

1
2
3
4
5
6
7
8
9
10
11
12
13

Enzymatic Hydrolyzed Potato Powder Stabilizes the Properties and Rheological Performance of Yogurt

Ishtiaq Ahmad^a, Zhouyi Xiong^{b**}, Hanguo Xiong^{a*}, Zia-ud-Din,^c Allah Bakhsh Javaid^a, Asad Nawaz^a, Noman Walayat^a

^a College of Food Science and Technology, Huazhong Agricultural University, Wuhan 430070, China

^b Fisheries research institute, Wuhan academy of agricultural sciences, Wuhan 430207, China

^c Department of Agriculture, University of Swabi, Anbar-23561, Khyber Pakhtunkhwa, Pakistan.

*Corresponding author: Hanguo Xiong; xionghanguo@163.com

**Co- Corresponding author: Zhouyi Xiong; xzy646590@163.com

14 **Abstract:**

15 The current study emphasizes on optimizing a suitable ratio of enzymatically hydrolyzed potato
16 powder (EHPP) and whole milk powder (WMP) to produce a quality yogurt by evaluating the
17 physicochemical and rheological properties. The results showed that the addition of EHPP decreased
18 the pH towards acidic conditions which resulted in the high acidity of yoghurt. The proximate
19 composition showed that EHPP increased the ash, protein, water holding capacity (WHC) while fat,
20 syneresis, color parameters and total solid were decreased when compared to control yoghurt (CY)
21 sample. Moreover, texture profile (TPA) analysis showed that the addition of EHPP decreased the
22 hardness and cohesiveness while springiness did not show significant difference. Furthermore,
23 rheological properties revealed that EHPP decreased the storage modulus (G') and loss modulus (G'')
24 when compared with control. In addition to this, sensory analysis revealed that the treatment P4M
25 (1:3) was found as optimum ratio regarding taste, flavor, and aroma. Besides this, scanning electron
26 microscopy (SEM) confirms that the high amount of EHPP resulted in the void holes while CY
27 showed dense gel structure. The prepared yogurt with EHPP provides an excellent flavor, satisfying
28 sweetness and homogeneous texture. These findings suggest the optimum formulation ratio of
29 prepared yogurt was found to be 25% EHPP (P4M) for desirable attributes and consumer acceptance.
30 The prepared yogurt from the EHPP presents the potential industrial applications.

31 **Key words:** Flavored yogurt; potato powder; physiochemical properties; microstructure; rheological
32 properties

33

34 **1. Introduction**

35 Yogurt and other fermented dairy products are popular foods in the world with significant
36 nutritional benefits. Yogurt is milk based product having high nutritional value with concentrated

37 amount of protein, carbohydrates, fat, minerals and vitamins [1, 2]. Frozen yogurt, stirred yogurt,
38 drinkable yogurt and firm yogurt are among the common types of yogurt, whereas, drinkable yogurt is
39 considered as a stirred yogurt with low viscosity and its demand is increasing day by day³. The
40 attractiveness of the yogurt depends upon sensory properties including flavor, aroma, texture, and
41 appearance. These characteristics of yogurt and other fermented dairy foodstuffs depend on type of
42 culture, milk, solid content in milk, fermentation time and temperature[3]. Moreover, textural and
43 rheological properties play a vital role in the sensory attributes of end products. Lactic acid bacteria
44 (LAB) have been extensively studied in the production of fermented milk. LAB occur naturally in
45 various foods and have been utilized for centuries in the fermentation of food with multiple health
46 benefits[4].

47 Recently, numerous efforts have been made to modify the quality of yogurt and to find the
48 protected supplements from natural ingredients [3, 5]. Recent studies reported that numerous fruits and
49 vegetables may be used in various fermented dairy products as functional ingredients and improve the
50 sensory properties in these products³. Beside this, yogurt was prepared with the addition of different
51 fruits, vegetables and fruit seed extracts to enhance the health promoting effects[6]. Thus variety of
52 ingredients are used in yogurt formulations including dietary fiber, antioxidants, vegetable oils, fruit
53 powders, inulin, probiotics, vitamins and minerals which have been successfully incorporated to
54 improve the nutritional value and customer satisfaction[3, 5].

55 Potato (*Solanum tuberosum*) is a tuber crop having starch, vitamins, carbohydrates, high value
56 protein and low fat content, so it plays a very important role in the human health. Potato powder (a
57 processed product of potato) has highest stability during cooling and heating process. It is used as
58 color, thickener and flavor enhancing agent in different value added products. Some other important
59 characteristics including gel forming properties, dough rheology, swelling ability determines the

60 suitability of potato powder in different food formulation [7, 8]. The properties of potato powder can
61 be modified by different enzymatic and chemical treatments. The amylases (glucoamylases β -
62 amylases and α -amylases) are authoritative families of enzymes in the biotechnology, with α -amylases
63 having widest range of industrial applications. A number of studies have been conducted on the
64 kinetics of glycolysis of starch during α -amylases hydrolysis[9]. Similarly, yogurt was prepared using
65 enzymatically hydrolyzed purple sweet potato powder by addition of α -amylase for hydrolysis [10].

66 Keeping in view the limited availability of studies on the effects of potato powder on
67 physicochemical characteristics, the current study emphasizes on optimizing a suitable ratio of
68 enzymatically hydrolyzed potato powder and whole milk powder to produce quality yogurt by
69 evaluating the physicochemical and rheological properties. Moreover, the effects of enzymatically
70 hydrolyzed potato powder supplementation with whole milk powder on pH, titratable acidity, and
71 water holding capacity, syneresis, microstructure, sensory evaluation and color of the yogurt after
72 storage at 4°C for 24 h were carried out during this research. This study provides useful information
73 regarding the use of potato powder in the processing and especially the optimal ratio for the
74 development of desirable characteristics of yogurt.

75 **2. Material and Methods**

76 **2.1. Materials**

77 Potato powder (moisture content 7%, protein 8.6%, fat 1%, carbohydrates 81.7% and sodium
78 35 mg/100g) was obtained from Zhang Jiakou Yunbie Potato Development Co., Ltd., Hubei, and P.R.
79 China. Skimmed milk powder (protein 33.4%, fat 0.8%, carbohydrates 54% and sodium 390
80 mg/100g), fromGMP Dairy Co., Ltd.,Beijing P.R. China. Whole milk powder (protein 23.5%, fat
81 26%, carbohydrates 41.5% and sodium 320 mg/100g) was obtained from Qiqihar Dairy Co., Ltd.,
82 Jingxing, Heilongjiang, P.R.China and sugar was procured from supermarket within Huazhong

83 Agricultural University, Wuhan, Hubei, China. A mixed starter culture of *Lactobacillus* (*Lactobacillus*
84 *bulgaricus*, and *lactobacillus Acidophilus*) from Beijing Chuanxiu Technology, Co., Ltd., Beijing, P.R
85 China was used and α -amylase (4000 U/g) was purchased from Beijing Culture Media Product Field
86 Beijing P.R China. All chemicals used in this study were of analytical grade and used without further
87 purification.

88 2.2. Methods

89 2.2.1. Starter culture preparation

90 Yogurt starter culture, a combination of *Lactobacillus bulgaricus* and *Lactobacillus*
91 *acidophilus* was obtained in freeze-dried form. The skimmed milk powder (SMP) with water (1:6) was
92 mixed and heated at 90 °C for 30 min using water bath. After cooling, inoculating microbial culture
93 using percentage of 0.12% (w/v) was mixed and culture was incubated at 42 °C for 5 to 7 h to prepare
94 the first generation of starter with three repetitions. Continuous cultivation led to the third generation
95 in order to obtain a fully active strain, the third starter generation was stored at 4 °C for 24 h in a
96 refrigerator.

97 2.2.2 Hydrolysis of potato powder

98 Potato powder was added in water according to (1:6) and α -amylase (0.015 g/100ml) was
99 added and mixed at 50 °C with magnetic stirrer (DF-101S type, Wuhan Hengtaida Instrument
100 Equipment Co., Ltd., Wuhan, P.R China). After enzymatic hydrolysis for 10 min, the mixture was
101 placed on boiling-water bath for the inactivation of enzyme for 10 min and cooled down at room
102 temperature for further processing.

103 2.2.3. Standardization of whole milk powder and formulations of yogurt

104 First of all whole milk powder (WMP) was mixed in water according to 1:6 (W/V) respectively.
105 Subsequently subjected EHPPY formulations P1M (1:6), P2M (1:5), P3M (1:4) P4M (1:3), P5M (1:2),
106 P6M (1: 1) (W/W) according EHPP with WMP respectively and CY (without EHPP) were prepared.

107 2.2.4. Preparation of EHPPY

108 Yogurt was prepared with WMP as previously reported methods[1, 2]. According to these
109 procedures, the WMP and white sugar were added in EHPP and uniformly mixed, the mixture was
110 sterilized for 15 min at 90 °C using water bath (HH-Z type, Changzhou Guohua Electric Co., Ltd.,
111 Changzhou, and P.R China). The sterilized mixture was rapidly cooled at room temperature and
112 inoculated by 6% activated strain of yogurt culture. The inoculated mixture was fermented for 5 h at
113 42 °C in an incubator (model; 303A-5, Shanghai Suopu Instrument Co., Ltd., Shanghai, P.R China).
114 After 5 h, the yogurt was placed for 24 h in a refrigerator at 4 °C for further maturation to obtain a
115 finished product. On the second day, the obtained yogurt was mixed for 1 min by using a glass rod and
116 was stored again at 4 °C until further use.

117 2.3. Physico-chemical analysis

118 The physico-chemical analysis of yogurt samples was done using the following methods[12].,
119 moisture (method No. 925-09), protein (method 920-87; N×6.38) fat (method 905.02), acidity (method
120 947.05), ash (method 923-03), total solids (method 990.20), pH during fermentation and after 24 h of
121 storage at 4°C were measured using pH meter (PHS-3 pH Shanghai Electronic Co., Ltd., Shanghai,
122 P.R China) by adopting method (No. 981.81), and acidity by method 947.05. The measurements were
123 done in triplicates and mean values were reported.

124

125 2.3.1. Water Holding Capacity (WHC)

126 WHC of yogurt was measured by previously reported method[12]. For the measurement of
127 WHC of the yogurt, 10 g sample was subjected to centrifugation at 8000 ×g at 4 °C for 15 min. The
128 percentage of WHC was calculated as Equation1.

$$129 \quad \text{WHC}\% = W1/W2 \times 100 \quad (1)$$

130 Where: W1 = Weight of drained gel, W2 = Weight of yogurt.

131 2.3.2. Syneresis

132 The centrifugation method was used to measure the physical stability of the yogurt after 24 h
133 of storage at 4 °C by following a method reported earlier [13]. Sample (10 g) was centrifuged at 7870
134 × g for 10 min at 4 °C; the percentage of syneresis was calculated as Equation 2.

$$135 \quad \% \text{ Syneresis} = (\text{expelled whey(g)})/(\text{Weight of yogurt(g)}) \times 100 \quad (2)$$

136 2.3.3. Color analysis

137 The color parameters of yogurt were measured as lightness (L^*), red/greenness (a^*), and
138 yellow/blueness (b^*), using a CR-300 Minolta colorimeter (Osaka, Japan). Before measuring the color
139 parameters, the colorimeter was calibrated and readings were taken in triplicate to calculate the mean
140 value.

141 2.4. Texture profile analysis

142 Texture profile analysis (TPA) was conducted following the previously described method¹⁵using
143 TA.XT2 plus Texture Analyzer (Stable Micro Systems, London, UK) equipped with a 1 kg load cell
144 was used to determine the texture priorities of yogurt with three replications in a 100 ml beaker of 50 g
145 yogurt that was stored for 24 h at 4 °C. The texture characteristics including hardness, adhesiveness,
146 springiness and cohesiveness were measured by following the pre-test speed 2 mm/s, post-test speed
147 0.5 mm/s, measuring speed 2 mm/s, minimum triggering force 3 g, test distance 8 mm and test time 5 s
148 with probe model P/36R. Samples were kept at 6-8 °C in ice till measurement was done at room

149 temperature. The measurements for all samples were done in triplicates in order to calculate the mean
150 value.

151 2.5. Rheology

152 The rheological characteristics of yogurt after 24 h storage at 4 °C were measured by following a
153 method described in a previous study [15]. A DHR 2 rotational rheometer (TA instruments Crawley,
154 UK) was used for the characterization of rheological characteristics. EHPPY and CY were stored in
155 refrigerator at 4 °C for 24 h and before measurement of the yogurt samples were whipped 10 times
156 clockwise and 10 times anticlockwise with a glass rod. A parallel plate geometry having 40 mm
157 diameter stainless steel plate probe sets distance between bottom and flat 1 mm was used during
158 analysis at 25 °C measuring temperature. The steady shear rheological measurement was done to get
159 with shear rate (0.1-500 s⁻¹) versus shear stress and viscosity curves. Fixed frequency 1Hz, strain
160 range from 0.01-100% scan was used to determine sample constant strain 1%. Power Law Model was
161 used to illustrate the data and demonstrate flow of yogurt samples. Shear stresses (τ) were determined
162 at the correspondent shear rates ($\dot{\gamma}$) obtained at 100 rpm at 25 °C. The experimental data were
163 adapted to Power Law Model $\tau = K\dot{\gamma}^n$, Where τ =yield stress, $\dot{\gamma}$ =shear rate K=consistency index and
164 flow index = n of these mathematical models were applied to illustrate the flow behavior of yogurt
165 samples[16]. Strain amplitude of 1% (within the LVR region) and 0.1-100Hz frequency ranges were
166 used to carry out the dynamic viscoelastic measurement of the yogurt samples. G', G'' and G''/G
167 (tan δ) were obtained as function of angular frequency.

168 2.6. Microstructure

169 The morphology of yogurt samples after 24 h storage at 4 °C by following the procedure of
170 previous study[17]. All the yogurt samples were freeze dried for 32 h under pressure 0.2 millibar fixed
171 in liquid nitrogen using freeze dryer (Beta 2-8 LD Freeze Dryer, CHRIST Co., Ltd.,Germany). These

172 freeze dried yogurt samples were mounted on aluminum stubs and coated by gold layer, observations
173 were carried out through scanning electron microscope (SEM) with different magnification (Model:
174 JSM-6390LV; NTC, Tokyo, Japan) and an accelerated voltage of 15 kV.

175 2.8. Sensory evaluation

176 The prepared yoghurt was subjected to sensory analysis in order to get the consumer's
177 response. Hedonic scale was applied to analyze the sensory properties of yogurts. Yoghurt samples
178 EHPPY and CY were analyzed for appearance, texture, taste, aroma, flavor and overall acceptability
179 after 24 h storage at 4 °C. Ten panelists (5 men and 5 women) having the knowledge about quality of
180 yogurt and food science, belong to the College of Food Science and Technology, Huazhong
181 Agricultural University, Wuhan, Hubei, China were selected to rate the samples based on the 5 points
182 hedonic scale [18]. Approximately 25 ml of yogurt per cup was presented to the panelists. The sensory
183 profiles were conducted on coded in three digits random number cups containing. These sensory score
184 included Excellent=5, Very good=4, Good=3, Average=2 and Poor=1. The panelists were also asked
185 to make some recommendations about the sensory parameter regarding appearance, texture, taste
186 aroma, flavor and overall acceptability of EHPPY and CY.

187 2.10. Statistical analysis

188 Software Statistix version 8.1 (stat Soft USA) was used to analyze the data by one-way
189 analysis of variance (ANOVA). All the measurements were done in three replications and presented as
190 a means and standard deviations. Comparisons between samples means were carried out using least
191 significant test (LSD) test at 5% level of significant.

192 3. Results and discussion

193 3.1. Physico-chemical characterizes

194 The results of pH and acidity of yoghurt after 24 h of storage are given in Table 1. The results showed
195 that the addition of EHPP significantly ($p < 0.05$) decreased the pH towards more acidic conditions
196 which resulted in the increased acidity of the yoghurt compared to the control sample. Our findings
197 regarding the changes in pH values are consistent with previous study [19], who observed lower pH
198 values of yogurt having higher buffering capacity. The pH values may decrease from 4.68 (CY) to
199 3.89 (P6M) by increasing the amount of EHPP because α - amylase convert the starch into sugar and
200 bacteria use this sugar as a food to grow more in number and convert lactose to in lactic acid, hence,
201 cause a reduction in the pH values of yogurt. The supplementation of EHPP significantly ($p < 0.05$)
202 affected the titratable acidity in P6M as compared to CY. After 24h of storage at 4°C, the lowest value
203 of titratable acidity was observed in CY while the highest value was recorded for P6M (Table1). In
204 yogurt, the titratable acidity was influenced by the different kind of proteins. It was reported that
205 titratable acidity affect the deformation of protein and formed the original viscous [20]. The results in
206 Table 1. revealed that the supplementation of EHPP significantly ($p < 0.05$) affected the moisture
207 content of EHPPY than that of CY. The moisture content was increased from 79.67 (CY) to 83.87%
208 (P6M) after 24 h of storage at 4 °C. The increased in moisture content might be due to the high water
209 absorption index of potato powder compared to the milk powder. Similar findings were reported who
210 fortified the yogurt with different minerals and their moisture contents were in the range of 80 to 85%
211 [21].The total solids contents were decreased with the increase in EHPP due to increased water
212 contents in yogurt (Table1). Similar findings regarding the solids were observed by the previous²¹
213 researchers who fortified yogurt with zinc, calcium and iron. The ash content was significantly ($p <$
214 0.05) increased with increased supplementation of EHPP because potato powder has more minerals
215 than milk powder and these minerals increased the ash contents in EHPPY. Similar results of ash
216 contents were found during the preparation of organic yogurt [5]. Moreover, another study also

217 revealed high ash contents in yoghurt with increased mineral contents²¹. The supplementation of
218 EHPP significantly ($p < 0.05$) affected the protein content in all the treatments compared to control
219 treatment (CY). This increase in proteins content with increase of EHPP supplementation in yogurt
220 samples may be due to protein content found in potato powder (8.6%). Similar protein contents were
221 found in pervious study of preparation of organic yogurt with oligofructose grape skin flour and purple
222 grape juice [5]. The addition of EHPP significantly ($p < 0.05$) decreased the fat content of yoghurt
223 compared to the control. This decrease in fat content of EHPP supplemented yogurt might be due to
224 minute amount of fat content of potato powder. Similar fat contents were found in pervious study of
225 preparation of yogurt with grape skin flour and purple grape juice[5].

226 31.1. Water holding capacity

227 The capability of yogurt to hold its own water is called the water holding capacity of yogurt.
228 The results regarding the WHC of yogurt samples are displayed in the Table 1. It is apparent from the
229 results that the WHC significantly ($p < 0.05$) increased from 40.74 to 51.07% in the CY and P6M
230 during the storage of 24 h at 4 °C, respectively. The WHC depends on the type of protein and the
231 amount of starch present in the formulation. The increased WHC of yoghurt with the addition of EHPP
232 might be due to the high starch content of EHPP compared to control. Interaction between water and
233 proteins is most significant in foodstuff systems and flavor, texture of foods affected by this
234 interaction. The value of WHC in this study is similar with the finding of previous study [22], who
235 observed the increased WHC with the addition of peanut (more starchy content)in milk yogurt.

236 3.1.2. Syneresis

237 Syneresis is an important attribute in determining the quality of yogurt. It is defined as the
238 separation of phases in a suspension or mixture and it is natural phenomenon occurs in dairy product
239 such as yogurt. Syneresis reflects the reduction of the gel and increase the separation of whey in

240 yogurt which indicates the weakness of gel [23, 24]. The results showed that the addition of EHPP
241 significantly ($p < 0.05$) decreased the values of syneresis from 59.27 (CY) to 48.90% in P6M,
242 respectively. This indicated that the addition of EHPP improved the gel in the yoghurt which is good
243 attributing regarding the quality of yoghurt. This decreasing effect of EHPP supplementation on
244 syneresis may be due to the high fat yogurt that early reported [25]. Similar results were found in
245 yogurt with the fiber supplementation that increased the WHC of yoghurt and decreased the syneresis
246 [20]. The P4M was found to be an optimized formulation with respect to the results of syneresis.

247 3. 1.3. Colors analysis

248 Color plays a significant role in determination of acceptability of numerous food products. The results
249 regarding the effect of EHPP supplementation on the color of control and supplemented yogurt
250 samples are displayed in Table 2. The results revealed that the addition of EHPP powder significantly
251 ($p < 0.05$) changed the color of EHHPY samples as compared to CY. The L^* value decreased from
252 initial value 98.94 in CY sample to 93.22 in P6M treatment. On the other hand, a^* and b^* values were
253 increased in CY compared to P6M. This change in color may be attributed by potato powder and its α -
254 amylase hydrolysis. Our findings are in line with the previously reported study of fortification of
255 yogurt with nano and micro sized calcium, iron and zinc check the effect on the physicochemical and
256 rheological properties [20]. On the basis of color parameters, the addition of high amount of EHPP
257 resulted in decreased lightness while increased in redness that is not acceptable for consumer point of
258 view. Thus, P4M was found to be in the range of color requirement for desirable yoghurt.

259 3.1.4. Texture profile analysis of yogurt

260 The TPA of different treatment of yogurt was evaluated after storage 24 h at 4 °C. The texture
261 characterizations of yogurt, including hardness, adhesiveness cohesiveness, and springiness were
262 obviously improved by EHPP (Table 2). Hardness is an important parameter for texture of yogurt;

263 higher values of hardness were observed in CY, because of higher rearrangement of protein found in
264 CY [26]. The addition of EHPP decreased the hardness significantly ($p < 0.05$) from 92.30 in CY to
265 24.76 in P6M. The adhesiveness increased with increasing the EHPP, so that a tendency of high fat
266 with EHPP becomes connected with the surface of the texture analyzer [27]. The addition of EHPP
267 caused a remarkable reduction in fat content of yogurt, however it cannot be comparable to low fat
268 yogurts, hence its behavior of texture properties is similar to high fat yogurt. The springiness of all the
269 yogurt samples was increased with the increase of EHPP (Table 2). The softness of the EHPPY may be
270 due to increase in the water due to the high absorption capacity of EHPP compared with milk powder.
271 Moreover, the increase in springiness may attribute to higher water in gel of yogurt that means sample
272 has ability of regain its own initial position [28]. It is apparent from the results that the addition of
273 EHPP contributed to the significant ($p < 0.05$) increase of cohesiveness in EHPP treated samples.
274 However, ²⁸ recorded higher values of cohesiveness in low fat yogurt samples. Similar results of TPA
275 were found in optimized yogurt by micro sized calcium, iron and zinc [20]. On the basis of texture, the
276 P4M considered as a good quality yogurt after comparison with previous published research.

277 3.2. Rheological properties of yogurt

278 Rheological characteristics are for huge attention, when study the properties of yogurt, because these
279 characteristics are directly connected to use of yogurt. Fig.1 (a-e) represents the rheological properties
280 of the yogurt with and without addition of EHPP.

281

282 3.2.1. Apparent Viscosity and shear stress

283 The results showed that the decreasing trend was found in apparent viscosity in the yogurt by
284 increasing the addition of EHPP. The CY sample showed the high viscosity values among yogurt all
285 samples. The results exposed that the addition of EHPP demonstrated decrease in viscosity values of

286 stirred yogurt. This might indicate that EHPPY positively changed the texture of the yogurt. Viscosity
287 influences the sensory characteristics and it is considered as vital quality parameter of yogurt.
288 Rheometric viscosity has a strong positive correlation with thickness [28]. Rheological study indicates
289 that the apparent viscosities of the entire yogurt sample decreased with increased shear rate. These
290 phenomena reflected the shear thinning behavior of EHPPY and CY samples. The viscosities of yogurt
291 of different EHPP are presented in all seven yogurt samples exhibits shear thinning behavior for the
292 shear rates from 0.1 to 500 s⁻¹. However, the viscosity profiles for these samples were almost
293 overlapped at most shear rates. The lower apparent viscosity of all EHPPY samples might result in
294 high moisture contents of yogurt. However, by increasing the shear rate for both CY and EHPPY the
295 viscosity was decreased (Fig.2a), a shear thinning behavior was found in all EHPP supplemented
296 yogurt by following the previous study [29]. Additionally, the shear stress for all the EHPPY was lower
297 than CY at the same shear rate in (Fig.1b). This phenomenon is indicative of that there is strong
298 interaction between molecules of starch found in potato powder and casein of milk. Shear stress
299 change EHPPY slowly, therefore, shear stress increases in EHPPY and reaches a certain value and the
300 curve angle first increases and then decreases, which is because of the external force of molecules
301 would gradually be disintegrated and the stress tends to be fixed. Power law model was well fitted
302 when the curve of shear stress with shear rate were analyzed ($R^2 > 0.99$) in all yogurt samples (Table 2).
303 The flow behavior index (n) $1 < n$ as all the samples exhibited pseudoplastic behavior. Flow behavior
304 index (n) was decreased from 0.562 in (CY) to 0.259 in (P6M) and consistency index was increased
305 from (CY) 4.67 to (P6M) 35.47(Pa.s), with the addition of EHPP which proved that the yogurt with
306 EHPP was more pseudo-plastic than that of CY; the entire yogurt sample exhibited the pseudo-plastic
307 behaviors. The steady shear viscosity coefficient of CY was higher than the yogurt samples of EHPP
308 at the relatively low shear rate. The addition of EHPP obviously affected the flow behaviors of

309 yogurts. The model with best fit was the Power-Law model [30]. Therefore, the yogurt with EHPP
310 exhibited improved shear thinning behavior which would compose an adequate contact between
311 yogurt and consumers. All the yogurt samples show exhibited extremely shear thinning behavior with
312 flow behavior index (n) as low as indicating the addition of EHPP enhanced the pseudo-plasticity of
313 yogurt due to the reduced entanglement between starch granules [31]. The experimental yogurts were
314 highly thixo-tropic and behaved as pseudo-plastic materials. We found significant differences ($p <$
315 0.05) in the rheological properties between yogurts made from milk powder and supplemented with
316 different concentrations of EHHP. Similar results were reported by the fortification of yogurt with
317 minerals [20]. Significantly decreased the viscosity of P4M considers as good quality yogurt as
318 compared to the other treatments of EHPPY by following the previous above mention study.

319 3.2.2. Frequency sweep

320 All the yogurt samples showed an escalating trend in G' and G'' values at lower frequencies
321 (Fig 1c; 1d). EHPP affected the viscoelastic properties of yogurts that indicate that EHPP had no
322 increasing effect on firmness of EHPPY (Fig 1c; 1d). The increase in G' values (Fig.1c) of CY are
323 negligible at higher frequency, suggesting a weak gel structure for yogurt and these results are
324 supported by initial findings of previous study[31].In contrast, more distinct increase in the G' values
325 (Fig.1 c) was noticed for the yogurt with EHPP representing that the structure properties of yogurt
326 have strengthened through the interaction among milk casein and molecules of starch of EHPP. The
327 G'' value (Fig.1d) for the yogurt with EHPP increased more notably with increase in angular frequency
328 as compared with the CY. The data is reinforced by the dynamic mechanical loss tangent ($\tan \delta = G''/G'$)
329 of yogurt that displayed escalating trend as shown in the (Fig.1e). The $\tan \delta$ gives the exact measure of
330 the behavior of material as liquid like to solid like and it is dimensionless parameter. The δ value of
331 controlled yogurt was lesser than EHPP supplemented samples. This result indicated a conversion

332 from liquid to solid behavior and a high value of δ for yogurt with EHPP which may be resulted due to
333 improved interaction among milk casein and starch molecules found in potato powder in the
334 continuous phase so as to appear a strong physical system [31]. However, at the relatively elevated
335 frequency the $\tan \delta$ value for EHPPY started to decline notably which recommended that the elastic
336 nature was succeeded above the viscous nature with EHPP addition.

337 **3.3. Microstructure**

338 Microstructure affects the texture properties, taste, flavor of food and understanding of
339 microstructure on food quality improvement guide. Microstructures of EHPPY samples and CY are
340 elucidated in (Fig.2).CY had dense gel structure of casein, large pore and uniform size of casein
341 particles. The microstructure of the yogurt without addition of EHPP was much branched and
342 displayed cross-linking in yogurt samples.SEM micrographs of the yogurts showed that the addition of
343 EHPP led to differences in gel network. The addition of EHPP resulted in void holes and weak gel
344 formation which resulted in low hardness of gel. With the supplementation of EHPP, the gel becomes
345 less dense and the particle becomes more uniform and small with fluffier and coarser microstructure.
346 Shorter fermentation time may be moderately explained as the lesser density of yogurt network. A
347 compact and more homogenous microstructure with minor pores and particles characterized yogurt
348 was prepared with EHPP. The same findings were found in yogurt prepared with goat milk by addition
349 of whey protein[32]. Similarly, The consistent findings were observed in the yogurt made from
350 modified whey protein formulations [17]. This could direct to the coarser network of the yogurts
351 containing EHPP in the current work, because it has been confirmed that the complexes comprised by
352 denatured whey protein and casein play a very important role in gel formation network³⁴Micrograph of
353 P4M showed different microstructure of yogurt as compared to the CY. This change in microstructure
354 of P4M improved the sensory characteristic and makes it a good quality yogurt.

355 3.4. Sensory evaluation of yogurt

356 Sensory evaluation of control and supplemented yogurt samples was conducted using the Hedonic
357 test (5-point scale) by 10 panelists (Fig.3). All the yogurt samples including CY and EPHH
358 supplemented samples were evaluated for taste, appearance, texture, aroma and overall acceptability.
359 The panelists rated higher scores to the appearance of EHPPY although there was significant ($p < 0.05$)
360 difference in appearance existed among all yogurts. P4M were given higher scores for appearance by
361 the panelists. The only P4M showed a significant ($p < 0.05$) difference in terms of appearance.
362 According to sensory properties, P4M was found more acceptable by the panelists among all other
363 treatments. For overall acceptability, appearance, taste, aroma, and texture the treatment P4M got
364 highest scores among CY and all other EHPP supplemented yogurt samples followed by CY. The
365 sensory scores of EHPP supplemented yogurt was related with a statistically significant ($p < 0.05$)
366 effect on sensory properties including color, flavor, and overall acceptability. The addition of EHPP to
367 yogurt improved the flavor. Appearance score of CY and EHPPY were different significantly ($p <$
368 0.05). Previous study also showed the improvement in flavor and appearance of yogurt fortified with
369 fruits and vegetables ³⁷.

370 4. Conclusion

371 In the current study, EHPPY was prepared with different ratios of EHPP, where the treatment
372 (P4M) 25% of EHPP showed improved physicochemical, texture, microstructure, rheological, color,
373 antioxidant activity with better sensory properties of stirred yogurt. Therefore, our study supports the
374 hypothesis that potato powder may be used as potential ingredient in yogurt due to its differential
375 characteristics. But the high amount of EHPP can negatively affect the desirable characteristics
376 especially WHC, texture, color and rheological properties. Thus, it is recommended that the addition
377 of EHPP increases the yogurt acceptability which could be scaled up further, providing a basis for

378 future studies on fermented dairy products. The storage study of newly prepared yoghurt can be a
379 limitation of this study.

380 **Acknowledgements**

381 This work was financially supported by the Nature Science Foundation of Hubei Province
382 (2018CFB269), innovation project of Wuhan Academy of Agricultural Sciences (CX201811) National
383 Key R&D Program of China (2018YFD0400702) and the National Natural Science Foundation of
384 China (31471699).

385 **References**

- 386 1. Mckinley MC (2005) The nutrition and health benefits of yoghurt. International Journal of
387 Dairy Technology 58:1–12. <https://doi.org/10.1111/j.1471-0307.2005.00180.x>
- 388 2. Tamime AY, Robinson RK (Richard K, Tamime AY (2007) Tamime and Robinson's yoghurt :
389 science and technology. CRC
- 390 3. Hashemi Gahrue H, Eskandari MH, Mesbahi G, Hanifpour MA (2015) Scientific and technical
391 aspects of yogurt fortification: A review. Food Science and Human Wellness 4:1–8.
392 <https://doi.org/10.1016/j.fshw.2015.03.002>
- 393 4. Saikali J, Picard C, Freitas M, Holt P (2004) Fermented Milks, Probiotic Cultures, and Colon
394 Cancer. Nutrition and Cancer 49:14–24. https://doi.org/10.1207/s15327914nc4901_3
- 395 5. Karnopp AR, Oliveira KG, de Andrade EF, et al (2017) Optimization of an organic yogurt
396 based on sensorial, nutritional, and functional perspectives. Food Chemistry 233:401–411.
397 <https://doi.org/10.1016/J.FOODCHEM.2017.04.112>
- 398 6. Oliveira A, Alexandre EMC, Coelho M, et al (2015) Incorporation of strawberries preparation
399 in yoghurt: Impact on phytochemicals and milk proteins. Food Chemistry 171:370–378.
400 <https://doi.org/10.1016/j.foodchem.2014.08.107>

- 401 7. Friedman M (1997) Chemistry, biochemistry, and dietary role of potato polyphenols. A review.
402 Journal of Agricultural and Food Chemistry 45:1523–1540. <https://doi.org/10.1021/jf960900s>
- 403 8. Van Hal M (2000) Quality of sweetpotato flour during processing and storage. Food Reviews
404 International 16:1–37. <https://doi.org/10.1081/FRI-100100280>
- 405 9. BravoRodriguez V, JuradoAlameda E, MartinezGallegos JF, et al (2006) Enzymatic Hydrolysis
406 of Soluble Starch with an α -Amylase from *Bacillus licheniformis*. Biotechnology Progress
407 22:718–722. <https://doi.org/10.1021/bp060057a>
- 408 10. Kim DC, Won SI, In M-J (2015) Substitution Effect of Enzymatically Hydrolyzed Purple Sweet
409 Potato Powder on Skim Milk in Yogurt Preparation. Journal of Applied Biological Chemistry
410 58:311–316. <https://doi.org/10.3839/jabc.2015.049>
- 411 11. AOAC (2006) Official Methods of Analysis (18th ed.)
- 412 12. Harte F, Luedecke L, Swanson B, Barbosa-Cánovas GV (2003) Low-Fat Set Yogurt Made from
413 Milk Subjected to Combinations of High Hydrostatic Pressure and Thermal Processing. Journal
414 of Dairy Science 86:1074–1082. [https://doi.org/10.3168/jds.S0022-0302\(03\)73690-X](https://doi.org/10.3168/jds.S0022-0302(03)73690-X)
- 415 13. Amatayakul T, Halmos AL, Sherkat F, Shah NP (2006) Physical characteristics of yoghurts
416 made using exopolysaccharide-producing starter cultures and varying casein to whey protein
417 ratios. International Dairy Journal 16:40–51. <https://doi.org/10.1016/J.IDAIRYJ.2005.01.004>
- 418 14. Varghese K. S, Mishra HN (2008) Modelling of acidification kinetics and textural properties in
419 dahi (Indian yogurt) made from buffalo milk using response surface methodology. International
420 Journal of Dairy Technology 61:284–289. <https://doi.org/10.1111/j.1471-0307.2008.00411.x>
- 421 15. Aprianita A, Purwandari U, Watson B, Vasiljevic T (2009) Physico-chemical properties of
422 flours and starches from selected commercial tubers available in Australia. International Food
423 Research Journal 16:507–520

- 424 16. Ramírez-Sucre MO, Vélez-Ruiz JF (2013) Physicochemical, rheological and stability
425 characterization of a caramel flavored yogurt. *LWT - Food Science and Technology* 51:233–
426 241. <https://doi.org/10.1016/J.LWT.2012.09.014>
- 427 17. Matumoto-Pintro PT, Rabiey L, Robitaille G, Britten M (2011) Use of modified whey protein in
428 yoghurt formulations. *International Dairy Journal* 21:21–26.
429 <https://doi.org/10.1016/J.IDAIRYJ.2010.07.003>
- 430 18. Stone H, Sidel JL (1992) Sensory evaluation practices. Academic Press
- 431 19. Zare F, Boye JI, Orsat V, et al (2011) Microbial, physical and sensory properties of yogurt
432 supplemented with lentil flour. *Food Research International* 44:2482–2488.
433 <https://doi.org/10.1016/J.FOODRES.2011.01.002>
- 434 20. Santillán-Urquiza E, Méndez-Rojas MÁ, Vélez-Ruiz JF (2017) Fortification of yogurt with
435 nano and micro sized calcium, iron and zinc, effect on the physicochemical and rheological
436 properties. *LWT - Food Science and Technology* 80:462–469.
437 <https://doi.org/10.1016/J.LWT.2017.03.025>
- 438 21. Sanz T, Salvador A, Jiménez A, Fiszman SM (2008) Yogurt enrichment with functional
439 asparagus fibre. Effect of fibre extraction method on rheological properties, colour, and sensory
440 acceptance. *European Food Research and Technology* 227:1515–1521.
441 <https://doi.org/10.1007/s00217-008-0874-2>
- 442 22. Isanga J, Zhang G (2009) Production and evaluation of some physicochemical parameters of
443 peanut milk yoghurt. *LWT - Food Science and Technology* 42:1132–1138.
444 <https://doi.org/10.1016/j.lwt.2009.01.014>
- 445 23. E. Ocak and Ş. Köse (2010) The effects of fortifying milk with Cu, Fe and Zn minerals on the
446 production and texture of yoghurt. *International Society for Food, Agriculture* 8:122–125

- 447 24. Celik S, Bakırcı I, Şat IG (2006) Physicochemical and Organoleptic Properties of Yogurt with
448 Cornelian Cherry Paste. *International Journal of Food Properties* 9:401–408.
449 <https://doi.org/10.1080/10942910600596258>
- 450 25. M. C. Staff (1998) *Cultured Milk and Fresh Cheeses*. In: *The Technology of Dairy Products*,
451 2nd Ed., R. Blackie Academic and Professional, London
- 452 26. Prasanna PHP, Grandison AS, Charalampopoulos D (2013) Microbiological, chemical and
453 rheological properties of low fat set yoghurt produced with exopolysaccharide (EPS) producing
454 *Bifidobacterium* strains. *Food Research International* 51:.
455 <https://doi.org/10.1016/j.foodres.2012.11.016>
- 456 27. Magenis RB, Prudencio ES, Amboni RDMC, et al (2006) Compositional and physical
457 properties of yogurts manufactured from milk and whey cheese concentrated by ultrafiltration.
458 *International Journal of Food Science and Technology* 41:560–568.
459 <https://doi.org/10.1111/j.1365-2621.2005.01100.x>
- 460 28. Pelaes Vital AC, Goto PA, Hanai LN, et al (2015) Microbiological, functional and rheological
461 properties of low fat yogurt supplemented with *Pleurotus ostreatus* aqueous extract. *LWT -*
462 *Food Science and Technology* 64:1028–1035. <https://doi.org/10.1016/J.LWT.2015.07.003>
- 463 29. Mohameed HA, Abu-Jdayil B, Al-Shawabkeh A (2004) Effect of solids concentration on the
464 rheology of labneh (concentrated yogurt) produced from sheep milk. *Journal of Food*
465 *Engineering* 61:347–352. [https://doi.org/10.1016/S0260-8774\(03\)00139-0](https://doi.org/10.1016/S0260-8774(03)00139-0)
- 466 30. Damin MR, Alcântara MR, Nunes AP, Oliveira MN (2009) Effects of milk supplementation
467 with skim milk powder, whey protein concentrate and sodium caseinate on acidification
468 kinetics, rheological properties and structure of nonfat stirred yogurt. *LWT - Food Science and*
469 *Technology* 42:1744–1750. <https://doi.org/10.1016/J.LWT.2009.03.019>

- 470 31. Nguyen PTM, Kravchuk O, Bhandari B, Prakash S (2017) Effect of different hydrocolloids on
471 texture, rheology, tribology and sensory perception of texture and mouthfeel of low-fat pot-set
472 yoghurt. *Food Hydrocolloids* 72:90–104. <https://doi.org/10.1016/J.FOODHYD.2017.05.035>
- 473 32. And JL, Guo M (2006) Effects of Polymerized Whey Proteins on Consistency and Water-
474 holding Properties of Goat's Milk Yogurt. *Journal of Food Science* 71:C34–C38.
475 <https://doi.org/10.1111/j.1365-2621.2006.tb12385.x>
- 476 33. Asuman Cinbas and Fehmi Yazici (2008) Effect of the Addition of Blueberries on Selected
477 Physicochemical and Sensory Properties of Yoghurts. *Food Technology and Biotechnology*
478 46:434–441
- 479