Effects of D-Tagatose and Inulin on Some Physicochemical, Rheological and Sensory Properties of Dark Chocolate

M. Shourideh, A. Taslimi, MH. Azizi, and MA. Mohammadifar

Abstract—**Chocolate is very popular among consumers of all ages. Replacing sucrose with low digestible carbohydrates (LDCs) will result in reduction of its energy content and glycemic index, as well as prevention of tooth decay. Inulin, and D-tagatose were used as sucrose substitutes in dark chocolate formulas. The inulin : D-tagatose ratios in the chocolate mixtures were 100:0, 75:25, 50:50, 25:75, and 0:100. With a reduction of the inulin content and increase of D-tagatose content, the moisture contents of the chocolate samples decreased, while their Aw increased. As the amount of D-tagatose increased in formula the hardness of the samples went up. In case of chocolate color indices, the least amounts of L*,a*,b*,c* and hue° were observed in chocolate samples with 100% inulin. The result indicated that reduction of the inulin resulted in decrease of the apparent and plastic viscosity and increases the yield stress and linear shear stress. Overall acceptability of the chocolate samples increased by increase of the D-tagatose level. It can be concluded that in chocolate samples with inulin: D-tagatose ratios of 25%-75% and 100%D-tagatose are proper for sucrose substitutes in dark chocolate formula.**

Index Terms—**Dark chocolate, D-tagatose, inulin, rheology, stevia.**

I. Introduction

Chocolate has got exclusive taste flavor texture and biologically active compounds which they have got antioxidant effects. In recent decade the consumption of low calorie foods is recommended for people with particular health problems like, diabetes, obesity and heart diseases. In order to reduce calories, glycemic index and also prevention from dental cavities, there is possibility to replace the sucrose with low digestible carbohydrates and bulking agents like inulin , simple sugar like D- tagatose, polyols and senthetice sweeteners [1].Among these substitutes, sugar alcohols have been used most frequently[2], but their laxative, cooling and hygroscopic properties are few limiting factors for their use in most foodstuffs[2], [3]. D-Tagatose a natural ketohexose, is an isomer of D-galactose and D-arabinose .it is a reducing sugar. The sweetness of D-tagatose is 92% of the sucrose when compared in 10% of their solutions. D-Tagatose has a sucrose-like taste with no cooling effect or after taste. It works as a flavoue enhancer texturizer (humectants and stabilizer). D-Tagatose has numerous health benefits it attenuates glycemic responce and reduces the symptoms associated with type 2 diabetes and also has prebiotic effect [4]. Inulin is a polydisperse fructan. It is a mixture of oligomers and polymers of of fructosyl units with one glucosyl unit at the end of the chain . Inulin reduces the calories and fat, acts as a fiber and increases the absorption of calcium by the body and it does not have influence onserum glucose. It is used for fat or sugar replacement as a low caloric bulking agent and acts as a texturizing agent [5], [6]. It seems there is a need to do new researchs to find favaorable substitutes with minimum side effects on the consumers as well as on the special characteristic of product. In this study Inulin, and D-tagatose were used as sucrose substitutes in dark chocolate formulas. Physical, chemical, rheological and sensory

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II. Materials and Methods

Alkalized cocoa powder (10-12%) (Shokinag cocoa, Germany), deodrised cocoa butter (KLK-Kepong, Malysia), Inulin GR (Orafti, Belgium), D-tagatose (Damhert,Belgium), Sucrose (Karoon Co., Iran), Soy Lecithin (Cargill, Netherland), Stevia (Stevian Biotechnology Co., Malysia) and vanilla powder (Panda, China) were used for the production of dark chocolates .Experimental samples were produced by mixing Sucrose, D-tagatose and Inulin powders(51.4%),cocoa powder(16%), cocoa butter (32%), lecithin(0.5%) and vanillin (0.1%). Since Inulin and D-tagatose are less sweet than the sucrose the samples were additionally sweetened with stevia. Concentration of stevia was adjusted from the following equation:

[0.92% × D-tagatose content (% w/w) + 0.1% × Inulin content (% w/w)] +120% × stevia content (% w/w) = 1% × sucrose content (% w/w)

Coefficients of 0.92, 0.1,120 and 1 reflect to the relative sweetness of D-tagatose, Inulin, stevia and Sucrose(standard).Chocolate masses were prepared in a laboratory ball mill (rotantional velocity 85 rpm, 50 ℃, 3 hours) and were ground until average paticle size reached approximately to 25 µm. vanillin and stevia added at the final stage. The chocolate samples were incubated for excess water loss at temperature 50 ℃ for 12 hours. On completion of grinding in the ball mill, the chocolate masses were tempered at laboratory temperer (rotantional velocity of 75 rpm), then chocolate masses were poured to polycarbonate mould, cooled and demoulded from the mould. Chocolate bars were wrapped in aluminum foil and stored at refrigerator

(10±2 ℃). For rheological measurements samples were stored in sealed plastic containers at ambient temperature (20±2℃).

The moisture content, reducing sugar (before and after hydrolysis), fat content, protein and pH of chocolate samples were determined according to 931.04, 933.04, 945.34, 939.02 and 970.21 methods of AOAC, respectively [7]. Water activity (Aw) was also measured using the Aw-sprint TH500 (Novasina, Pfäffikon, Switzerland) at 25 ℃. Hardness of solid tempered chocolate was measured usingH50KS universal testing machine (Hounsfield, Surrey, UK) with a penetration of a cylindrical flat-end probe(d = 1.6 mm, 500 newton cell load). Maximum penetration was 6mm in sample (100 ×20, depth 10 mm) with penetration speed of 1mm/s and was reported as the maximum force (*N*). HunterLab Colorimeter was used for colour measurements of solid chocolate size (150×150mm). Colour images of chocolate surfaces were converted into XYZ tristimulus values, which were converted to CIELAB system: *L**, luminance ranges were from zero(black) to 100(white); a*(green to red) and b*(blue to yellow) with values from -120 to +120 . hue angle (h ℃)=arctan(*b**/ *a**); chroma (*C**)=[(*a**)2 +(*b**)2]1/2 calculated .

Rheological properties of dark chocolates were determined using a rheometer (Anton paar - MCR 301) operating in a controlled shear rate rotation mode with concentric cylinder geometry (Z3 DIN) according to IOCCC methods . The samples were incubated at 50℃ for 90 min for melting. Samples were pre-sheared at $5s^{-1}$ for 500 seconds at 40 ℃ for homogenizing and temperature control .Shear stress was measured at 40 ℃ (±0.1 ℃) as a function of increasing shear rate from 5 to 50 s^{-1} 1 with 18 positions 180 s (upward), then decreasing from 50 to 5 s^{-1} duration 180s (downward).

Sensory attributes of dark chocolates like texture, snapness, sweetness, flavor, taste, melting rate, mouth feel, color, and overall acceptability (sum of aforementioned eight sensory attributes) were evaluated using ranking test (high quality = 6 and low quality =1) based on a balanced incomplete block design by 15 trained panelists. Chocolates were presented randomly in three-digit coded containers to the panelists.

Analysis of ANOVA, variance and least significant differences tests were also used (P <0.05) and differences between mean values were expressed by using SPSS software (Version 14.0, SPSS Inc., Chicago, IL).

III. Results and Discussion

A. Effects of Sugar Substitutes on Physicochemical , Hardness and Color Parameters of the Chocolate

Increase of the inulin leads to an increase in moisture content "Table I". Inulin posseses hydrophilic groups on its chemical formula which is the cause of increasing and preserving of the moisture in samples with high content of inulin. Inulin and oligofructose can keep breads and cakes moist and fresh for longer period of time [8]. It was reported that cookies with inulin and less fat tend to have a higher moisture content and therefore less crisp [9]. In milk chocolate with inulin (up to 50 %), the moisture content is diminished, whereas with further addition of inulin, the moisture content was conversely increased [10]. Cardarelli et.al (2008) reported that petit-suisse cheese with high levels of inulin had high moisture content[11].

The lowest moisture contents belongs to chocolates containing 100% D-tagatose, there were no difference between this formula and control. Originally tagatose powder has lower moisture to compare with inulin powder. D-tagatose itself showed less tendency to absorb and preserve the moisture. Gaio reported that moisture content of the dark chocolate samples with D-tagatose were lower than control with sucrose[12].

As D-tagatose contents were increased, aw contents increased too and there was no difference between 75% D-tagatose-25%inulin, 100% D-tagatose and control (P< 0.05). Inulin increased

TABLE I: Effects of Various Formulations on Mean Values of Physicochemical and Mechanical Characteristic and color of Dark Chocolates

Formulation Weight ratio of sugar substitutes (g/100g chocolate)	Moisture content (g)	*A*w	Hardness (N)	Reducing sugar before hydrolysis (%)	Reducing sugar after hydrolysis(%)	Energy (Kcal/100 g)	calorie reduction%
100%inulin	1.79±0.015^{a}	0.281±0.001^{a}	21.50±0.22^{a}	2.20± 0.06	50.98±0.12	379.6	27.5
75%inulin-25% D-tagatose	1.52±0.03^{b}	0.315±0.004^{b}	22.65±0.93^{ab}	13.98±0.13	50.9±0.36	383.5	26.7
50%inulin-50% D-tagatose	1.34±0.01^{c}	0.325±0.003^{c}	24.35±0.38^{b}	25.6±0.33	51.2±0.03	387.4	26
25%inulin-75% D-tagatosse	1.05±0.041^{d}	0.335±0.005^{d}	26.12±0.76^{bc}	39.1±0.2	51.18±0	391.2	25.2
100% D-tagatose	0.71±0.02^{e}	0.336±0.002^{d}	27.82±0.58^{c}	51.3±0	51.4±0.09	395.1	24.5
control(100%sucrose)	0.68±0.01^{e}	0.333±0.004^{cd}	26.80±1^{c}	0.37±0.02	51.9±0.17	523.6	-

Different letters within the columns indicating the significant differences between different dark chocolate formulations (p<0.05) moisture content but decreased Aw , inulin can bind with water molecules and decreases free moisture and Aw .Oligofructose and inulin will reduce water activity ensuring high microbiological stability [8].

The mean values of fat content of all chocolates were the same (33.41%) and equal to the added cocoa butter fat .

The amount of reducing sugars before hydrolysis increased as D-tagatose content was increased.

Determination of the amount of reducing sugar after hydrolysis proved that inulin and D-tagatose were compeletely hydrolysed.

The hardness of chocolates were related to the type of sugar in chocolates .Sample with 100%inulin was softer than the other formulas and hardness increased as D-tagatose content were raised. Formulation with 100% D- tagatose was the hardest and was similar to control (100% sucrose). Inulin absorbs the moisture and it causes the hardness. Drewnowski (1997) reported that cookies with inulin (low fat) were less crisp [9] . Gaio also reported that the hardness of the chocolate with sucrose or tagatose was the same [12].

CIELAB parameters *L**, *b** , *C** and h◦ and c* Increased as Inulin contents decreased (p<0.05) "Table II". Inulin absorbs moisture, light scattering and lightness decreased and chocolate seemed darker. Bolenz et al. (2006) reported that chocolate samples with 20% inulin was the most brown, and had the lowest l* among other texurizing agen in milk chocolate. Dark chocolate became lighter as D-tagatose content increased. D-tagatose as a reducing sugar caused fading of the color and l*, a* and b* increased as tagatose content increased. Darkness of chocolate sample with D-tagatose was different significantly from control (chocolate sample with sucrose [12]

TABLE II: INFLUENCE OF VARIOUS FORMULATIONS ON COLOR PARAMETERS OF DARK CHOCOLATE

samples	L^*	a^*	b^*	C^*	hue °
100%inulin	16.09±0.11^a	4.95±0.04^a	3.08±0.01^a	5.83±0.04^a	35.7±0.26^a
75%inulin-25% D-tagatose	18.4±0.14^b	6.05±0.06^b	4.11±0.01^b	7.26±0.12^b	38.93±0.32^b
50%inulin-50% D-tagatose	19.58±0.07^c	6.6±0.03^c	4.75±0.01^c	8.14±0.05^c	41.06±0.30^c
25%inulin-75% D-tagatosse	20.08±0.07^d	7.48±0.03^d	6.65±0.02^d	10.04±0.07^d	50.94±0.37^d
100% D-tagatose	22.17±0.12^e	9.47±0.04^e	9.53±0.03^e	13.53±0.18^e	57.63±0.07^e
Control	17.6±0.02^f	7.48±0.01^d	6.01±0.02^f	9.63±0.08^f	45.99±0.12^f

Different letters within the columns indicating the significant differences between different dark chocolate formulations (p<0.05)

B. Effects of Sugar Substitutes on Sensory Properties

In sensory evaluation, as the amounts of the D- tagatose increased ,sweetness, , flavor, taste, texture snapness ,melting rate, mouth feel, color and overall acceptability gained higher scores "Table III" and Samples 25%inulin-75% D-tagatosse and 100% D-tagatose had no difference with control (p<0,05). Nevertheless, in terms of the other sensory parameters, chocolates formulated with high ratios of inulin received lower sensory scores which were significantly different from the other samples(P<0.05). Sample with 100% inulin had the darkest color and was more suitable for dark chocolate.

Moreover, samples 100% D-tagatose and 100% inulin showed the highest and lowest overall acceptability respectively. Golob et al. (2004) reported that stickiness, mouth loading and solubility of three types of chocolates with inulin were evaluated as low quality chocolate [13]. the supplement of inulin influences on the viscosity and low solubility of the product [14]. D-tagatose were improved the sensory parameters as compared to inulin. Gaio (2004) claimed that at a dosage of 0.2-1%, D-tagatose improves the flavour profile of the soft drinks made with high intensity sweeteners. Also, Gaio (2004) reported that D-Tagatose was the compound which comes close to sucrose when used in dark chocolate [12].

C. Effects of Sugar Substitutes on the Rheological Properties

Rheograms of control and dark chocolates are drawn "Fig. 1".

Hysteresis loops were observed in all samples and control during upward and downward shear rate sweeps. Yield stress and viscosity decreased in the same shear rate. Evalution of the rheological behavior of the chocolate samples, the shear stress vs. shear rate data was fitted on few applicable mathematical models including windhab (1), Herschel-Bulkley (2), Casson (3), Bingham (4) and Power law (5):

$$\tau = \tau_0 + (\tau_1 - \tau_0).[1 - \exp(-\dot{\gamma}/\dot{\gamma}^*)] + \eta_\infty.\dot{\gamma} \quad (1)$$

$$\tau = \tau_0 + \eta_p(\dot{\gamma})^n{}_1 \quad (2)$$

$$\tau^{0.5} = \tau_0{}^{0.5} + k_1(\dot{\gamma})^{0.5} \quad (3)$$

$$\text{Eq.4: } \tau = \tau_0 + \eta_{pl}\ \dot{\gamma} \quad (4)$$

$$\text{Eq.5: } \tau = k_2 \, \dot{\gamma}^{n_2} \quad (5)$$

where τ is shear stress (Pa), τ_1 linear shear stress (Pa) , η_∞ infinite viscosity (Pa s), $\gamma_\circ^*$ characteristic shear rate (s^{-1}), η_{pl} plastic viscosity (Pa s), $\dot{\gamma}$ shear rate (s^{-1}), τ_0 yield stress (Pa) , K_1 Casson viscosity [(Pa s)$^{0.5}$] , k_2 consistency coefficient (Pa sn) , n_1 flow behaviour index (dimensionless) and n_2 is power low index (dimensionless).

TABLE III: INFLUENCE OF VARIOUS FORMULATIONS ON SENSORY PROPERTIES OF DARK CHOCOLATES

Sample	Color	Flavor	Taste	Melting rate	Sweetness	Mouth feeling	Hardnees	Texture	Overall Acceptabillity
100%inulin	5.33^{a}	1.66^{a}	1.93^{a}	2.06^{a}	2^{a}	1.8^{a}	1.66^{a}	1.8^{a}	14.86^{a}
75%inulin-25% D-tagatose	4.6^{ab}	2.1^{a}	2.33^{a}	2.6^{a}	2.73^{a}	2.26^{a}	2.66^{b}	2.26^{a}	18.26^{b}
50%inulin-50% D-tagatose	3.6^{c}	3.46^{b}	3.2^{b}	3.9^{b}	3.73^{b}	4^{b}	3.33^{bc}	3.53^{b}	28.60^{c}
25%inulin-75% D-tagatosse	1.86^{d}	4.8^{c}	4.4^{c}	4.4^{b}	4.06^{b}	4.46^{b}	4.13^{cd}	4.66^{c}	35.46^{d}
100% D-tagatose	1.4^{d}	4.4^{bc}	4.7^{c}	4.3^{b}	4.2^{b}	4.33^{b}	4.8^{d}	4.46^{c}	36.93^{d}
control	4.3^{b}	4.53^{c}	4.66^{c}	3.8^{b}	4.33^{b}	4.13^{b}	4.46^{d}	4.26^{cb}	33.46^{d}

Different letters within the columns indicating the significant differences between different dark chocolate formulations ($p<0.05$)

TABLE IV: EFFECTS OF SUGAR SUBSTITUTES ON FITTING OF EXPERIMENTAL DATA ON THE MATHEMATICAL MODELS BASED ON DETERMINATION COEFFICIENT AND STANDARD ERROR PARAMETERS

Sample	model	R^2	SE		model	R^2	SE
100%inulin	Windhab	0.99999	0.70149	25%inulin-75% D-tagatosse	Windhab	0.99999	0.14727
	Hershel-Bulkley	0.99998	0.75222		Hershel-Bulkley	0.99996	0.42377
	Casson	0.99969	3.7528		Casson	0.9991	2.117
	Bingham	0.99986	6.2756		Bingham	0.99751	3.5236
	Ostwald-de Waal	0.99529	14.504		Ostwald-de Waal	0.98482	8.6659
75%inulin-25% D-tagatose	Windhab	0.99996	0.18151	100% D-tagatose	Windhab	0.99998	0.19568
	Hershel-Bulkley	0.99997	0.55242		Hershel-Bulkley	0.99979	0.77177
	Casson	0.99938	3.0326		Casson	0.99893	2.4355
	Bingham	0.99825	4.9815		Bingham	0.99494	3.9452
	Ostwald-de Waal	0.99121	11.382		Ostwald-de Waal	0.96511	9.8542
50%inulin-50% D-tagatose	Windhab	0.99998	0.2285	control	Windhab	0.99998	0.42941
	Hershel-Bulkley	0.99997	0.48609		Hershel-Bulkley	0.99981	1.0525
	Casson	0.99905	2.6329		Casson	0.99979	1.5806
	Bingham	0.99664	3.4961		Bingham	0.99837	4.3604
	Ostwald-de Waal	0.97036	9.5716		Ostwald-de Waal	0.9781	11.379

In order to find the approperate model and The fitting of experimental data with suitable model was evaluated on the basis of the coefficient of determination (R^2) and standard error (S.E.) parameters. Statistical calculation indicated that the windhab, Herschel-Bulkley and Casson models, provided the highest R^2 values and lowest standard errors respectively "Table IV". As a result, the substitution of sucrose with inulin and D-tagatose had influence on rheological parameters, but had no effect on the fitting of mathematical models "Table V", by reduction of the inulin and increasing of D- tagatose contents yield stress increased highest and lowest yield stress achieved in control and sample

100%inulin. Sample 100% D-tagatose had no difference with control (P <0.05). Linear yield stress also increased as D-tagatose content increased. The apparent viscosities (shear rate 40 s^{-1}) of samples were shown in Table V.

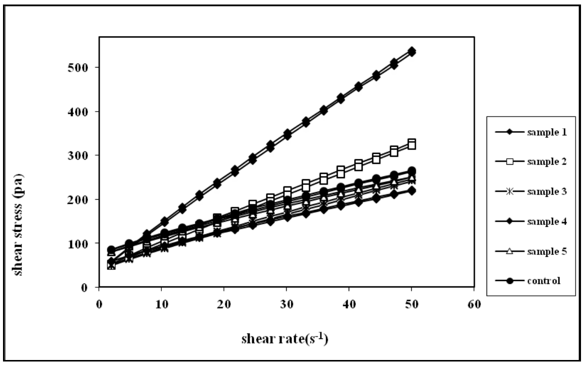

Fig. 1. Effects of sugar substitutes on shear stress versus shear rate rheograms of dark chocolates.

Sample 100%inulin had the highest apparent viscosity and its difference with other samples and control was notable. Apparent viscosity decreased, as inulin content decreased. The lowest apparent viscosity was shown in sample 25% inulin-75% D-tagatose. Sample with 100%tagatose had no difference with control. By decreasing of the inulin content , plastic viscosity was decreasing .The highest plastic viscosity belonged to sample 100%inulin. Samples 25%inulin-75% D-tagatosse and 100% D-tagatose had no difference with a control (*P* <0.05). flow behaviour index decreased as inulin decreased. The lowest content was shown in control and had no differences with samples 25%inulin-75% D-tagatosse and 100% D-tagatose (*P* <0.05). The inulin reduces the yield stress in chocolate. Low yield stress in the samples with high percent of inulin indicating that interaction forces between inulin particles were weak and for overcome to these forces and make flow of the chocolate less force in needed. The difference on the structures of inulin (oligosaccharide) with D-tagatose (monosaccharide) and sucrose (disaccharide) will be one of the factors for the interaction between the particles in chocolate making and their resistance to flow. The reduction of yield stress happened when inulin used as afiller in chocolate making [15].The samples with high percent of D-tagatose had only slight difference in yield stress as compared with the control. This is because of the structural similarities of this two sugars.

Results of this study indicated that when inulin was used in high percent, it will cause the increment of the plastic and apparent viscosity. In low percent of inulin flow property improves but apparent viscosity reduces. Inulin in low and non fat foods like: chocolate, salad dressing, and mouss desert increases their viscosity and texture also improves [16]. Since inulin has potential to absorb water molecules and protect the moisture, so the viscosity increases [13]. Grittenden and Playne (1997) said that high molecular weight oligosachrides and polysachrides increase viscosity and also improves texture and mouth feeling [17]. Inulin as a texturizing agent in chocolate has adverse effect on viscosity and solubility of chocolate but in low amount reduces viscosity (13). Inulin with 20% concentration in chocolate reduced the viscosity and yield stress [15]. The lowest viscosity was achieved when the ratio of inulin and tagatose was 25% and 75% respectively. Viscosity of sample with 100% tagatose did not show significant difference from the control and it had acceptable viscosity, this was also reported for milk and dark chocolates which were contained tagatose and sucrose[12].

TABLE V: INFLUENCE OF VARIOUS COMBINATIONS OF THE SUGAR SUBSTITUTES ON THE RHEOLOGICAL PARAMETERS BASED ON THREE MODELS FOR DARK CHOCOLATE

Models / Sample	Windhab model			Casson model	Herschel-Bulkley model		Apparent viscosity* (Pa s)
	Real Yield stress	liner Yield stress	η_∞ Infinite viscosity	Casson viscosity (Pa s)	Herschel-Bulkley index	Plastic viscosity (Pa s)	
100%inulin	33.96±0.88^a	61.24±1.1^a	9.44±0.33^a	2.73±0.04^a	0.94±0.01^a	12.21±0.3^a	10.95±0.26^a
75%inulin-25% D-tagatose	36.93±0.73^b	62.66±1.1^{ab}	5.34±0.20^b	1.93±0.03^b	0.90±0.02^{ab}	8.53±0.38^b	6.84±0.15^b
50%inulin-50% D-tagatose	41.14±0.77^c	65.14±1.6^b	3.63±0.11^c	1.51±0.01^c	0.87±0.02^{bc}	6.86±0.4^c	5.23±0.18^c
25%inulin-75% D-tagatosse	50.39±0.84^d	77.92±1.4^c	2.88±0.12^{de}	1.27±0.03^d	0.83±0.03^{cd}	6.69±0.25^c	4.78±0.14^d
100% D-tagatose	72.56±0.718^e	113.41±1.3^d	2.53±0.11^d	1.26±0.01^d	0.82±0.01^d	6.78±0.4^c	5.55±0.15^e
control	75±0.80^f	127.87±1.1^e	3.05±0.14^e	1.32±0.02^d	0.80±0.04^d	7.41±0.7^c	5.82±0.14^e

Different letters within the columns indicating the significant differences between different dark chocolate formulations (p<0.05)
*apparent viscosities are reported at a constant shear rate (40 s^{-1}) for comparison purposes

IV. Conclusion

It can be concluded that in chocolate samples with inulin-D-tagatose ratio of 25%-75% and 100% D- tagatose are the proper sucrose substitutes. since inulin is a dietetic fiber and D-tagatose and inulin both have prebiotic properties, chocolate samples prepared with these sugars are also desirable from a nutritional point of view and they can be considered as functional foods.

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