 1F and the Supplementary Materials. The final DPPH scavenging ability decreases except for GT-70 (+4.46%), AT-10 (+5.99%), and AT-40 (+38.08%) assays. Decline values change from 0.86% to 31.97% for herbal Kombucha teas and 2.92% to 17.50% for fruit Kombucha teas. The maximal decrease is calculated to be 31.97% for LT-40. This decline could be about substrate and starter culture types. Because of research about the influence of starter culture on Kombucha fermentation, results show that DPPH scavenging ability is slightly increased in the first 3 days and decreased after day 3 of fermentation [15]. The final antioxidant values are lower than the initial values of this research. The highest final DPPH scavenging ability value is 66.59% for the GT-40 at the end of the fermentation (Figure 1F). In a study [19], half of eight different Kombucha samples showed a regular increase in antioxidant activity, while the rest of them had irregular and variable results. It was predicted that Kombucha is affected by different environments, sugar quantity, fermentation conditions, and ionization change that occurs during fermentation may cause this variability. The highest initial and final DPPH scavenging ability results are calculated in green tea samples (Figure 1F). All other herbal and fruit tea Kombucha samples are lower than green tea samples (Figure 1F and Supplementary Materials). It is also reported that the highest DPPH scavenging ability is generally obtained from green tea samples [41]. Moreover, the change in antioxidant activity is affected by tea type and fermentation temperature. DPPH scavenging ability, despite decreases and increases during fermentation, generally increases at the end of fermentation [41].

3.4. Kinetic Characterization

Based on the kinetic results given in Table 2, the minimum and maximum ΔS are determined as 8.52 g/L in ST-10 and 55.40 g/L in LT-70, respectively. As the sugar concentration in the fermentation medium increases, ΔS increases. However, this is not alone as an indicator that indicates the success of fermentation. Therefore, other kinetics regarding substrate depletion, Q_S, and η, were estimated. The results indicate that the lowest and

highest values of Q_S are 1.29 g/L/d in ST-10 and 11.28 g/L in LT-70, which are the same as ΔS. Moreover, as the substrate concentration increases, Q_S increases. The η was also determined, and the values of η range from 26.02 in BBT-70 to 100% in AT-10, RT-10, and PT-10. When the sugar amount added into the fermentation medium was minimum, almost all the sugar was consumed by the Kombucha culture. However, when the substrate concentrations in the fermentation medium are 40 g/L and 70 g/L, the η varies from 29.39% in ST-40 to 56.65% in GT-40 and 26.02% in BBT-70 to 72.78% in LT-70, respectively. Therefore, we can say that as the substrate concentration in the fermentation environment increases, η decreases in general.

Similar to the kinetics regarding substrate consumption, when the fermentation medium is enriched with 10 g/L sucrose, the minimum ΔX is 0.72 g/L for ST-10, whereas its maximum value is 2.20 g/L for PT-10. When 40 g/L sucrose is added into the fermentation medium, the lowest and highest values of ΔX are 1.74 g/L and 4.71 g/L in ST-10 and RT-40, respectively. Similarly, when the substrate concentration in the medium is 70 g/L, the ΔX varies from 3.55 to 8.77 g/L in BBT-70 and PT-70. Therefore, as the substrate concentration increases, ΔX increases. As for the Q_X, when the fermentation medium is supplemented with 10 g/L, 40 g/L, and 70 g/L of sucrose, the lowest and highest values of Q_X are calculated as 0.12 g/L/d and 0.91 g/L/d (ST-10 and PT-10), 0.51 g/L/d and 2.17 g/L/d (ST-40 and RT-40), and 0.14 g/L/d and 1.66 g/L/d (LT-70 and PT-70), respectively. Between both the minimum and maximum Q_X values, the highest Q_X values are yielded when 40 g/L substrate is added into the medium. Moreover, when the fermentation medium is supplemented with 10 g/L, 40 g/L, and 70 g/L, the lowest values of $Y_{X/S}$ are 0.06 g/g, 0.08 g/g, and 0.07 g/g, whereas their highest values are 0.21 g/g, 0.20 g/g, and 0.20 g/g, respectively. The minimum and maximum values of $Y_{X/S}$ at different substrate concentrations are highly close to each other. Although ΔX increases depending on the substrate concentration, this situation is not valid for the $Y_{X/S}$.

Regarding the kinetic results related to the total acidity, the minimum and maximum ΔTA values are determined as 0.17% and 1.07%, 0.87% and 4.10%, and 1.42% and 2.77% of ST-10 and RT-10, GT-40 and ST-40, and GT-70 and RT-70 when 10 g/L, 40 g/L, and 70 g/L sucrose are inserted into the medium, respectively. Except for the ΔTA values of the Kombucha fermentation of ST, as the substrate concentration increases, ΔTA values increase (Table 2). The lowest and highest Q_{TA} values are found as 0.08%/d and 0.36%/d (ST-10 and RT-10), 0.27%/d and 0.60%/d (LT-40 and ST-40), and 0.28%/d and 0.82%/d (ST-70 and RT-70) with 10 g/L, 40 g/L, and 70 g/L of sucrose concentration added into the medium. As it is in the values of ΔTA, except for Q_{TA} values of ST, Q_{TA} values increase with an increase in substrate concentration (Table 2).

The lowest values of ΔPH are 130.66 mg/L, 295.36 mg/L, and 227.06 mg/L with 10 g/L, 40 g/L, and 70 g/L sucrose concentrations inserted into the RT-10, RT-40, and LT-70 media, respectively. Contrarily, its maximum values are yielded as 1217.60 mg/L from GT-10, 1311.92 mg/L from BBT-40, and 1268.40 mg/L from GT-70. Except for the ΔPH values of ST, GT, and BT, the highest ΔPH values are obtained when 40 g/L sucrose is used in the medium (Table 2). Additionally, the minimum and maximum values of Q_{PH} are 21.39 mg/L/d and 410.68 mg/L/d, 49.54 mg/L/d and 245.69 mg/L/d, and 48.14 mg/L/d and 264.16 mg/L/d for RT-10 and GT-10, LT-40 and GT-40, and LT-70 and GT-70, respectively. It is realized that the Kombucha teas from GT supplemented with 10 g/L, 40 g/L, and 70 g/L sucrose give the highest phenolic substance amounts. As the substrate concentration in the GT-based medium increase, the values of Q_{PH} decrease. Conversely, the Q_{PH} increase with an increase in substrate concentration added into the ST- and PT-based media. For the rest of Q_{PH}, the maximum peak values of Q_{PH} are yielded when 40 g/L substrate concentration is added into the fermentation medium. Concerning the $Y_{PH/S}$, its lowest values are obtained to be 8.47 mg PH/g, 10.28 mg PH/g, and 4.10 mg PH/g substrate when the fermentation media are RT-10, RT-40, and LT-70, respectively. Maximum $Y_{PH/S}$ values are also calculated as 100.38 mg PH/g, 81.74 mg PH/g, and 36.57 mg PH/g substrate for GT-10, BBT-40, and BBT-70, respectively. As the sugar concentration in the

medium increases, the maximum $Y_{PH/S}$ value decreases. Moreover, it is determined that $Y_{PH/S}$ values decrease with an increase in the substrate levels of GT- and PT-based media. For the remaining media, except for that of the LT, the highest $Y_{PH/S}$ peak values are yielded when the media are enriched with 40 g/L sucrose (Table 2).

3.5. Kinetic Modeling

The observed values of total acidity were estimated by the LPM, while the actual values related to sugar depletion were predicted by the LM (Figure 2). Concerning the prediction of sugar depletion values, the observed and estimated sugar depletion curves are plotted vs. time in Figure 2. As indicated in Figure 2, ST, LT, GT, AT, RT, PT, and BBT, the experimental and estimated sugar depletion values are generally in good agreement, except for that of the Kombucha fermentation performed with ST supplemented with 40 g/L sucrose because its R^2 value (0.6038) is lower than 0.75 (Table 3). Indeed, the experimental data on days 4–7 of fermentation are overestimated by the LM (Figure 2, ST). Additionally, if the R^2 value is higher than 0.75, meaning that the model can be used to estimate the fermentation experimental data [42]. Therefore, the yielded R^2 values are found between 0.7731 and 0.9750, except for R^2 = 0.6038, demonstrating that the proposed model for substrate depletion adequately fits the actual data of substrate depletion. Additionally, the values of $\mu_{m,S,}$ and S_f are between 0.17 and 1.14 d^{-1} and 0.34 and 9.64 g/L, respectively. The minimum and maximum values of $\mu_{m,S}$ are obtained when the BBT supplemented with 40 g/L and 10 g/L sucrose is used in the production of Kombucha tea, respectively. The highest $\mu_{m,S}$ values are achieved when 10 g/L sucrose is added to the fermentation media (Table 3). The lowest and highest S_f values are also yielded with ST supplemented with 10 g/L sucrose and RT supplemented with 70 g/L sucrose, respectively. Nevertheless, the values of S_f increase with an increase in sucrose concentration added into the fermentation media in general (Table 3). Mahdinia et al. [35] predicted the observed substrate depletion data of Menaquinoe-7 fermentation in the biofilm reactor with glucose- or glycerol-based medium using the LM. Based on the modeling results, it was declared that the R^2 values of the process were determined between 0.953 and 0.991, indicating that the suggested models fit well with the experimental substrate consumption data. The values of $\mu_{m,S}$ for glucose- and glycerol-based media were 0.059 h^{-1} and 0.054 h^{-1}, respectively [35], which are all higher than those of this study (Table 3). Ilgın et al. [43] estimated the substrate consumption data of *Aspergillus niger* inulinase fermentation from carob extract under shake flask fermentation circumstances using the LM. From the calculations, it was reported that the values of $\mu_{m,S,}$ and S_f were found to be 0.062 h^{-1} and 0.93 g/L, respectively, showing that the $\mu_{m,S}$ value is greater than the results yielded from the current study while the S_f value is compatible with those of this study (Table 3). In another study in which *A. niger* inulinase fermentation of sugar beet molasses in the large-scale stirred tank bioreactor was taken place [44], the experimental data of substrate consumption were forecasted by using the LM; thus, the $\mu_{m,S}$, and S_f values are computed as 0.042 h^{-1} and 1.18 g/L, respectively. It is determined that the computed $\mu_{m,S,}$ and S_f values are highly consistent with the present study (Table 3). It was reported that the proposed model successfully fitted the experimental data of substrate depletion with a high R^2 value (R^2 = 0.9778) [44].

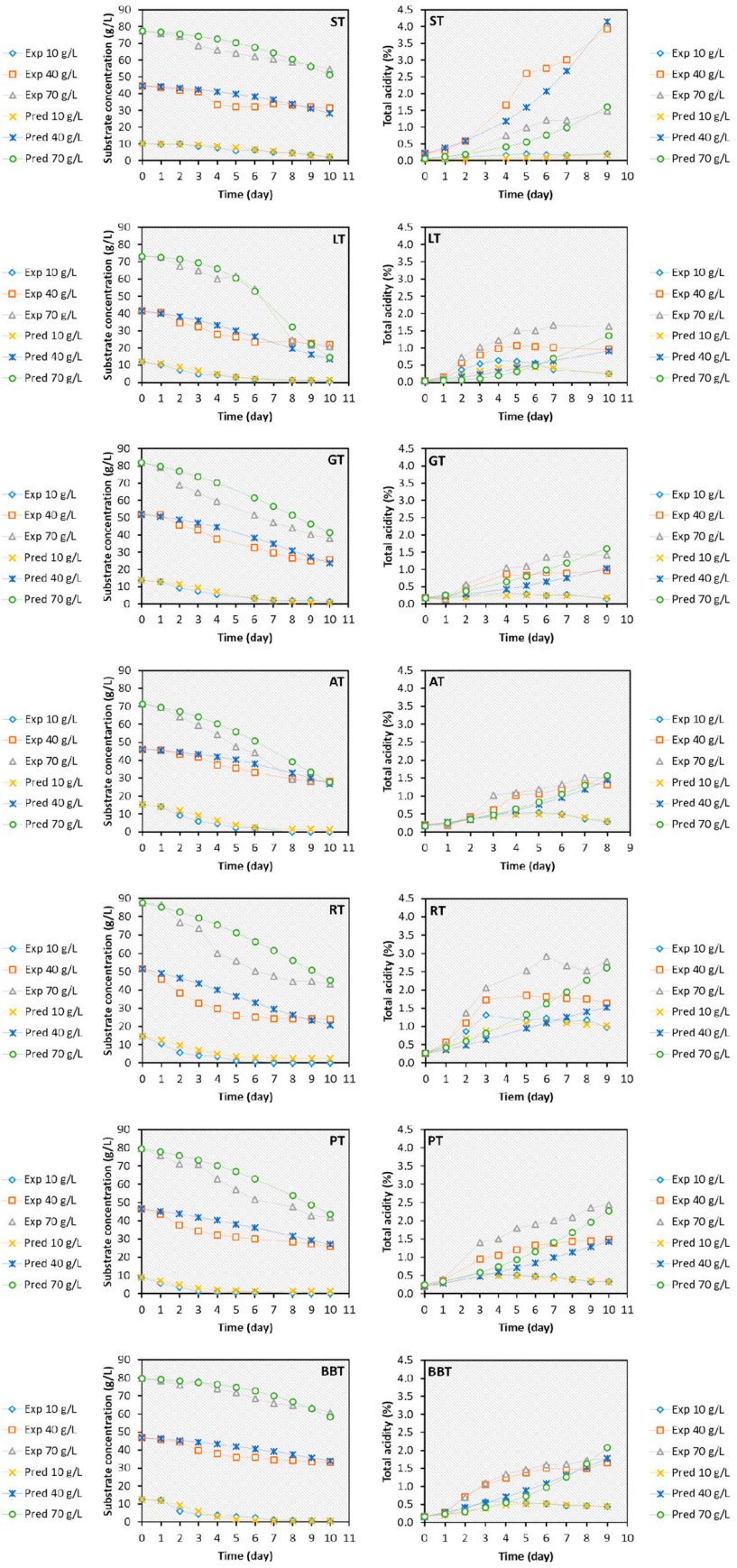

Figure 2. Substrate consumption and total acidity curves fitted by the LM and LPM.

Table 3. The model parameters calculated for kinetically modeling the Kombucha fermentation.

Tea	[Substrate]	Kinetics for Substrate Consumption				Kinetics for Acidity			
		$\mu_{m,S}$ (1/d)	S_m (g/L)	S_f (g/L)	R^2	β (%/gS.d)	α (%/gS)	A > β Fold	R^2
ST	10 (g/L)	0.4738	10.24	0.34	0.9438	0.0001	0.0201	360.19	0.5675
	40 (g/L)	0.2752	44.58	1.87	0.6038	0.0076	0.2584	34.00	0.9064
	70 (g/L)	0.2864	77.45	2.54	0.8572	0.0013	0.0662	49.85	0.8481
LT	10 (g/L)	0.7821	11.99	1.19	0.9672	−0.0058	0.0631	10.91	0.8060
	40 (g/L)	0.3462	41.42	3.89	0.8055	−0.0006	0.0343	60.27	0.5337
	70 (g/L)	0.5947	73.16	0.83	0.9655	0.0004	0.0211	55.41	0.4809
GT	10 (g/L)	0.7168	13.62	0.84	0.9487	−0.0024	0.0142	5.87	0.7586
	40 (g/L)	0.2909	52.03	4.44	0.9185	0.0005	0.0315	63.17	0.7220
	70 (g/L)	0.2503	82.03	9.18	0.9413	0.0001	0.0394	282.80	0.8259
AT	10 (g/L)	0.7798	15.34	1.24	0.9636	−0.0075	0.0468	6.24	0.9228
	40 (g/L)	0.2757	46.12	2.46	0.9012	0.0019	0.0863	45.75	0.8479
	70 (g/L)	0.3324	71.27	5.07	0.9563	0.0015	0.0365	23.65	0.8251
RT	10 (g/L)	0.7978	14.64	2.49	0.9671	−0.0030	0.0876	29.15	0.8465
	40 (g/L)	0.2765	51.46	9.47	0.7731	−0.0003	0.0475	145.00	0.6153
	70 (g/L)	0.2459	87.58	9.64	0.8645	−0.0006	0.0671	110.88	0.7225
PT	10 (g/L)	1.0969	8.94	1.39	0.9750	−0.0039	0.0554	14.30	0.9526
	40 (g/L)	0.2011	46.40	6.97	0.8137	0.0008	0.0568	75.30	0.8141
	70 (g/L)	0.2589	79.37	6.13	0.9332	0.0013	0.0493	39.13	0.8193
BBT	10 (g/L)	1.1422	12.71	0.52	0.9166	−0.0023	0.0371	16.34	0.7676
	40 (g/L)	0.1671	46.97	4.90	0.8257	0.0010	0.1349	129.31	0.7937
	70 (g/L)	0.3043	79.67	1.49	0.9088	0.0008	0.1122	135.44	0.6790

$\mu_{m,S}$, specific sugar consumption rate (1/d); S_m, maximum substrate concentration (g/L); S_f, final substrate concentration (g/L); R^2, determination of coefficient; β, empirical constant (%/gS.d); α, empirical constant (%/gS); ST, sage tea; LT, linden tea; GT, green tea; AT, apple tea; RT, rosehip tea; PT, pomegranate tea; and BBT, bilberry tea.

The actual and estimated total acidity values are also plotted vs. time and shown in Figure 2. It is detected that the values of R^2 range from 0.4809 to 0.9526. It can be said that those with R^2 values higher than 0.75 (in this case, they are ST-40, ST-70, LT-10, GT-10, GT-70, AT-10, AT-40, AT-70, RT-10, PT-10, PT-40, PT-70, BBT-10, and BBT-40) are adequately fitted by the LPM (Table 3). Therefore, most of the Kombucha fermentation from different tea extracts supplemented with different concentrations of sucrose is satisfactorily fitted by the LPM with the R^2 value greater than 0.75. Moreover, α and β values, which can change based on the fermentation circumstances, were estimated (Table 3). If $\alpha \neq 0$ and $\beta = 0$, then the total acidity is associated with substrate consumption. If $\alpha = 0$ and $\beta \neq 0$, then the total acidity is non-associated with substrate consumption. The values of β vary from −0.0075 to 0.0076%/gS.d, which are so close to zero, while the values of α range from 0.0142 and 0.2584%/gS. The values of α are 5.87 to 360.19 times greater than those of β (Table 3). Therefore, we can say that the total acidity is associated with substrate consumption. Mahdinia et al. [35] studied the kinetic modeling of Menaquinoe-7 fabrication from glucose and glycerol in the biofilm reactor using the LPM. The model parameters of the LPM, which are α and β, were calculated to be −0.138 mg/g and 0.00010 mg/g/h for the production in the glucose-based medium and −0.089 mg/g and 0.00301 mg/g/h for the production in glycerol-based medium, respectively. Therefore, the values of α were 1380- and 29.57-fold higher than those of β, respectively, showing that Menaquinoe-7 fabrication was associated with substrate consumption [35], as it is in the current study. To the best of our knowledge, there is no study regarding the kinetic modeling of Kombucha fermentation using different tea extracts enriched with different concentrations of sucrose as a carbon source. Therefore, this study is important in terms of contributing to science because of the information it contains.

4. Conclusions

In conclusion, the study focused on the fermentation performance of a commercial Kombucha culture using different herbal and fruit tea extracts with varying sugar concentrations. The results showed that the Kombucha culture effectively consumed all sugar in the fermentation medium when supplemented with AT-10, RT-10, PT-10, and BT-10, but when higher sugar concentrations were present, not all substrate was consumed. The highest sugar consumption value was 55.40 g/L for LT-70 herbal Kombucha tea, while AT-70 fruit Kombucha tea had the highest value at 48.39 g/L. These sugar consumption values were correlated with biomass production, showing an increase with higher sugar consumption. The pH and total acidity of the Kombucha teas changed during fermentation, with herbal teas showing more significant fluctuations than fruit teas. The organic acid production and microbial by-products during fermentation decreased pH and increased total acidity. Additionally, the total phenolic compounds and antioxidant activity increased during fermentation, with higher increases observed in fruit kombucha teas compared to herbal teas. Kinetic characterization revealed the relationships between sugar depletion, biomass production, total acidity, and phenolic compounds during fermentation. The values of different kinetic parameters varied depending on the sugar concentration in the fermentation medium. However, this study had some limitations, such as the lack of detailed analysis of the specific enzymes involved in the breakdown of complex phenolic compounds and the influence of environmental factors on fermentation variability. Furthermore, only a specific commercial Kombucha culture was used, which may not fully represent the diversity of Kombucha cultures available. Despite these limitations, this study provides valuable insights into the fermentation performance of Kombucha cultures under different conditions, which could be beneficial for further research and industrial applications.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/pr11072100/s1, Figure S1: Change of substrate concentration, biomass production, pH, total acidity, phenolic, and antioxidant activity during Kombucha fermentation performed on media containing different initial sugar concentrations. (A, H, and O): Sage tea (ST); (B, I, and Q): Linden tea (LT); (C, J, and P): Green tea (GT); (D, K, and R): Apple tea (AT); (E, L, and S): Rosehip tea (RT); (F, M, and T): Pomegranate tea (PT); and (G, N, and U): Bilberry tea (BBT).

Author Contributions: K.T.K. conceptualization, investigation, methodology, project administration, and writing—original draft preparation; G.A. conceptualization, investigation, methodology, project administration, and writing—original draft preparation; S.T. conceptualization, investigation, methodology, project administration, and writing—original draft preparation; E.Y. writing—review and editing, project administration, supervision, and formal analysis; M.G. writing—review and editing, formal analysis, software, and modeling; I.Y. writing—review and editing, formal analysis, software, and modeling; I.T. writing—review and editing, resources, supervision, conceptualization, and methodology. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: Not applicable.

Acknowledgments: This study was supported by the TUBITAK 2209-University Student Research Project Support Program.

Conflicts of Interest: The authors declare no conflict of interest.

Abbreviations

Abbreviations	Full Name
ISC	Initial sucrose concentration
AAB	Acetic acid bacteria
R^2	Determination of coefficient
α and β	Empirical constants
N	Normal
HCl	Hydrochloric acid
LPM	Luedeking–Piret model
LM	Logistic model
GT	Green tea
ST	Sage tea
LT	Linden tea
BT	Black tea
BBT	Bilberry tea
RT	Rosehip tea
AT	Apple tea
PT	Pomegranate tea
NaOH	Sodium hydroxide
GAE	Gallic acid equivalents
DPPH	α, α-diphenyl-β-picrylhydrazyl
ΔS	Substrate consumption, g/L
Q_S	Maximum substrate consumption rate, g/L/d
η	Substrate utilization yield, %
ΔX	Biomass production, g/L
Q_X	Maximum biomass production rate, g/L/d
$Y_{X/S}$	Biomass yield, g biomass/g substrate
TA	Total acidity, %
Q_{TA}	Maximum total acidity production rate, %/d
ΔPH	Phenolic production, mg/L
Q_{PH}	Maximum phenolic production rate, mg/L/d
$Y_{PH/S}$	Phenolic yield, mg phenolic/g substrate
$-dS/dt$	Substrate consumption rate, g/L/d
$\mu_{m,S}$	Specific sugar consumption rate, 1/d
S	Residual substrate concentration at the time "*t*", g/L
S_m	Maximum substrate concentration, g/L
S_f	Final substrate concentration, g/L
dP/dt	Total acidity rate, %/d
SAS	Statistical Analysis System

References

1. Dutta, H.; Paul, S.K. Kombucha drink: Production, quality, and safety aspects. In *Production and Management of Beverages*; Elsevier: Amsterdam, The Netherlands, 2019; pp. 259–288.
2. Jayabalan, R.; Malbasa, R.V.; Loncar, E.S.; Vitas, J.S.; Sathishkumar, M. A review on kombucha tea: Microbiology, composition, fermentation, beneficial effects, toxicity, and tea fungus. *Compr. Rev. Food Sci. Food Saf.* **2014**, *13*, 538–550. [CrossRef] [PubMed]
3. Teoh, A.L.; Heard, G.; Cox, J. Yeast ecology of Kombucha fermentation. *Int. J. Food Microbiol.* **2004**, *95*, 119–126. [CrossRef] [PubMed]
4. Malbaša, R.; Lončar, E.; Djurić, M. Comparison of the products of Kombucha fermentation on sucrose and molasses. *Food Chem.* **2008**, *106*, 1039–1045. [CrossRef]
5. Şafak, S.; Mercan, N.; Aslim, B.; Beyatli, Y. A study on the production of poly-β-hydroxybutyrate by some eukaryotic microorganisms. *Turk. Electron. J. Biotechnol.* **2002**, *1*, 11–17.
6. Mayser, P.; Fromme, S.; Leitzmann, G.; Gründer, K. The yeast spectrum of the 'tea fungus Kombucha'. *Mycoses* **1995**, *38*, 289–295. [CrossRef]
7. Tsilo, P.H.; Basson, A.K.; Ntombela, Z.G.; Maliehe, T.S.; Pullabhotla, R.V. Isolation and optimization of culture conditions of a bioflocculant-producing fungi from Kombucha tea SCOBY. *Microbiol. Res.* **2021**, *12*, 950–966. [CrossRef]
8. Greenwalt, C.; Steinkraus, K.; Ledford, R. Kombucha, the fermented tea: Microbiology, composition, and claimed health effects. *J. Food Prot.* **2000**, *63*, 976–981. [CrossRef]

9. Kurtzman, C.P.; Robnett, C.J.; Basehoar-Powers, E. *Zygosaccharomyces kombuchaensis*, a new ascosporogenous yeast from 'Kombucha tea'. *FEMS Yeast Res.* **2001**, *1*, 133–138. [CrossRef]
10. Liu, C.-H.; Hsu, W.-H.; Lee, F.-L.; Liao, C.-C. The isolation and identification of microbes from a fermented tea beverage, Haipao, and their interactions during Haipao fermentation. *Food Microbiol.* **1996**, *13*, 407–415. [CrossRef]
11. Chakravorty, S.; Bhattacharya, S.; Chatzinotas, A.; Chakraborty, W.; Bhattacharya, D.; Gachhui, R. Kombucha tea fermentation: Microbial and biochemical dynamics. *Int. J. Food Microbiol.* **2016**, *220*, 63–72. [CrossRef]
12. Morales-de la Peña, M.; Miranda-Mejía, G.A.; Martín-Belloso, O. Recent Trends in Fermented Beverages Processing: The Use of Emerging Technologies. *Beverages* **2023**, *9*, 51. [CrossRef]
13. Nummer, B.A. Kombucha brewing under the Food and Drug Administration Model Food Code: Risk analysis and processing guidance. *J. Environ. Health* **2013**, *76*, 8–11. [PubMed]
14. Villarreal-Soto, S.A.; Beaufort, S.; Bouajila, J.; Souchard, J.P.; Taillandier, P. Understanding kombucha tea fermentation: A review. *J. Food Sci.* **2018**, *83*, 580–588. [CrossRef] [PubMed]
15. Malbasa, R.V.; Loncar, E.S.; Vitas, J.S.; Canadanovic-Brunet, J.M. Influence of starter cultures on the antioxidant activity of kombucha beverage. *Food Chem.* **2011**, *127*, 1727–1731. [CrossRef]
16. Greenwalt, C.J.; Ledford, R.A.; Steinkraus, K.H. Determination and characterization of the antimicrobial activity of the fermented tea kombucha. *LWT-Food Sci. Technol.* **1998**, *31*, 291–296. [CrossRef]
17. Battikh, H.; Bakhrouf, A.; Ammar, E. Antimicrobial effect of kombucha analogues. *LWT-Food Sci. Technol.* **2012**, *47*, 71–77. [CrossRef]
18. Bhattacharya, D.; Bhattacharya, S.; Patra, M.M.; Chakravorty, S.; Sarkar, S.; Chakraborty, W.; Koley, H.; Gachhui, R. Antibacterial activity of polyphenolic fraction of kombucha against enteric bacterial pathogens. *Curr. Microbiol.* **2016**, *73*, 885–896. [CrossRef]
19. Chu, S.C.; Chen, C.S. Effects of origins and fermentation time on the antioxidant activities of kombucha. *Food Chem.* **2006**, *98*, 502–507. [CrossRef]
20. Malbasa, R.; Loncar, E.; Djuric, M.; Dosenovic, I. Effect of sucrose concentration on the products of kombucha fermentation on molasses. *Food Chem.* **2008**, *108*, 926–932. [CrossRef]
21. Zubaidah, E.; Yurista, S.; Rahmadani, N. Characteristic of physical, chemical, and microbiological kombucha from various varieties of apples. In *IOP Conference Series: Earth and Environmental Science*; IOP Publishing Ltd.: Bristol, UK, 2018; Volume 1, p. 012040.
22. Tarhan, K. Use of Different Substrate Resources in the Production of Kombucha Tea. 2017. Available online: https://agris.fao.org/agris-search/search.do?recordID=TR2019000120 (accessed on 18 February 2023).
23. Tamer, C.E.; Temel, Ş.G.; Suna, S.; Karabacak, A.Ö.; Özcan, T.; Ersan, L.Y.; Kaya, B.T.; Çopur, Ö.U. Evaluation of bioaccessibility and functional properties of kombucha beverages fortified with different medicinal plant extracts. *Turk. J. Agric. For.* **2021**, *45*, 13–32.
24. Jayabalan, R.; Marimuthu, S.; Swaminathan, K. Changes in content of organic acids and tea polyphenols during kombucha tea fermentation. *Food Chem.* **2007**, *102*, 392–398. [CrossRef]
25. Battikh, H.; Chaieb, K.; Bakhrouf, A.; Ammar, E. Antibacterial and antifungal activities of black and green kombucha teas. *J. Food Biochem.* **2013**, *37*, 231–236. [CrossRef]
26. Lončar, E.S.; Kanurić, K.G.; Malbaša, R.V.; Đurić, M.S.; Milanović, S.D. Kinetics of saccharose fermentation by Kombucha. *Chem. Ind. Chem. Eng. Q.* **2014**, *20*, 345–352. [CrossRef]
27. Ismaiel, A.A.; Bassyouni, R.H.; Kamel, Z.; Gabr, S.M. Detoxification of patulin by kombucha tea culture. *CyTA-J. Food* **2016**, *14*, 271–279. [CrossRef]
28. Germec, M.; Karhan, M.; Demirci, A.; Turhan, I. Kinetic modeling, sensitivity analysis, and techno-economic feasibility of ethanol fermentation from non-sterile carob extract-based media in *Saccharomyces cerevisiae* biofilm reactor under a repeated-batch fermentation process. *Fuel* **2022**, *324*, 124729. [CrossRef]
29. Alboreadi, M.A.; Al-Najdawi, M.M.; Jarrar, Q.B.; Moshawih, S. Evaluation of hair growth properties of topical kombucha tea extracts. *Adv. Tradit. Med.* **2021**, *22*, 155–161. [CrossRef]
30. Kruk, M.; Trząskowska, M.; Ścibisz, I.; Pokorski, P. Application of the "SCOBY" and Kombucha tea for the production of fermented milk drinks. *Microorganisms* **2021**, *9*, 123. [CrossRef]
31. Cemeroğlu, B. *Gıda Analizleri, Gıda Teknolojisi Derneği Yayınları*; Bizim Grup Basımevi: Ankara, Turkey, 2010.
32. Germec, M.; Karahalil, E.; Yatmaz, E.; Tari, C.; Turhan, I. Effect of process parameters and microparticle addition on polygalacturonase activity and fungal morphology of *Aspergillus sojae*. *Biomass Convers. Biorefin.* **2021**, *12*, 5329–5344. [CrossRef]
33. Miller, G.L. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal. Chem.* **1959**, *31*, 426–428. [CrossRef]
34. Škerget, M.; Kotnik, P.; Hadolin, M.; Hraš, A.R.; Simonič, M.; Knez, Ž. Phenols, proanthocyanidins, flavones and flavonols in some plant materials and their antioxidant activities. *Food Chem.* **2005**, *89*, 191–198. [CrossRef]
35. Mahdinia, E.; Mamouri, S.J.; Puri, V.M.; Demirci, A.; Berenjian, A. Modeling of vitamin K (Menaquinoe-7) fermentation by *Bacillus subtilis natto* in biofilm reactors. *Biocatal. Agric. Biotechnol.* **2019**, *17*, 196–202. [CrossRef]
36. Watawana, M.I.; Jayawardena, N.; Gunawardhana, C.B.; Waisundara, V.Y. Health, wellness, and safety aspects of the consumption of kombucha. *J. Chem.* **2015**, *2015*, 591869. [CrossRef]
37. Muhialdin, B.; Osman, F.; Muhamad, R.; Che Wan Sapawi, C.; Anzian, A.; Voon, W.; Hussin, A. Effects of sugar sources and fermentation time on the properties of tea fungus (kombucha) beverage. *Int. Food Res. J.* **2019**, *26*, 481–487.

38. Kallel, L.; Desseaux, V.; Hamdi, M.; Stocker, P.; Ajandouz, E. Insights into the fermentation biochemistry of kombucha teas and potential impacts of kombucha drinking on starch digestion. *Food Res. Int.* **2012**, *49*, 226–232. [CrossRef]
39. Bhattacharya, S.; Gachhui, R.; Sil, P.C. Effect of kombucha, a fermented black tea in attenuating oxidative stress mediated tissue damage in alloxan induced diabetic rats. *Food Chem. Toxicol.* **2013**, *60*, 328–340. [CrossRef]
40. Emiljanowicz, K.E.; Malinowska-Pańczyk, E. Kombucha from alternative raw materials—The review. *Crit. Rev. Food Sci. Nutr.* **2020**, *60*, 3185–3194. [CrossRef] [PubMed]
41. Fu, C.L.; Yan, F.; Cao, Z.L.; Xie, F.Y.; Lin, J. Antioxidant activities of kombucha prepared from three different substrates and changes in content of probiotics during storage. *Food Sci. Technol.* **2014**, *34*, 123–126. [CrossRef]
42. Carlson, H.A. Check your confidence: Size really does matter. *J. Chem. Inf. Model.* **2013**, *53*, 1837–1841. [CrossRef]
43. Ilgın, M.; Germec, M.; Turhan, I. Statistical and kinetic modeling of *Aspergillus niger* inulinase fermentation from carob extract and its partial concentration. *Ind. Crops Prod.* **2020**, *156*, 112866. [CrossRef]
44. Germec, M.; Turhan, I. Kinetic modeling and sensitivity analysis of inulinase production in large-scale stirred tank bioreactor with sugar beet molasses-based medium. *Biochem. Eng. J.* **2021**, *176*, 108201. [CrossRef]