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Effect of vacuum drying and tray drying on the physicochemical properties during storage of chocolate

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Abstract
The aim of the present work was to study the effects of storage period on the sensory analysis degradation of polyphenol and flavonoid of the dark chocolate. The chocolate was developed from the beans that are dried using two different drying methods vacuum drying and tray drying from the lab scale fermented cocoa beans. The developed chocolates were investigated for their sensory characteristics, shelf-life and microbial analysis. The TPC of VT and TT were at 42.24±0.107g/mg of GAE and 28.33±0.11 during 0th day by the end 150th day it was reduced to 32.44±0.151 g/mg of GAE and 21.77±0.057 g/mg of GAE for chocolate prepared from vacuum dried and tray dried cocoa bean.

Keywords: Chocolate, polyphenol degradation, vacuum drying, tray drying

Introduction
Cacao (*Theobroma cacao* L.) is one of the richest source of polyphenol which is transformed to chocolate through various industrialisation process. Polyphenols are key molecules for the cocoa bean and chocolate quality which is associated with various health benefits (Peláez-Soto *et al*., 2020) [1]. Many studies have reported that consumption of dark chocolate has positive effect on cardiovascular system while lowering blood pressure (EFSA, 2012) [2]. It is also a good carrier of nutrients owing to its low moisture and water activity. Each ingredient present in the chocolate and their proportion contribute an important role in obtaining a high-quality product in terms of structure as well as their degradation during storage (Lonchampt and Hartel, 2004) [3]. Phenols were important components turned out to be a major factor in explaining the sensory acceptance of customers. In this study, stability of dark chocolate was studied throughout the storage period using the sensory data and chocolate polyphenol composition from the chocolate produced from lab scale fermented treatment of vacuum-dried (VT) and tray cocoa beans (TT).

Material and Methods
Preparation of dark chocolate bars
Dark chocolate bars were prepared using the beans that are dried using two different drying methods vacuum drying (VT) and tray drying (TT) at a temperature of 60 and 70℃ respectively from the lab scale fermented cocoa beans. Fermentation was performed using defined starter culture with the equal ratio of Y:LAB: AAB inoculum. The developed chocolates were investigated for their sensory characteristics and physiochemical.

Shelf-life studies
The shelf life of chocolate made from vacuum dried and tray dried cocoa bean was stored at 5 ºC for 150 days. The control and samples were analysed at interval of 30 days for sensory evaluation, water activity along with total polyphenol and flavonoid.

Sensory evaluation
Sensory evaluation of developed chocolate was carried out using the 9-point hedonic scale (1=extremely weak or dislike and 9=extremely strong or like) (Amerine *et al*., 2013) [4] by a panel comprising of 15 members that consisted of university students. All the samples were appropriately coded before subjected to sensory evaluation.

Water activity meter
The Novasina lab touch water activity meter was used to determine the water activity of chocolate bars. The partial temperature-regulation and the integrated sample surface temperature measurement allow precise and reproducible measurement of values through this equipment.

Total polyphenol content (TPC)
The TPC of the samples was determined by Folin–Ciocalteu method according to a method of Ioannone *et al*. (2015) [5] and reported as milligram Gallic acid equivalents per gram (mg g-1 GAE) of defatted cocoa.

Statistical analysis
The results were expressed as Mean ± SE by SPSS®20.0 software using one ANOVA for windows to determine the significant differences as per the standard procedure (Snedecor and Cochran, 2004) [6].

Result and Discussion
Sensory Evaluation
Statistical analysis showed that a highly significant ($P< 0.01$) difference was observed in all treatment throughout the storage period with regard to colour, gloss, snap, melt in mouth, sweetness, flavour and overall acceptability as shown in Table. 1. The ANOVA analysis of the sensory attributes showed statistically significant differences for the factor time in both processes ($P< 0.01$). Figure 1 and 2 shows sensory scores of different storage time have been represented pictorially using radar diagram. It also shows that different drying process intervene in the organoleptic quality of the final product (Guehi, *et al*., 2010) [7].

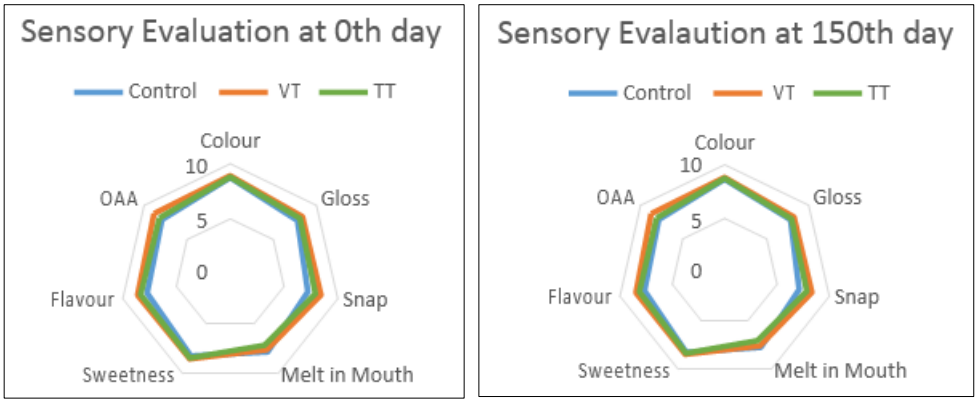

Fig 1 and 2: Sensory evaluation of chocolate at 0th and 150th of storage

Colour
The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 8.763±0.042, 8.933±0.043 and 8.816±0.042 respectively while on 150th day it was 8.641±0.043, 8.83±0.046 and 8.748±0.048 respectively. Statistical analysis revealed highly significant difference ($P< 0.01$) in the organoleptic sensory scores of colour during the storage period.

Gloss
The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 7.815±0.046, 8.361±0.045 and 8.086±0.044 respectively while on 150th day it was 7.845±0.038, 8.24±0.052 and 7.958±0.053 respectively. Statistical analysis revealed highly significant difference ($P< 0.01$) in the organoleptic sensory scores of gloss during the storage period.

Snap
The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 7.226±0.053, 8.441±0.05 and 7.941±0.054 respectively while on 150th day it was 7.126±0.052, 8.343±0.056 and 7.831±0.044 respectively. Statistical analysis revealed highly significant difference ($P< 0.05$) in the organoleptic sensory scores of snap during the storage period.

Melt in mouth
The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 7.848±0.06, 7.611±0.045 and 7.161±0.047 respectively while on 150th day it was 7.748±0.047, 7.571±0.045 and 7.058±0.047 respectively. Statistical analysis revealed highly significant difference ($P< 0.05$) in the organoleptic sensory scores of melt in mouth during the storage period.

Sweetness
The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 8.245±0.058, 8.52±0.053 and 8.43±0.046 respectively while on 150th day it was 8.25±0.053, 8.466±0.051 and 8.35±0.056 respectively. Statistical analysis revealed highly significant difference ($P< 0.05$) in the organoleptic sensory scores of sweetness during the storage period.

Table 1: Sensory Evaluation of Vacuum dried and Tray dried Chocolate

Parameters	Chocolate Variants	Days						F value
		0	30	60	90	120	150	
Colour	Control	8.763±0.042a	8.755±0.047a	8.693±0.047a	8.686±0.047a	8.653±0.05a	8.641±0.043a	1.179NS
	VT6	8.933±0.043b	8.83±0.043a	8.853±0.045c	8.826±0.046a	8.833±0.043b	8.83±0.046b	0.855 NS
	TT6	8.816±0.042ab	8.751±0.053a	8.766±0.045ab	8.741±0.046a	8.775±0.055ab	8.748±0.048ab	0.315 NS
	F Value	4.189**	0.839*	3.032**	2.268*	3.329**	4.173**	
	Control	7.815±0.046a	7.845±0.048a	7.823±0.051a	7.831±0.048a	7.838±0.046a	7.845±0.038a	0.067 NS
	VT6	8.361±0.045c	8.333±0.053c	8.291±0.048c	8.243±0.048c	8.248±0.058b	8.24±0.052b	1.007 NS

Gloss	TT6	8.086±0.044b	8.1±0.055b	8.078±0.052b	8.043±0.05b	7.976±0.043a	7.958±0.053a	1.416 NS
	F Value	35.734**	21.624**	21.337**	17.515**	17.410**	17.362**	
Snap	Control	7.226±0.053a	7.168±0.049a	7.188±0.043a	7.138±0.047a	7.178±0.051a	7.126±0.052a	0.528 NS
	VT6	8.441±0.05c	8.313±0.051c	8.366±0.053c	8.341±0.054c	8.373±0.052c	8.343±0.056c	0.673 NS
	TT6	7.941±0.054b	7.943±0.049b	7.961±0.045b	7.953±0.048b	7.933±0.054b	7.831±0.044b	0.928 NS
	F Value	133.765**	136.357**	156.668**	147.397**	130.274**	141.268**	
Melt in mouth	Control	7.848±0.06c	7.868±0.05c	7.855±0.054c	7.79±0.05b	7.813±0.051b	7.748±0.047c	0.752 NS
	VT6	7.611±0.045b	7.665±0.047b	7.65±0.054b	7.661±0.044b	7.686±0.045b	7.571±0.045b	0.790 NS
	TT6	7.161±0.047a	7.095±0.048a	7.065±0.049a	7.06±0.054a	7.053±0.047a	7.058±0.047a	0.717 NS
	F Value	45.654**	67.156**	60.283**	61.445**	71.325**	57.971**	
Sweetness	Control	8.245±0.058a	8.235±0.052a	8.226±0.062a	8.253±0.054a	8.211±0.052a	8.25±0.053a	0.080 NS
	VT6	8.52±0.053b	8.52±0.045c	8.5±0.048b	8.451±0.049b	8.391±0.048b	8.466±0.051b	1.006 NS
	TT6	8.43±0.046ab	8.375±0.049ab	8.355±0.051ab	8.353±0.049ab	8.365±0.052ab	8.35±0.056ab	0.347 NS
	F Value	7.000**	8.395**	6.320**	3.732**	3.600**	4.059**	
Flavour	Control	7.76±0.051a	7.745±0.055a	7.765±0.054a	7.663±0.05a	7.65±0.055a	7.651±0.05a	1.137 NS
	VT6	8.636±0.043c	8.603±0.051c	8.583±0.053c	8.568±0.055c	8.583±0.046c	8.476±0.055c	1.105 NS
	TT6	8.346±0.045b	8.306±0.056b	8.265±0.056b	8.261±0.049b	8.246±0.054b	8.17±0.046b	1.324 NS
	F Value	90.546**	63.957**	56.506**	79.541**	81.581**	67.568**	
OAA	Control	7.931±0.057a	7.87±0.056a	7.841±0.046a	7.863±0.05a	7.84±0.05a	7.85±0.053a	0.425 NS
	VT6	8.835±0.043c	8.86±0.047c	8.845±0.043c	8.818±0.041c	8.833±0.038c	8.753±0.058c	0.658 NS
	TT6	8.213±0.049b	8.17±0.046b	8.133±0.046b	8.091±0.041b	8.155±0.043b	8.121±0.049b	0.830 NS
	F Value	84.342**	101.634**	128.359**	125.115**	130.519**	73.726**	

Flavour

The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 7.76±0.051, 8.636±0.043 and 8.346±0.045 respectively while on 150th day it was 7.651±0.05, 8.476±0.055 and 8.17±0.046 respectively. Statistical analysis revealed highly significant difference ($P< 0.05$) in the organoleptic sensory scores of flavour during the storage period.

Overall Acceptability

The mean ± SE sensory scores of colour obtained for 0th of storage at 4˚C for control, VT and TT were 7.931±0.057, 8.835±0.043 and 8.213±0.049 respectively while on 150th day it was 7.85±0.053, 8.753±0.058 and 8.121±0.049 respectively. Statistical analysis revealed highly significant difference ($P< 0.05$) in the organoleptic sensory scores of overall acceptability during the storage period.

Water activity of chocolate during storage

The water activity of the control and developed chocolate showed a significant difference ($P\leq 0.05$) which was analysed using water activity meter. Table 2 shows the water activity of the chocolate samples during 150 days storage period. The water activities of control, VT and TT at 0th day were 0.321±0.01, 0.29±0.005, 0.361±0.017 respectively and 150th day were 0.44±0.023, 0.42±0.008,
0.516±0.012 respectively. The significant difference between the samples may be due to the different drying methods of cocoa bean and results were similar to the findings by Tolve *et al*., 2018 [8]. Water activity remained low throughout the storage period thus preventing the growth of undesirable microorganism.

Table 2: Water activity of chocolate during 150 days of storage

	0	30	60	90	120	150	F Value
Control	0.321±0.01Aab	0.356±0.007ABb	0.371±0.019ABCb	0.406±0.009BCDb	0.421±0.013CDb	0.44±0.023Eb	8.724**
VT6	0.29±0.005Aa	0.308±0.008ABa	0.34±0.008BCa	0.368±0.007CDa	0.4±0.005DEa	0.42±0.008Ea	48.116**
TT6	0.361±0.017Ab	0.405±0.014ABc	0.445±0.019BCc	0.466±0.009CDc	0.486±0.024CDc	0.516±0.012Dc	11.071**
F Value	8.684**	21.527**	10.429**	31.810**	7.687**	10.218**	

Microbial analysis of chocolate during storage

The microbial studies by standard plate count, coliform count, yeast and mold count during storage intervals of 0 to 150 days for the control and chocolate made from vacuum dried and tray dried chocolate were studied. At 0th day the SPC of were 1.631±0.083, 1.581±0.1, 1.697±0.102 respectively for control, VT and TT while at 150th day the values were 2.887±0.043, 2.851±0.035, 2.944±0.037 respectively as shown in **Table 3**. It was observed that no coliform and mold count in any of the samples during different storage intervals of 0 to 150 days at 4 ºC, which was due to low water activity and low moisture content along with hygienic practices during production and storage.

Table 3: Microbial analysis of chocolate during 150 days of storage

Sample	0	30	60	90	120	150	F Value
Control	1.631±0.083Aa	2.097±0.053Bb	2.347±0.018Ca	2.585±0.041Da	2.74±0.038Ea	2.887±0.043Fa	84.446**
VT6	1.581±0.1Aa	1.906±0.048Ba	2.281±0.029Ca	2.531±0.038Da	2.707±0.039DEa	2.851±0.035Ea	81.255**
TT6	1.697±0.102Aa	2.217±0.046Bb	2.383±0.022Bb	2.637±0.033Ca	2.785±0.04CDa	2.944±0.037Da	70.257**
F Value	0.373NS	9.976NS	4.738NS	1.897NS	0.982NS	1.447NS	

Total Polyphenol content

The stability of total polyphenol content in individual dark chocolate samples at 4°C over the course of 150 days of storage is presented in Fig.3. Initially at 0th day the TPC of control, VT and TT were 18.166±0.102, 42.241±0.107 and 28.33±0.11 and at 150th day it was decreased significantly to

10.73±0.1, 32.446±0.151 and 21.77±0.057 respectively. Chocolate prepared from vacuum dried cocoa bean is found to contain higher polyphenol due to uniform heating at short time with lesser oxidation preventing the action of polyphenol oxidase.

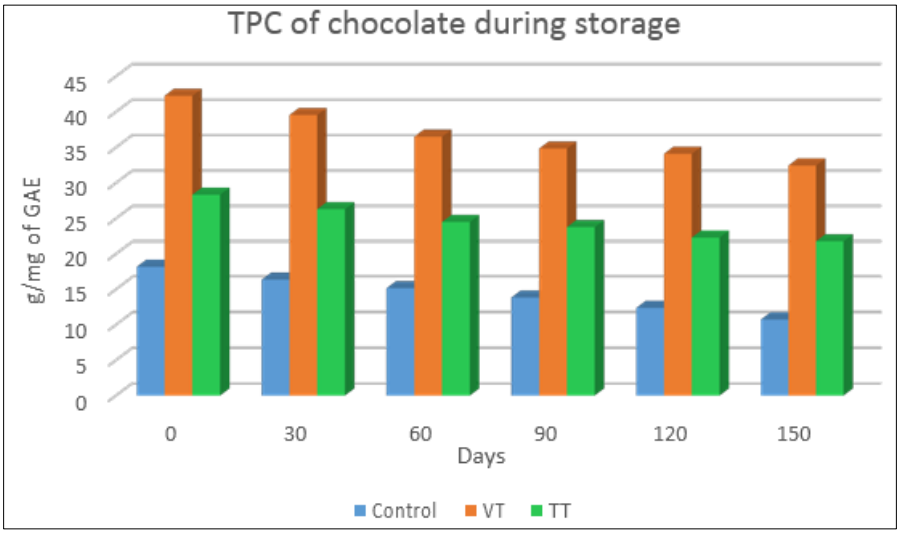

Fig 3: Total Polyphenol content of dark chocolate

Conclusion

The chocolate was prepared by different drying methods had significant difference in their physicochemical analysis such as sensory attributes, shelf-life, water activity, microbial analysis and total polyphenol content. The study revealed that the quantity of total polyphenol content in dried cocoa beans could be related to the sensory evaluation attribution. The degradation of polyphenol increases as the storage days with significant effect on sensory attribute has between the treatments. The storage process has also an impact on the water activity, microbial analysis of chocolate but was was in line with the permissible limit.

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Conflict of interest

The authors declare no conflict of interest pertaining to this manuscript.

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