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Theoretical Evaluation on The Interaction Between Triglycerides and Methylxanthines Using Density Functional Theory B3LYP/6-31G(d,p) and Molecular Electrostatic Potential

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The binding, interaction and distortion energies between the main triglycerides, palmitic-oleic-stearic (POS) in cocoa butter *versus* palmitic-oleic-palmitic (POP) in refined, bleached and deodorized (RBD) palm oil with cocoa's methylxanthines (caffeine, theobromine, and theophylline) during the production of chocolate were theoretically studied and reported. The quantum mechanical software package of Gaussian09 at the theoretical level of density functional theory B3LYP/6-31G(d,p) was employed for all calculations, optimization, and basis set superposition errors (BSSE). Geometry optimizations were carried out to the minimum potential energy of individual species and binary complexes formed between the triglycerides, methylxanthines and polyphenols. The interaction energies for the optimized complexes were then corrected for the BSSE using the counterpoise method of Boys and Bernardi. The results revealed that the binding energy and interaction energy between methylxanthine components in cocoa powder with triglycerides were almost of the same magnitude (13.6-14.5 and 3.4-3.7 kJ/mol, respectively), except for the binary complex of POS-caffeine (25.1 and 10.7 kJ/mol, respectively). Based on the molecular geometry results, the hydrogen bond length and angle correlated well with the interaction energies. Meanwhile, the POS-caffeine complex with two higher and almost linear bond angles showed higher binding and interaction energies as compared to the other methylxanthines. Therefore, a donor-acceptor analysis showed that the hydrogen bond strength was proven using the molecular electrostatic potential (MEP), which resulted in parallel outcomes. The research results were believed to be one of the factors that contributed to the rheological behaviour and sensory perception of cocoa products, especially chocolate.

Keywords: DFT study, Cocoa butter, Triglycerides, Methylxanthines.

INTRODUCTION

Cocoa butter is the key element in the production of chocolate. However, natural cocoa fat production is constantly decreasing from day to day, as well as cocoa cultivation worldwide. These situations have encouraged the food industries, especially chocolate manufacturers, to search for cocoa butter replacers and substitutes. Numerous studies have been conducted to produce quality cocoa butter replacers, which are expected to have similar properties and are compatible with cocoa butter as a mixture for chocolate manufacturing [1-4]. However, to date, no reports have been published on the synergistic interactions between selected species of cocoa and cocoa butter products, such as between triglyceride and methylxanthines (*i.e.* caffeine, theobromine and theophylline). Rios *et al.* [2] reported that the different formulation of triglyceride and any other lipids in food products will greatly influence the rheological and sensory properties of the product.

In various compounds of chocolate, the methylxanthine compounds are known to have psychoactive effects [3]. The levels of methylxanthines (*i.e.* caffeine, theobromine and theophylline) can affect the flavor of the cocoa beans, which simultaneously impacts the taste of chocolate [4]. This study presents an evaluation of the binding, distortion, and interaction energies between the important components in cocoa and cocoa butter products. These components are potentially manipulated and simultaneously affect the rheological behavior and sensory perception of cocoa products, especially chocolate. Donor-acceptor analysis and molecular electrostatic potential (MEP) are performed and calculated accordingly to show and substantiate the hydrogen bond strength present between the complexes.

Binding energy refers to stabilization, total interaction, total bond or bond dissociation energy. There are two energies that contribute toward the binding energy, namely the interaction and distortion energies. In the calculation of binding energy, the relaxation effects occur when the dimer or complex breaks apart and allows the monomers to become isolated from each other [5]. Therefore, in the binding energy calculation, each monomer must be optimized separately ($\Delta E_{Binding} = E_{opt\ complex} - (E_{opt\text{-}monomer\ A} + E_{opt\text{-}monomer\ B}$)

Interaction energy is the contribution to the total energy that is caused by an interaction between the objects being considered. In the interaction energy calculation, each isolated monomer is not allowed to relax into its lowest energy configuration. Thus, the contributions of the relaxation energy will be neglected. Ghost function or ghost atom is used in the interaction energy calculation [6]. Therefore, $\Delta E_{Interaction} = E_{opt\ complex} - (E_{monomer\ AAB} + E_{monomer\ BAB})$

Distortion energy refers to relaxation, deformation or preparation energy and can be calculated as $\Delta E_{Distortion} = \Delta E_{Binding} - \Delta E_{Interaction}$

The Boys and Bernardi counterpoise correction (CP) is a method for removing basis set superposition errors (BSSE), which minimizes the error produced when studying an intermolecular reaction using an incomplete basis set [6]. In the counterpoise correction, the artificial stabilization is countered by letting the separate monomers improve their basis sets *via* borrowing the functions of an empty basis set. Ghost function or ghost atom is used to realize such empty basis set. The ghost function or atom has the basic set of the according monomer or atom, but no electrons to fill it. This procedure corrects the BSSE for both monomer and atom [6]. Previously, the theoretical prediction of palm oil stability using optimized B3LYP/6-31G(d,p) has been reported [7]. It was stated that the different compositions of fatty acid moieties resulted in different physical parameters (bond angles, bond indexed and stabilization energy).

This work aims to study and determine the reason behind the differences in physical properties between chocolate products that use either cocoa butter or palm oil as their filling. This theoretical study mainly focuses on evaluating the binding, interaction, and distortion energies between the main triglycerides [*i.e.* palmitic-oleic-stearic (POS) in cocoa butter *versus* palmitic-oleic-palmitic (POP) in refined, bleached and deodorized (RBD) palm oil] with methylxanthines (caffeine, theobromine, and theophylline) in the cocoa powder. The energy obtained is then used to correlate such differences in the products that use either cocoa butter or palm oil as their fillings. The research results are believed to be one of the factors that contribute toward the rheological behaviour and sensory perception of cocoa products, especially for chocolate.

COMPUTATIONAL METHODS

In this study, all molecular structures and complexes were constructed using the computational software Gauss View 5.0 [8] and optimized using the density functional theory (B3LYP) methods at the 6-31G(d,p) basis set level. The individual structures of triglycerides, polyphenols (epicatechin and procyanidin) and methylxanthines (caffeine, theobromine and theophylline) were firstly constructed and optimized. Their calculation energy was then calculated, followed by the optimization of triglycerides with the polyphenols and methylxanthines complexes and measurement of their binding and interaction energies. The interaction energies for the optimized complexes were corrected for the BSSE using the counterpoise method of Boys and Bernardi [6] *via* the 6-31G(d, p) basis set. All the calculations were carried out using the Gaussian 09 computational package [9]. The binding and interaction energies were next used to correlate the differences found in products that use either cocoa butter or palm oil as their fillings.

RESULTS AND DISCUSSION

Molecular geometries of methylxanthines: The general molecular structure of methylxanthines consist of different functional groups, R_1 and R_2 for caffeine, theobromine and theophylline. The optimized molecular structures in the most stable form are shown in Fig. 1. Density functional theory (DFT) calculations were performed and the calculated optimized structures of all molecules were computed at B3LYP/6-31G(d,p) level of theory. Recently, a work published by Pandohee *et al.* [10] reported the B3LYP/6-31G(d) methods for fatty acids and triglyceride molecule models. The geometrical parameters (bond lengths and bond angles) of all methylxanthines are calculated in order to later optimized with methylxanthines

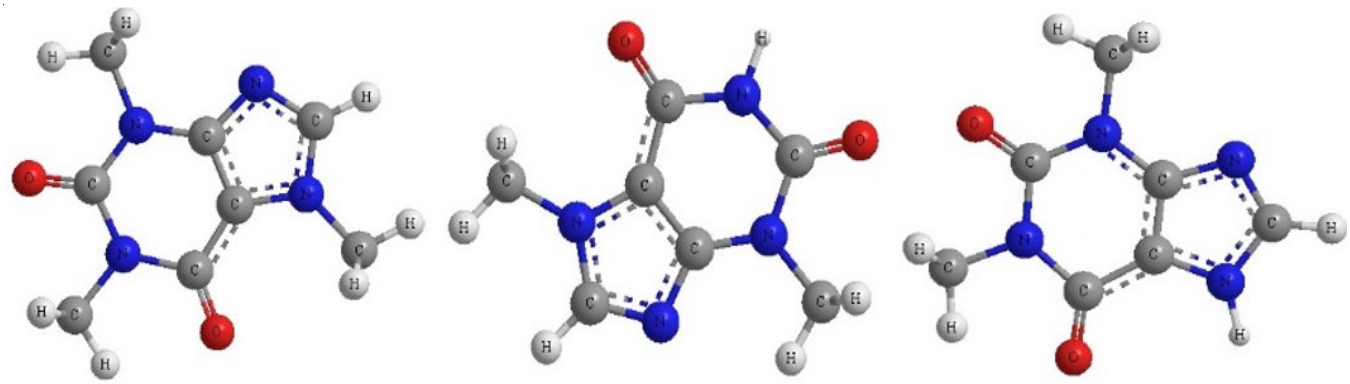

Fig. 1. Optimized structures of caffeine, theobromine and theophylline

complex (Table-1). In general, the calculated bond length and bond angle of caffeine differed from the reference values by 0.001-0.004 Å and 0.003-0.442º, for theobromine by 0.000-0.001 Å and 0.010-0.061º; and for theophylline by 0.000-0.005 Å and 0.021-0.363º. The differences were not significant although the reference values of theophylline used a different basis set, which was B3LYP 6-311G(d,p).

Based on the calculated results, the shortest and strongest bond values for all methylxanthines difference were not significant at the bond C(8)–N(9) with value 1.33 Å. However, the longest and weakest bond lengths between all methylxanthines were found to be different. The weakest bond length shown by caffeine was at the bond C(6)-N(1), while both theobromine and theophylline were at the bond C(5)-C(6). Nevertheless, the weakest bond value for all methylxanthines was not significantly different, which was between 1.42-1.44 Å.

The most linear and strongest bond angle result for all methylxanthines obtained was at the same angle between C(4)-N(9)-C(8) with caffeine, showing the lowest value of 103.5º, while theobromine and theophylline values were the same at 103.8º. This result was due to the lone pair of electrons at the N9 atom that was not part of the aromatic system and extended in the plane of the ring. Although the difference in bond angle value between all methylxanthine was not significant, this value showed that between all methylxanthines, the angle at N9 (nitrogen atom which was not attached to the -CH_3 group) at caffeine was the most unstable angle and had a higher potential to be an electron donor to form hydrogen bonding with the triglyceride molecules as compared to theobromine and theophylline.

Methylxanthine molecules offered several possible donor sites to form hydrogen bonding with the triglyceride molecules.

TABLE-1
GEOMETRICAL PARAMETERS [BOND LENGTHS (Å), BOND ANGLES (°)] FOR CAFFEINE, THEOBROMINE AND THEOPHYLLINE

Bond lengths (Å)	Isolated		Complex with TAG POS	Complex with TAG POP	Bond angle (°)	Isolated		Complex with TAG POS	Complex with TAG POP
	Calculated B3LYP 6-31G (d,p)	Ref.[a] B3LYP 6-31G (d,p)	Calculated B3LYP 6-31G (d,p)	Calculated B3LYP 6-31G (d,p)		Calculated B3LYP 6-31G (d,p)	Ref.[a] B3LYP 6-31G (d,p)	Calculated B3LYP 6-31G (d,p)	Calculated B3LYP 6-31G (d,p)
Caffeine									
C(2)–N(1)	1.4090	1.4080	1.4075	1.4077	C(2)–N(3)–C(4)	119.600	119.700	119.633	119.658
C(2)–N(3)	1.3960	1.3920	1.3936	1.3932	N(3)–C(4)–C(5)	121.700	121.300	121.426	121.459
C(4)–C(5)	1.3810	1.3810	1.3815	1.3811	C(4)–C(5)–C(6)	123.700	123.900	123.844	123.812
C(4)–N(3)	1.3750	1.3750	1.3758	1.3751	C(5)–C(6)–N(1)	111.000	111.000	111.033	111.028
C(4)–N(9)	1.3600	1.3590	1.3603	1.3597	C(2)–N(1)–C(6)	127.200	126.900	126.899	126.901
C(5)–C(6)	1.4330	1.4330	1.4328	1.4334	C(4)–C(5)–N(7)	105.100	105.100	105.124	105.167
C(5)–N(7)	1.3870	1.3870	1.3880	1.3880	C(5)–C(4)–N(9)	111.900	111.800	111.641	111.640
C(6)–N(1)	1.4180	1.4170	1.4181	1.4178	C(4)–N(9)–C(8)	103.500	103.600	103.889	103.866
C(8)–N(7)	1.3570	1.3560	1.3543	1.3549	N(7)–C(8)–N(9)	113.800	113.800	113.354	113.491
C(8)–N(9)	1.3310	1.3300	1.3343	1.3321	C(5)–N(7)–C(8)	105.700	105.700	105.991	105.837
					N(1)–C(2)–N(3)	116.800	117.200	117.149	117.134
Theobromine									
C(2)–N(1)	1.4010	1.4000	1.4005	1.4011	C(2)–N(3)–C(4)	119.600	119.600	119.538	119.372
C(2)–N(3)	1.3910	1.3910	1.3926	1.3950	N(3)–C(4)–C(5)	122.000	122.100	122.209	122.569
C(4)–C(5)	1.3850	1.3840	1.3845	1.3842	C(4)–C(5)–C(6)	123.400	123.400	123.270	123.096
C(4)–N(3)	1.3800	1.3790	1.3790	1.3779	C(5)–C(6)–N(1)	109.600	109.600	109.623	109.531
C(4)–N(9)	1.3590	1.3590	1.3592	1.3599	C(2)–N(1)–C(6)	129.900	129.900	129.949	130.239
C(5)–C(6)	1.4360	1.4350	1.4363	1.4362	C(4)–C(5)–N(7)	105.200	105.200	105.214	105.269
C(5)–N(7)	1.3860	1.3860	1.3867	1.3857	C(5)–C(4)–N(9)	111.600	111.600	111.422	111.429
C(6)–N(1)	1.4080	1.4080	1.4082	1.4081	C(4)–N(9)–C(8)	103.800	103.800	104.032	103.948
C(8)–N(7)	1.3560	1.3560	1.3546	1.3551	N(7)–C(8)–N(9)	113.700	113.700	113.434	113.525
C(8)–N(9)	1.3300	1.3300	1.3321	1.3314	C(5)–N(7)–C(8)	105.700	105.700	105.897	105.829
					N(1)–C(2)–N(3)	115.400	115.400	115.402	115.188
Theophylline									
C(2)–N(1)	1.4010	1.4000	1.4005	1.4011	C(2)–N(3)–C(4)	119.600	119.600	119.538	119.372
C(2)–N(3)	1.3910	1.3910	1.3926	1.3950	N(3)–C(4)–C(5)	122.000	122.100	122.209	122.569
C(4)–C(5)	1.3850	1.3840	1.3845	1.3842	C(4)–C(5)–C(6)	123.400	123.400	123.270	123.096
C(4)–N(3)	1.3800	1.3790	1.3790	1.3779	C(5)–C(6)–N(1)	109.600	109.600	109.623	109.531
C(4)–N(9)	1.3590	1.3590	1.3592	1.3599	C(2)–N(1)–C(6)	129.900	129.900	129.949	130.239
C(5)–C(6)	1.4360	1.4350	1.4363	1.4362	C(4)–C(5)–N(7)	105.200	105.200	105.214	105.269
C(5)–N(7)	1.3860	1.3860	1.3867	1.3857	C(5)–C(4)–N(9)	111.600	111.600	111.422	111.429
C(6)–N(1)	1.4080	1.4080	1.4082	1.4081	C(4)–N(9)–C(8)	103.800	103.800	104.032	103.948
C(8)–N(7)	1.3560	1.3560	1.3546	1.3551	N(7)–C(8)–N(9)	113.700	113.700	113.434	113.525
C(8)–N(9)	1.3300	1.3300	1.3321	1.3314	C(5)–N(7)–C(8)	105.700	105.700	105.897	105.829
					N(1)–C(2)–N(3)	115.400	115.400	115.402	115.188

TAG = Triglyceride

The most basic nitrogen in methylxanthines was nitrogen atom, which was not attached to the -CH_3 group. This nitrogen was basic nitrogen because the lone pair was in the sp^2 hybrid orbital pointing away from the ring system; therefore, it was able to act as a proton acceptor. Nitrogen atom attached to -CH_3 group was not basic nitrogen because the lone pair was in the unhybridized *p*-orbital and part of the aromatic system. Therefore, it was unable to act as a proton acceptor. It was observed that the complexes may be formed through C-H···O and C-H···N type hydrogen bonds. This is supported by Karthika *et al.* [11] by studying the hydrogen bonding between caffeine-theophylline complexes. The optimized methylxanthine molecules interacted at hydrogen at β-position of the triacylglycerols. It was observed that when both molecules interacted at this position, the highest number of hydrogen bonding might be formed through C-H···O and C-H···N type hydrogen bonds.

The calculated results showed that the shortest and strongest bond length values at the bond C(8)–N(9) for all methylxanthines increased after the formation of complex with both triglycerides. This result showed that the bond became weaker due to the lone pair at N9 atom that was donated due to the interaction with both triglycerides. However, the bond length value at bond C(8)–N(9) for caffeine showed the highest difference after the interaction with triglyceride POS as compared to other methylxanthines. This result proved that the complex between triglyceride POS and caffeine had the strongest interaction.

The highest bond angle value at the angle C(4)-N(9)-C(8) for all methylxanthines increased after the formation of complex with both triglycerides. This result proved that the bond became more stable due to the electronegativity of N9 atom that decreased during the formation of the hydrogen bond. Consequently, this caused the repulsion between the bond pairs to increase and therefore, the bond angle also increased. However, the bond angle value at the bond C(4)-N(9)-C(8) for caffeine showed the highest difference after the interaction with triglyceride POS as compared to other methylxanthines. This result suggested that the complex between triglyceride POS and caffeine had the strongest interaction.

Molecular geometries of triglyceride during complexation with methylxanthines: Fig. 2 shows the complexation of triglyceride with methylxanthines. The distances and angles of hydrogen bonds could also be considered as a character of hydrogen bonding strength. The geometrical parameters of the intermolecular hydrogen bond forms in all complexes are presented in Table-2. Hydrogen bond strength and length for the optimized complex between triglyceride-methylxanthines showed that all optimized complexes had the strongest hydrogen bonding at C-H···N position with hydrogen at β-position of triglyceride as a proton donor and basic nitrogen of methylxanthines as a proton acceptor. The hydrogen bond strength values varied from 10.04 to 21.25 kJ/mol, while the bond length values varied from 2.08 to 2.56 Å. The strongest hydrogen bond strength was found for the complex between triglyceride and theophylline with hydrogen bond strength of 20.13 to 21.25 kJ/mol and hydrogen bond lengths of 2.43 and 2.41 Å. However, for the optimized triglyceride POS-caffeine, it was observed that there were two strong hydrogen bonds at the C-H···N and C-H···O positions. For hydrogen-bond at C-H···O position, C-H of caffeine acted as the proton donor while oxygen of triglyceride POS acted as the proton acceptor with bond strength 10.17 kJ/mol and bond length 2.33 Å. Similarly, stability of the caffeine-theophylline complex was strongly influenced by hydrogen bonding interactions, especially when additional hydrogen bonds were formed (C-H···O) [11].

The calculated bond angle for the optimized complexes as presented in Table-2 indicates that hydrogen bond was almost linear. According to Nagy [12], the closer the angle was to 180º, the stronger the hydrogen bond. In present study, it was observed that the intermolecular angle for C-H···O in the triglyceride-caffeine and triglyceride-theobromine was stronger than the intermolecular angle for C-H···N. The intermolecular

TABLE-2
THEORETICAL OPTIMIZED GEOMETRICAL STRUCTURE PARAMETERS [BOND LENGTHS (Å), BOND ANGLES (°)] FOR COMPLEX CALCULATED AT B3LYP/6-31G (d,p) LEVEL OF THEORY

Complex	Hydrogen bond	Bond lengths (Å)	Bond angle (°)	Total strength of hydrogen bonding (KJ/mol)
Triglyceride POS-Caffeine	C28–H29···N189	2.56358	156.851	10.04
	C180–H181···O65	2.69631	161.287	4.23
	C170–H171···O41	2.32646	130.562	10.17
Triglyceride POP-Caffeine	C28–H29···N183	2.50453	134.047	10.13
	C174–H175···O10	2.95895	163.094	1.59
	C164–H165···O65	2.63478	125.82	3.39
Triglyceride POS-Theobromine	C28–H29···N175	2.50269	134.165	10.04
	C176–H179···O10	2.93437	161.850	1.80
	C170–H171···O65	2.6457	125.305	3.05
Triglyceride POP-Theobromine	C28–H29···N169	2.46944	141.293	13.56
	C164–H165···O10	2.88198	122.286	1.21
	C170–H173···O65	2.65465	162.321	4.90
Triglyceride POS-Theophylline	C49–H50···N19	2.42607	175.939	20.13
	C13–H15···O86	2.77881	119.599	1.63
	C13–H14···O63	2.7314	155.085	3.56
Triglyceride POP-Theophylline	C28–H29···N178	2.40852	177.184	21.25
	C172–H174···O65	2.80647	118.854	1.46
	C172–H173···O42	2.73681	155.768	3.51

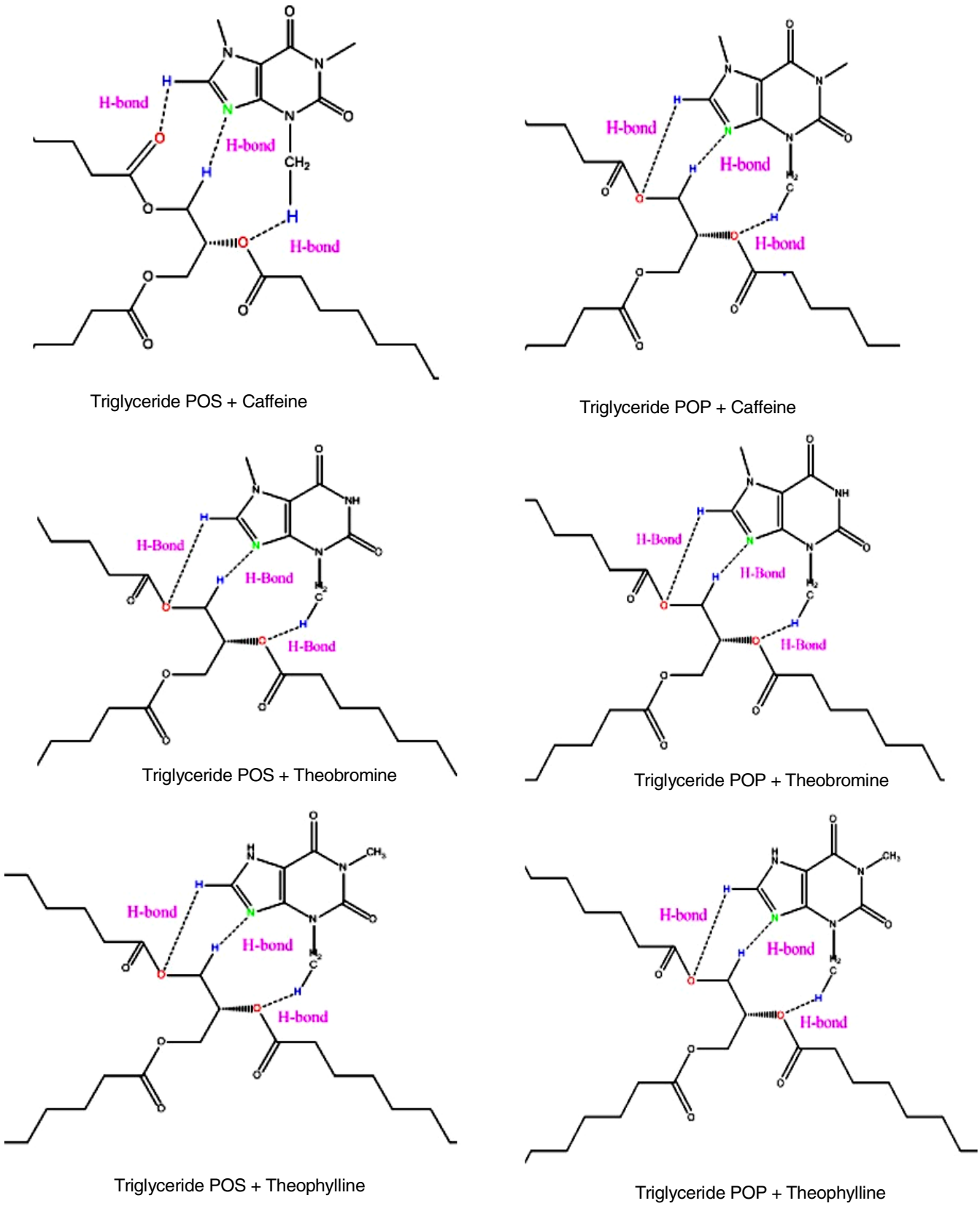

Fig. 2. Optimized structures of triglyceride-methylxanthines complexes

angle for C-H···O in the triglyceride-caffeine and triglyceride-theobromine was in the range between 122.3º and 163.1º. The intermolecular angle for C-H···N in triglyceride-caffeine and triglyceride-theobromine was in the range of 134.0 to 156.9º. However, the intermolecular bond for triglyceride-theophylline complex was observed to be stronger at their C-H···N bond

(175.9-177.2º) as compared to the C-H···O bond (118.9-155.8º). This value paralleled with the strongest hydrogen bond strength and bond length obtained for the triglyceride-theophylline complex.

Donor acceptor analysis of triglyceride during complexation with methylxanthines: The donor-acceptor analysis for complexes at B3LYP/6-31G(d,p) level of theory was calculated. Based on the donor-acceptor analysis conducted, it was found that the strongest hydrogen bond strength between triglyceride-methylxanthines complex was largely contributed by the lone pair electron of nitrogen atom from the methylxanthines, which acted as the electron donor. This was also contributed by hydrogen at the β-position of triglyceride, which acted as the electron acceptor. However, it was also found another strong hydrogen bond between triglyceride POS-caffeine complex at the C=O position, which was contributed by the lone pair electron from the oxygen atom of triglyceride POS and hydrogen of caffeine acted as the electron acceptor. Compared to other triglyceride-methylxanthine interactions, hydrogen bonding was formed at the C-H···O position, not at C=O position. This is the reason why the binding and interaction energies of triglyceride POS-caffeine were higher as compared to other triglyceride-methylxanthine complexes [11, 13].

Binding energy of triglyceride during complexation with methylxanthines and counterpoise corrected interaction energy of triglyceride during complexation with methylxanthines: The calculated binding energies are summarized in Table-3. While, the interaction energy of all the complexes is calculated after correcting the BSSE by the full counterpoise procedure (CP) of Boys and Bernardi equation [6]. Based on the data obtained, both the binding energy and the interaction energy between caffeine and triglyceride POS was almost two times higher as compared to the binding with triglyceride POP. It is also found that the same pattern observed with the distortion energy of the complexes. Distortion energy between caffeine and triglyceride POS was almost two times higher as compared to the distortion energy with triglyceride POP. Distortion energy refers to relaxation, deformation or preparation energy [14]. These findings were supported by the hydrogen bond strength, bond length and bond angle results obtained, which showed that there were two strong hydrogen bonds at C-H···N and C-H···O positions for the binding between triglyceride POS-caffeine. The binding energy between other two methylxanthines (theobromine and theophylline) in cocoa powder and both triglycerides obtained were almost of the same magnitude.

The binding energy obtained between theobromine and both triglycerides (POS and POP) was between 13.6 and 13.7 kJ/mol, while the binding energy obtained between theophylline and both triglycerides was between 14.1 and 14.5 kJ/mol. Additionally, the interaction energy obtained between theobromine and both triglycerides (POS and POP) was between 3.42 and 3.45 kJ/mol, while it was between 3.70 and 3.72 kJ/mol obtained between theophylline and both triglycerides. Therefore, this study theoretically proved that the binding energy between the caffeine in cocoa with the main triglyceride in cocoa butter and RBD palm kernel oil might be one of the factors explaining why chocolate made from cocoa butter has excellent quality and sensory properties as compared to the chocolate made from RBD palm kernel oil. This is because the higher the binding energy, the higher the stabilization of the complex formed. Previously, a study reported the structure and hydrogen bond interactions of caffeine and theophylline studied using density functional theory (DFT) and Møller Plesset perturbation theory

TABLE-3
BINDING ENERGY, COUNTERPOISE CORRECTED INTERACTION ENERGY AND DISTORTION ENERGY FOR COMPLEX CALCULATED AT B3LYP/6-31G (d,p) LEVEL OF THEORY

Complex	Total electronic energy (a.u.)	Electronic energy of molecule A (a.u.)	Electronic energy of molecule B (a.u.)	Electronic energy of molecule A with ghost function (a.u.)	Electronic Energy of molecule B with ghost function (a.u.)	Binding energy (KJ/mol)	BSSE energy (KJ/mol)	Counterpoise corrected interaction energy (KJ/mol)	Distortion energy (KJ/mol)
	$E_{complex}$	EA^{A}	EB^{B}	EA^{AB}	EB^{AB}	$\Delta E_{Binding}$	BSSE	$\Delta E_{Interaction}$	$\Delta E_{Distortion}$
TAG POS (H-β)-Caffeine	-3290.5157	-680.3906	-2610.1156	-680.3930	-2610.1186	-25.1054	14.0640	-10.7349	14.3705
TAG POP (H-β)-Caffeine	-3211.8783	-680.3906	-2531.4824	-680.3926	-2531.4844	-13.7978	11.1431	-3.3788	10.4189
TAG POS (H-β)-Theobromine	-3251.1998	-641.0790	-2610.1156	-641.0809	-2610.1176	-13.6903	11.1485	-3.4470	10.2433
TAG POP (H-β)-Theobromine	-3172.5666	-641.0790	-2531.4824	-641.0809	-2531.4844	-13.5662	10.9618	-3.4191	10.1472
TAG POS (H-β)-Theophylline	-3251.1965	-641.0756	-2610.1156	-641.0772	-2610.1179	-14.0633	10.6663	-3.7186	10.3447
TAG POP (H-β)-Theophylline	-3172.5636	-641.0756	-2531.4824	-641.0772	-2531.4849	-14.4804	10.7313	-3.6983	10.7820

Molecule A = Caffeine, Theobromine, Theophylline; Molecule B = TAG POS, TAG POP; TAG = Triglyceride

(MP2) [15]. They revealed that the strong bonds resulted from the highest interaction energy of the compounds.

Based on all the results obtained, it is interesting to note that hydrogen bond length and angle correlated well with the binding energies. Complex triglyceride POS-caffeine had two higher and almost linear bond angles, which showed higher binding energy as compared to the other two methylxanthines. triglyceride-theophylline with stronger bond length and angle had higher binding energy as compared to the complex triglyceride-theobromine. Triglyceride-procyanidin with the bond angle nearer to linear had a higher binding energy as compared to triglyceride-epicatechin.

Molecular electrostatic potential (MEP) of complex triglyceride POS and triglyceride POP with caffeine: Fig. 3a shows the MEP diagram of methylxanthines (caffeine, theobromine and theophylline) After the formation of a complex between triglyceride POS and caffeine, it can be observed that the areas of a nitrogen atom (not attached to the CH_3 group) on caffeine initially tended to show yellow and red tones changing to color tones (tended toward green). This change in color tones showed that nitrogen in caffeine was initially the region rich with electrons and indicated negative values of electrostatic potential, which donated its electrons to the electron-poor hydrogen beta on the triglyceride due to the hydrogen bond formation (Fig. 3b). Additionally, the same finding was observed for complex triglyceride POP with caffeine.

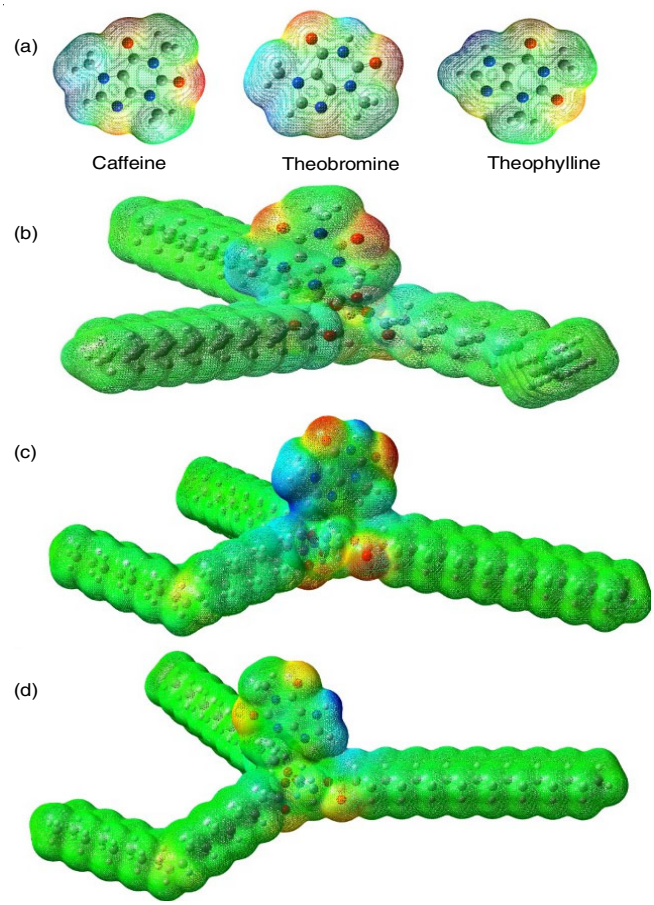

Fig. 3. MEP image for (a) triglyceride POS-caffeine complex (b) triglyceride POS-theobromine complex and (c) triglyceride POS-theophylline

For triglyceride POS and caffeine complex, it can be observed that oxygen atom (focusing on the area of C=O) on triglyceride POS initially tended to show yellow and red changing to color tones (tended toward green) after the formation of the complex. Additionally, another oxygen atom on triglyceride POS (C-O) that initially tended to yellow also changed to the color tones (tended toward green) after the formation of the complex. On the contrary, the triglyceride POP with caffeine complex showed that two oxygen atoms (C-O) on triglyceride POP that initially tended to show yellow had changed to the cooler tones of green after the formation of this complex (figure not shown).

The color changes showed that oxygen atoms were initially regions rich with electrons and showed negative values of electrostatic potential, which donated their electron to the electron-poor hydrogen on the caffeine due to the hydrogen bonds formation.

The MEP result obtained was parallel with the donor-acceptor analysis result, which showed that two strong hydrogen bonds were formed between TAG POS and caffeine at N183–H29 and O41–H171. Based on MEP, it proved that the color at both regions N183–H29 (10.04 KJ/mol) and O41–H171 (C=O, 10.17 KJ/mol) tended more toward the green tones, which meant that strong hydrogen bonding was formed. The MEP result also proved the donor-acceptor analysis that hydrogen bonding formed at O65–H181 (C-O, 4.23 KJ/mol) was a weak hydrogen bond. This is because although the color in this region tended to show green, the color appeared as yellowish-green, which proved that the hydrogen bond formed in this region was weak.

Meanwhile, the MEP result obtained for triglyceride POP with caffeine showed the formation of a strong hydrogen bond between N183–H29 (10.13 KJ/mol). Similarly, the MEP result also proved the donor-acceptor analysis of the formation of weak hydrogen bonds at O10–H175 (C-O, 1.59 KJ/mol) and O65–H165 (C-O, 3.39 KJ/mol).

Molecular electrostatic potential (MEP) of complex triglyceride POS and triglyceride POP with theobromine: After the formation of respective complexes between triglyceride POS and triglyceride POP with theobromine, it can be observed that the nitrogen atom (not attached to the CH_3 group) on theobromine initially tended to yellow and red before changing to color tones (tended toward green). This atom that was rich with electrons showed negative values of electrostatic potential; therefore, it is a potential atom to become an electron donor. Molecular electrostatic potential (MEP) is used to determine the electronic distribution and other molecular charge distribution of a molecule or complexes [16].

This color change showed that nitrogen on theobromine was initially a region rich with electrons and showed negative values of electrostatic potential, which donated electrons to the electron-poor hydrogen at β-position on triglyceride POS and triglyceride POP due to the hydrogen bond formation (Fig. 3c). Besides, it can be observed that two oxygen atoms (C-O) on both complexes initially tended to yellow and then changed to color tones (green) after the formation of the complex. This color change showed that the oxygen atoms in triglyceride

POS and triglyceride POP were initially regions rich with electrons and showed negative values of electrostatic potential, in which they donated electrons to the electron-poor hydrogen on the theobromine due to the hydrogen bond formation.

The MEP result obtained was parallel with the donor-acceptor analysis result, which showed only one strong hydrogen bonding was formed between triglyceride POS and theobromine at N175–H29. Based on MEP, it was proven that the color at region N183–H29 (10.04 KJ/mol) showed tendency toward the green tones, which meant that strong hydrogen bonding was formed. The MEP result also ascertained the donor-acceptor analysis that hydrogen bonds formed at O10–H179 (C-O, 1.80 KJ/mol) and O65–H171 (C-O, 3.05 KJ/mol) were weak hydrogen bonds. Although the color in this region tended toward green, the color appeared as yellowish-green, which proved that the hydrogen bond formed in this region was weak. Furthermore, for triglyceride POP, the MEP result obtained was also parallel with the donor-acceptor analysis result, which showed that only one strong hydrogen bonding was formed between triglyceride POP and theobromine at N169–H29 (13.56 KJ/mol) and showed tendency toward the green tones. Similar to triglyceride POS, the donor-acceptor analysis confirmed that hydrogen bonding formed at O10–H165 (C-O, 1.21 KJ/mol) and O65–H173 (C-O, 4.90 KJ/mol) consisted of weak hydrogen bonds.

Molecular electrostatic potential (MEP) of complex triglyceride POS and triglyceride POP with theophylline: After the formation of the complex between triglyceride POS and theophylline, it can be observed that a strong hydrogen bonding was formed between triglyceride POS and theophylline at N19–H50. Meanwhile, for the complex of triglyceride POP with theophylline, strong hydrogen bonding was formed between N178–H29. Based on MEP, it was proven that both colors at regions N19–H50 (20.13 KJ/mol) and N178–H29 (21.25 kJ/mol) showed tendency toward the green tones, which attributed to the strong hydrogen bonding. The MEP result also proved the donor-acceptor analysis that weak hydrogen bonds were formed at O63–H14 (C-O, 3.56 KJ/mol) and O86–H15 (C-O, 1.63 KJ/mol) for triglyceride POS, as well as O65–H174 (C-O, 3.51 kJ/mol) and O42–H173 (C-O, 1.46 kJ/mol) for triglyceride POP complexes (color appeared as yellowish-green) (Fig. 3d).

Conclusion

Binding and interaction energies between triglyceride POS with caffeine play an important role that contributes toward the excellent quality and sensory properties of chocolate made from cocoa butter as compared to chocolate made from refined, bleached and deodorized (RBD) palm oil. Binding and interaction energies results showed that the presence of methylxanthine caffeine was one of the main contributors why chocolate made from cocoa butter has excellent quality and sensory properties as compared to the chocolate made from RBD palm oil. The donor-acceptor analysis showed that the hydrogen bond strength result obtained was proven using MEP and the result was parallel. Molecular electrostatic potential (MEP) showed the region with stronger hydrogen bonds tended toward green color, while weaker hydrogen bond tended toward yellow or red color. The results were believed to be one of the factors that contributed toward the rheological behaviour and sensory perception of cocoa products, especially chocolate.

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CONFLICT OF INTEREST

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

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