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# SHELF-LIFE ASSESSMENT OF FORTIFIED WHOLEMEAL PASTA WITH Amaranthus tricolor AND Polygonum minus

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# Abstract

#### Keywords:

Bio preservation; shelf-life; antimicrobial; flavonoids; pasta Fresh pasta is known to contain a good nutritional profile and moisture content of greater than 24.0%. However, under certain conditions, pasta can be an ideal vessel for microbial growth and requires the addition of preservatives for a longer shelf-life. In plants, flavonoids are synthesised in response to microbial threats. With the concept of bio-preservatives from these natural plant sources, this study aims to observe fresh pasta shelf-life by fortifying wholemeal pasta with dried powder of Amaranthus tricolor and Polygonum minus individually, stored in refrigeration (4°C) and in ambient temperature (24°C). From preliminary evaluation, P. minus aqueous extract recorded higher flavonoid and flavonol content at 21.6 mg/g QE and 34.5 mg/g QE, compared to A. tricolor aqueous extract at 17.0 mg/g QE and 23.5 mg/g QE, respectively. The fortified wholemeal pasta had shown to significantly affect optimum cooking time and swelling index by almost two-fold compared with control pasta. Interestingly, after observing and analysing the total plate count during a period of 120 hrs, wholemeal pasta with dried powder of Amaranthus tricolor stored at 4°C had shown to have a better shelflife, as it significantly contained within the borderline pasta spoilage at 5.46 log cfu/ml. Fortification of wholemeal pasta with these plant powders also had similar overall acceptance with no significant differences to the control pasta. This shows the potential that the addition of plant powder could serve as potent bio-preservative and antimicrobial agents and consumer acceptance.

#### **INTRODUCTION**

The concept of bio-preservation was developed to minimise the usage of chemical preservatives. Food products, particularly of high-water content can be easily subjected to bacterial and fungal contamination, leading to the deterioration of food flavour, colour, odour, textural and sensory properties of the food. Various studies had explored different methods of shelf life prolongation, including the common approach with chemical preservatives such as acetic acids and sodium dehydroacetate (1). Nonetheless, the usage of chemical preservatives had increasingly attracted negative attention due to possible detrimental effects on consumers as well as the environment (2). Therefore, the search for natural bio-preservatives had been greatly promoted with the exploration of antimicrobial compounds derived from microorganism, animal and plant origin (3). Plants are excellent sources of bio-active compounds with great antimicrobial potential. Flavonoids are one of the phytochemicals that had been extensively studied for its antimicrobial properties (4). These bio-compounds present in plants as the most abundant secondary plant metabolites, providing protective functions against many foreign stressors including herbivores and microbial threats (5). The selected plants for this study are *Polygonum minus* (*P. minus*) and *Amaranthus tricolor* (*A. tricolor*). Both species are indigenous among the Southeast Asian region. It contains excellent amount of minerals, antioxidant leaf pigments, carotenoids, vitamins, phenolics, as well as, flavonoids (6,7), with demonstrating moderate levels of antibacterial and anticancer activities (8).

Pasta had gain international popularity due to its versatility, nutritional profile and its consumption acceptability by people from all social and age groups. Storage condition and water content are some of the main factors affecting pasta's shelf life. Notably, fresh pasta experience much shorter shelf life as it is susceptible to rapid microbial proliferation, due to its moisture content of more than 24.0% (9). Therefore, careful storage consideration and the addition of preservatives is necessary to prolong its shelf life. Incorporation of plants in food matrix vessels had deemed effective in prolonging the shelf life. This was observed in fortified broccoli pasta (9) and in fresh fortified barley noodles (10). However, the main challenge is the lack of consumers' acceptance which stems from undesirable sensory quality such as bitter and acrid taste from incorporating plant derivatives in food products (11). Thus, with the above considerations, the aim of this study was to evaluate the efficacy of two common vegetables in Brunei; P. minus and A. tricolor to improve pasta shelf life, while maintaining pasta formulation with the acceptable sensory qualities for consumption. Plant powders were prepared and subsequently fortified into fresh wholemeal pasta and the effects were verified through microbial and sensory evaluation.

## MATERIALS AND METHODS

#### Materials

Plate count agar and sodium chloride were obtained from Merck Millipore. *A. tricolor* (NT-305) and *P. minus* (NT-2601) were purchased from the local market and rinsed thoroughly under running water. The leaves were set to oven-dry at 40 °C for 5 hours. The dried leaves were grounded to powder using a domestic blender (Prestige Deluxe 750 Watts Power, India).

#### Methods

# **Preparation of Plant Extracts**

Aqueous plant extracts for each individual plants were prepared by soaking and stirring plant powder 1:10 to distilled water using a magnetic stirrer at medium speed for 24 hours. It was then subjected to vacuum filtration and rotary evaporator. The crude extracts were then stored at -5 °C until further use.

# **Total Flavonoid and Flavonol Content**

The determination of both flavonoid and flavonol content were evaluated following the proposed method by Iqbal et al. (12) using aluminum chloride colorimetric method, where

standard calibration curve of quercetin was prepared with the range of 0-200 ug/ml. For total flavonoid content, the standard (0.5 ml) and extract (0.5 ml) were added in different test tubes, followed by 10% aluminium chloride (0.1 ml), 1 M potassium acetate (0.1 ml), 80% methanol (1.5 ml) and distilled water (2.8 ml). A blank was similarly prepared with the exception of extract or standard and aluminum chloride and were replaced by distilled water. The mixtures were incubated at room temperature for 30 minutes and the absorbance was read at 415 nm. The concentration of flavonoid was expressed as mg quercetin equivalent (QE) per gram of extract. In the determination of total flavonol content, extracts (1.0 ml) and standard (1.0 ml) were placed in different test tubes, followed by 2.0% aluminium chloride (1.0 ml) and 5.0% sodium acetate (3.0 ml). The mixtures were mixed vigorously to get a clear solution and left to stand for 30 min at room temperature. The absorbance was read at 440 nm and the results were expressed as mg quercetin equivalent (QE) per gram extract.

# **Pasta Preparation**

Pasta preparation followed the methods of Li et al. (13), with few modifications. Four pasta formulations were prepared for this study (Table. 1). The basic pasta components including wholemeal flour, plant powder and water (30.0 ml) were mixed and combined to facilitate even water distribution. The dough was set to rest for 20 minutes before subjecting into the domestic pasta maker. The pasta dough was further rolled to flatten and cut into pasta with 1.0 mm thickness and 30.0 mm length. These raw pastas were stored in sealed airtight containers at 4°C until further analysis.

**Table 1.** Pasta formulations of control pasta (CO), wholemeal pasta (WM), wholemeal pasta fortified with *P. minus* (PWM) and wholemeal pasta fortified with *A. tricolor* (AWM)

Pasta samples	Plain flour (g)	Wholemeal flour (g)	Plant powder (g)	
СО	100	0	0	
WM	50	50	0	
PWM	50	50	2	
AWM	50	50	2	

#### **Pasta Cooking Quality Evaluation**

Each pasta sample was analysed for the optimum cooking time (OCT) and cooking loss (CL) according to approved method No 66-50 (AACC, 2000). Pasta samples were cooked in boiling distilled water (1:10). The optimal cooking time corresponds to the time required to gelatinise the starch and for the disappearance of the white core (non-gelatinised starch), after compressing between two pieces of glass at intervals of 30s. The cooking water was oven-dried at 105°C until constant weight is achieved. Cooking loss was determined by the amount of solid lost into the cooking water

and the percentage cooking loss was expressed through the formula:

Cooking loss (%) = 
$$\frac{\text{Weight of dried residue (g)}}{\text{Raw pasta before cooking (g)}} \times 100$$

Swelling index of cooked pasta was evaluated by drying pasta at 105°C until constant weight was achieved. The swelling index was expressed as follows:

Swelling index (g) = 
$$\frac{\text{Cooked pasta (g)} - \text{Dried pasta (g)}}{\text{Dried pasta (g)}}$$

# **Total Plate Count Analysis**

The method for total plate count (TPC) was adapted from Li et al. (14) with slight modifications. All apparatus including test tubes, pipette tips, beakers, saline solution and agar were autoclaved at 121°C for 1.5 hours. Cooked pasta samples of 10.0 g were homogenized with 90.0 ml of sterile saline solution (0.85% NaCl). Ten-fold dilution of pasta samples were prepared by pipetting 1.0 ml of homogenised sample into sterile tube with 9.0 ml of sterile saline solution. Consequently, serial dilutions of 10-1 to 10-6 were also prepared. The pour plate method was used to execute this experiment. Exactly 1.0 ml of each dilution were pipette into sterile petri dish and warm plate count agar (PCA) was poured over it. The petri dish was gently swirled to allow mixing. Petri dishes were incubated at 37.0°C for 48 hours to detect any growth of aerobic mesophilic bacteria (AMB). This can be done by observing whether colonies are formed on the surface of PCA plates for each plate. Only 25 to 250 colonies were taken into consideration in calculating the colony forming unit (cfu/ml) of bacterial colonies grown on PCA plates. Plates with less than 25 colonies were recorded as too few to count (TFTC) while too numerous to count (TNTC) was recorded for plates with more than 250 colonies. The colony forming unit was determined using the following formula:

Colony forming unit 
$$\left(\frac{ctu}{ml}\right)$$
  
=  $\frac{\text{Number of colonies counted on plate } \times \text{Dilution factor}}{\text{Volume of culture plate (ml)}}$ 

#### **Pasta pH Evaluation**

Pasta samples (10.0 g) was homogenised in 90.0 ml distilled water using a domestic blender. The pH was measured using pH meter (F-22 E HORIBA, Japan) in a period of 5 days, following the total plate count analysis.

#### **Pasta Moisture Analysis**

Cooked pasta samples were subjected to the moisture analyser (AND MX-50, Japan). Briefly, 5.0 g of sample were weighed before placing in the moisture analyser. The percentage moisture was determined using the moisture analyser with the formula:

Moisture (%) = 
$$\frac{W0 - W1}{W0} \times 100$$

where, W0 is the initial weight of the sample in grams and W1 is the dry weight of the sample. The moisture content of pasta samples was monitored for 5 days and recorded.

#### **Sensory Evaluation**

Pasta samples were cooked in boiling water without the addition of salt. A panel of healthy adults evaluated the pasta attributes based on its appearance, chewiness, colour, firmness, stickiness, taste and overall acceptability. The sensory evaluation was employed based on hedonic scale of food acceptability of nine points, where 1 = dislike extremely, 9 = like extremely and the middle point at 5 = neither like nor dislike.

#### **Statistical Analysis**

All statistical analyses were conducted in R 4.0.2 (R Development Core Team, 2020). The variables were analysed using one-way analysis of variance (ANOVA), to compare differences between each pasta samples. This was followed by Tukey's honest significant difference (Tukey's HSD) test to make a pair-wise multiple comparison of means among the samples. Pearson's correlation was used to determine the correlation of data between the total plate count, pH and moisture. Data obtained were reported as mean  $\pm$  standard deviation.

#### **RESULTS AND DISCUSSION**

# Total Flavonoid and Flavonol Content of *A. tricolor* and *P. minus* Aqueous Extracts

The total flavonoid and flavonol were evaluated as preliminary screening prior to the total plate count analysis. Flavonoids are secondary metabolites with polyphenolic structure produced by plants in response to various biological activities. It is known to synthesise in sites that are attributable to the plants' colour and aroma, which consequently aid in pollination and germination process (15). While, flavonol is the subclass of flavonoids, consisting of quercetin, myricetin, and kaempferol, which are all commonly found in fruits, vegetables and medicinal plants. Based on the data obtained in Table 2, *P. minus* had significantly higher flavonoid and flavonol content of 21.6 mg/g QE and 34.5 mg/g QE, respectively, in contrast to *A. tricolor* with flavonoid content of 17.0 mg/g QE and flavonol

at 23.5 mg/g (p < 0.05). This was similarly observed in Sumazian et al. (16), whereby boiled aqueous *P. minus* extract had prominently regarded with the highest flavonoid content at 6.28 mg/g dry weight. During pathogenic invasion, plants trigger its innate immunity defense which includes biosynthesis of flavonoids (17). This aids in deterring pathogenic attacks which includes microbial threats. This preliminary flavonoid and flavonol screening suggest *P. minus* would have the superior potential as bio preservative in prolonging shelf life of pasta.

 Table 2. Total flavonoid and flavonol content of A. tricolor and P. minus

	A. tricolor	P. minus
Flavonoid (mg/g QE)	$17.0\pm0.12^{\rm a}$	$21.6\pm0.30^{\rm a}$
Flavonol (mg/g QE)	$23.5\pm0.52^{b}$	$34.5\pm0.90^b$

Values with the same superscript letters across each row signifies significant differences (p < 0.05)

# **Cooking Quality Analysis of Pasta**

Fortification of food with foreign compositions often affect cooking parameters of the product. The specificity of raw materials included in a formulation could influence the texture and overall quality (18). Hence, it is of great importance to monitor and assess the cooking quality. Fortification of pasta with wholemeal flour and plant powders had significantly increased the cooking time to almost 2-fold (Figure 1A). Control pasta (CO) recorded with minimum optimum cooking time (OCT) of 3.2 minutes, while wholemeal pasta (WM), wholemeal pasta fortified with *P. minus* (PWM) and wholemeal pasta fortified with *A. tricolor* (AWM) required 5.0, 5.0 and 5.2 minutes for OCT, respectively. The range of cooking time was also in

reasonable agreement with the findings of Attanzio et al. (19) in their fortified Opuntia ficus-indica pasta. In the present study, the addition of wholemeal flour had strengthened the gluten-matrix complex which reinforce the overall structure of pasta, providing elasticity and retain moisture (20), resulting in longer cooking time required for WM, AWM and PWM. As part of the cooking quality parameter, the capacity of pasta to absorb water during cooking was also recorded. This swelling index was determined by the index value of weight increased after cooking. AWM had the highest weight increase at 1.30 g (Figure 1B), as with it the highest OCT (Figure 1A). It was reported that the ability of pasta to absorb water correlates significantly with its cooking time (21). The discontinuity of gluten matrix due to the addition of other ingredients may loosen the textural integrity, thus weakening such starch-protein network had led to increased water absorption capacity of fortified pasta (22). And as a result, increasing the overall OCT required to achieve an acceptable cooked pasta structure. Notably, Sobota et al. (18) highlighted few other factors that could be of influence, including the raw materials used, structure and shape of the pasta, and fractional composition of gluten protein. With regards to the amount of cooking loss (CL), it was reported in Ma et al. (23), that an acceptable pasta quality should retain CL of not more than 10%. The dry matter loss is associated with the protein component in pasta products (19). All pasta samples in this study had maintained cooking loss within 10%, with insignificant differences (p >0.05). The presence of gluten in wheat flours, especially with added wholemeal flour component in this pasta formulation produces strong protein-carbohydrate network, which is essential in determining high cooking quality of pasta product with minimal dry matter losses.

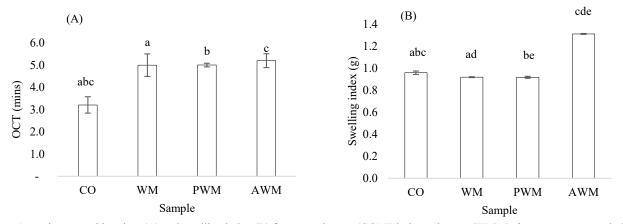


Figure 1: Optimum cooking time (A) and swelling index (B) for Control pasta (CO), Wholemeal pasta (WM), Polygonum minus + wholemeal pasta (PWM) and Amaranthus tricolor + wholemeal pasta (AWM). Bar columns with the same superscript letters have significant differences (p < 0.05)

#### **Microbial Growth Evaluation in Pasta**

The initial microbial growth of pasta was recorded and monitored for 120 hours under two storage conditions; refrigerated (4 °C) and at ambient temperature (24 °C  $\pm$  2.0). According to Ghaffar et al. (24) and Li et al. (13), 10<sup>6</sup> cfu/ml is the borderline level of food spoilage, which implies that foods exceeding this level may have detrimental effect on consumers. The present study adheres to this standard and considers 6 log cfu/ml in pasta samples as spoiled with no visible mould decay. As can be seen in Table 3, all pasta samples stored in ambient temperature deteriorated with samples having recorded over 10<sup>6</sup> cfu/ml at 48 hours. In contrast with samples stored in refrigerated condition, the pastas at 48 hours were within the range for safe consumption of less than 6 log cfu/ml and had only exceeded the spoilage mark at 72 hours. Interestingly, AWM stored in refrigerated condition revealed a longer shelf-life, as it had extended another 48 hours indicating microbial stability and managed to contain microbial growth within the borderline spoilage mark at 120 hours. This distinct effect of fortified pasta with plant powder as bio-preservative was mainly due to the reduction of microorganism growth. The Amaranthus species had been reported with active phytochemical constituents including flavonoids, polyphenols, alkaloids, tannins and terpenoids (25,26) which had been deemed effective against foodborne pathogens, particularly against Staphylococcus aureus strains (27) and Cronobacter sakazakii (28). Furthermore, having kept pastas at 4 °C had deterred rapid microbial growth and gives better shelf-life. Most foodborne pathogens including Escherichia coli, S. aureus and C. sakazakii are mesophiles, which thrives best in room temperature as these microbes reach optimal growth at 20 °C to 45 °C (29). However, cooler conditions can cause reversible bacterial cell damage, in which it may restore its former structure under much favourable storage conditions (30,31). This indicates the importance of storage condition in controlling and maintaining microbial growth.

**Table 3.** Total plate count of Control pasta (CO), Wholemeal pasta (WM), Polygonum minus + wholemeal pasta (PWM) and Amaranthus tricolor + wholemeal pasta (AWM), under ambient temperature and refrigerated conditions

Time of preservation	Ambient temperature (Log cfu/ml)					
(hrs)	СО	WM	PWM	AWM		
0 nil		nil	nil	nil		
24	<b>24</b> $5.01 \pm 0.09^{a}$		$4.73\pm0.51^{\rm A}$	$5.07\pm0.17^{b}$		
<b>48</b> $6.56 \pm 0.06^{\text{Bab}}$		$6.10\pm0.10^{Bac}$	$6.18\pm0.11^{Bbd}$	$6.66\pm0.03^{Bcd}$		
<b>72</b> TNTC		$6.53\pm0.06^{\rm a}$	$5.75\pm0.03^{\rm a}$	TNTC		
120	120 TNTC		TNTC TNTC			
Time of preservation (hrs)	Refrigerated (Log cfu/ml)					
0			nil	$\begin{array}{c} \text{nil} \\ 0.00^{\text{A}} & 4.56 \pm 0.06 \end{array}$		
24			$3.52\pm0.00^{\rm A}$			
<b>48</b> $5.31 \pm 0.19^{Ba}$		$3.52\pm0.00^{Babc}$	$5.61\pm0.17^{Bb}$	$5.61\pm0.09^{Bc}$		
72 $6.17 \pm 0.06^{a}$ 120 $6.01 \pm 0.13^{a}$		$6.14\pm0.08^{\text{b}}$	$6.01\pm0.07^{\rm c}$	$5.58\pm0.23^{abc}$		
		$6.10\pm0.02^{\text{b}}$	$6.12\pm0.39$	$5.46\pm0.05^{ab}$		

Values with the same superscript letters across each row and same capital superscript letters within each column signifies significant differences (p < 0.05)

Based on the preliminary screening result. *P. minus* had higher flavonoid and flavonol content which would have been the more effective plant species against microbial threat in contrast to A. tricolor. This phenomenon was not uncommon as it was rather expected, especially in the application of fortified pasta. Similar trend was observed in Tuersuntuoheti et al. (32) whereby the total phenol content and total flavonoid content had decreased after noodle processing, from 77.5 mg/100g and 39.5 mg/100g in uncooked noodles to 71.8 mg/100g and 36.5 mg/100g, respectively after cooking. Plant phytochemicals are heatsensitive compounds, which could have been affected during pasta processing, thus catalysing the degradation of phytochemicals in the presence of heat and water. The reduction of antimicrobial activity due to thermal degradation of complex flavonoid and phenolic structures as well as leeching of phytochemicals into the cooking water (33) could have explained the decreased ineffectiveness of PWM.

#### Moisture and pH Evaluation of Pasta

Moisture content slightly varies across pasta samples from 58.2% - 64.9% in refrigerated condition and 52.6% - 64.9% when stored in ambient temperature, but the differences between these two conditions and among pasta samples were

insignificant (p > 0.05). These results had aligned with the distinctive moisture content of cooked pasta ranged from 51.2% - 68.0% (18). The evolution of pH values in pasta samples were depicted in Figure 2. According to the Food and Drug Administration Center for Food Safety and Applied Nutrition (2007), the approximate pH for conventional macaroni, spaghetti and noodle products are ranged between 5.10 - 6.50 (34). With the increment of storage time, pH of pasta stored in ambient temperature was seen varied within the comparable range of 5.06 - 6.46(Figure 2A) and 5.13 - 6.65 for pasta stored in refrigerated condition (Figure 2B). A prominent increase of pH was observed in WM and PWM pasta in both storage conditions (p < 0.05). The pH values of both samples remained stable during the early hours. It falls to pH 5 at 72 hours and subsequently the pH levels increased to 6.50 at 120 hours.

This increment of pH to a near neutral range of 6.00 - 7.00provides optimum growth environment for microbial growth especially E. coli (35,36). As pastas are nutrient-rich food with carbohydrates and proteins, these microbes utilise those carbohydrates with the added nutrient from plant powders for growth, development, reproduction and to produce metabolites such as acids that influence the pH values of pasta (37) throughout the storage. Furthermore, A. tricolor powder managed to maintain relatively high pH level within 120 hours in ambient temperature (6.00 - 5.73) and refrigeration (5.93 - 6.27), in contrast to other pasta formulations. Similar to other leafy green vegetables, A. tricolor has an excellent nutritional value, containing rich alkalinising minerals such as iron, calcium and magnesium (38,39), which may have contributed the less acidic and near to neutral pH value of AWM.

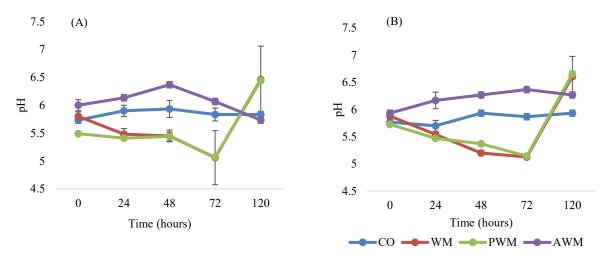


Figure 2: pH evaluation of Control pasta (CO), Wholemeal pasta (WM), *Polygonum minus* + wholemeal pasta (PWM) and *Amaranthus tricolor* + wholemeal pasta (AWM), under different storage conditions: (A) ambient temperature and (B) refrigeration

Pearson's correlation test was conducted to measure the correlation between total plate count, moisture content and pH of all pasta samples between storage conditions. Significant positive correlation was observed when stored in refrigerated condition between total plate count and pH content in CO pasta (R = 0.916, p < 0.05) and AWM pasta (R = 0.968, p < 0.05). This implies that increasing the storage time, similar increment trend was observed in both pH and TPC value of pasta (Figure 3). Previous literature also

highlighted the significant impact of storage temperature as a factor influencing the microbial evolution, structure and growth, which subsequently affects the overall pH value of samples as well (1,40). With regards to both TPC and pH values, A. tricolor powder was considered the more effective bio-preservative for fresh pasta, due to its ability to produce less acidic pasta quality as observed in AWM as compared to PWM, while maintaining within the pasta spoilage standard of  $10^6$  cfu/ml for 120 hours.

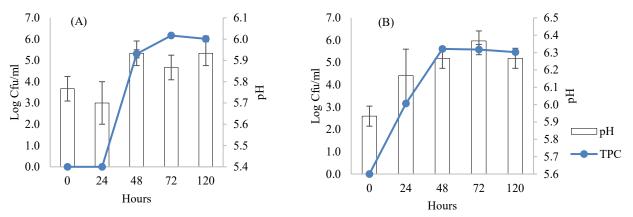


Figure 3: Total plate count (TPC) and pH values in (A) control pasta (CO) and (B) A. tricolor + wholemeal pasta (AWM) under refrigeration

#### **Sensory Evaluation of Pasta**

A 9-point hedonic rating scale was used to perform the panel test. All sensory parameters evaluated were not lower than the centre point of scale (5 = neither like nor dislike), indicating the fortified wholemeal pasta were not disliked. The highest overall acceptability was in CO pasta with 6.67 score (Table 4), as this closely resembles the more familiar commercial pasta. Fortification of plant powders contribute towards the overall colour profile, taste, aroma and appearance. When compared between fortified wholemeal pasta with plant powders, the overall acceptability of AWM

was highly favoured with the score of 6.11 in contrast to PWM with only 5.78 score. Moreover, AWM had the best appearance, chewiness, colour, firmness, taste and overall acceptability (Table 4). Enhancing functional food with plant powders generally triggers adverse consumer reaction stemming from the final product taste due to the presence of plant-derived bio-compounds, which are often associated with undesirable tastes such as bitter and astringent (11). Nevertheless, results had shown insignificant differences among all pasta samples, which implies that pasta fortification with these pasta formulations had been equally accepted by consumers.

Table 4. Sensory evaluation analysis of Control pasta (CO), Wholemeal pasta (WM), Polygonum minus + wholemeal pasta (PWM) and Amaranthus tricolor + wholemeal pasta (AWM)

Pasta samples	Sensory parameters						
	Appearance	Chewiness	Colour	Firmness	Stickiness	Taste	Overall acceptability
СО	$6.67\pm8.11$	$7.11\pm8.42$	$6.33\pm8.09$	$6.44 \pm 5.83$	$6.89 \pm 9.84$	$7.00\pm13.0$	$6.67\pm8.77$
WM	$6.22\pm5.38$	$6.33\pm9.19$	$5.89 \pm 8.31$	$5.89 \pm 7.32$	$5.78\pm 6.42$	$6.11\pm9.70$	$6.00\pm10.6$
PWM	$5.56\pm5.73$	$6.00\pm7.79$	$5.44\pm 6.88$	$5.56 \pm 8.25$	$5.67\pm7.07$	$5.67\pm5.87$	$5.78\pm8.53$
AWM	$6.33\pm5.68$	$6.00\pm5.41$	$5.89 \pm 6.81$	$5.67 \pm 6.61$	$5.56\pm 6.82$	$6.33\pm 6.61$	$6.11\pm6.79$

# CONCLUSION

Both *A. tricolor* and *P. minus* are excellent sources of flavonoid and flavonol with potential antimicrobial activity. An attempt was made to incorporate these plant powders into fortified wholemeal pasta, and to evaluate pasta shelf life at different storage conditions. Storage temperature holds a prominent role in prolonging shelf life of foods. With added potential bio-preservative, AWM showed better shelf-life as it had successfully contained within the borderline spoilage of  $10^6$  cfu/ml standard for 120 hours at 4°C. All fortified pasta in this study had maintained agreeable cooking quality format, with the standard cooking time and minimal dry matter loss, hence, considering it as high-quality pasta.

Furthermore, the overall acceptability evaluated through consumers' preference had argue AWM as the more favourable fortified pasta formulation in contrast to PWM. Although the differences were insignificant, this implies that all pasta formulations were equally acceptable. Based on the results obtained in this study, works will be prepared to further evaluate microbial growth; total plate count and particularly the yeast and mould count of fortified pasta with different formulations.

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# **CONFLICTS OF INTEREST**

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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