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Effects of Mycobacterium Tuberculosis Macrophage Interaction: Surface Energetic Mechanism

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Abstract— The mechanisms effects of mtb – macrophage using surface energetic in determining the interaction processes with the surface interfacial energies explained using van der Waals concept of particle – particle interactions. The Lifshitz derivations for van der Waals forces were applied as an alternative to the contact angle approach which has been widely used in other biological systems. The methodology involved taking sputum samples from twenty infected persons and from twenty uninfected persons for absorbance measurement using a digital Ultraviolet visible Spectrophotometer. Matlab software tools were used in the mathematical analysis of the data generated from the absorbance values. The values of A_{132abs} = $0.21631x10^{-21}$Joule (for mtb infected sputum) and $\tilde{A}_{132abs}$ = $0.18825x10^{-21}$Joule (for mtb/HIV infected sputum) were obtained. The free energies of adhesion calculated were found to be negative with combined Hamaker coefficient positive. The implication of this result is the positive value of the absolute combined Hamaker coefficient which entails net positive van der waals forces demonstrating an attraction between mtb and the macrophage. This however, implies that infection is very likely to occur. It was also shown that in the presence of HIV, the interaction energy is reduced by 13% confirming adverse effects observed in HIV patients suffering from tb. Negative Hamaker coefficient ($-0.22669x10^{-19}mJ/m^2$) indicated that separation of mtb is practical. The condition was sought for repulsion to occur and that condition was based on the value of A_{33} that would render the absolute

combined Hamaker coefficient A_{131abs} negative. Mathematically it was derived as $A_{33} \geq 0.9527 \times 10^{-21}$Joule which satisfies this condition for negative A_{132abs}. To achieve the condition of A_{33} above, possible additive(s) in form of drugs to the sputum should be required.

Keywords- Absorbance; surface energetics; hamaker coefficient; macrophage; mycobacterium tuberculosis; van der waals forces; wavelength.

I. Introduction

Tuberculosis, tb, is an airborne disease caused by the bacterium mycobacterium tuberculosis, mtb. This causative agent of tb is one of the world's most destructive human pathogens. The infectious bacteria is transmitted through air and most commonly affects the lungs, but can also attack other parts of the body, such as the brain, spine or kidneys, which is responsible for more than 75% of cases (Aandahl, 2012).

The World Health Organization, WHO, declared tb as a global emergency in 1993. Unfortunately, the efforts made by the discontinue tb strategy were not enough to impede the occurrence of 1.3 million deaths in 2009 (WHO, 2010). However, WHO estimates that the number of cases per capita peaked at 2004 and is slowly falling (WHO, 2012). Nonetheless, the battle against tb is far from being over, since mtb (the main causative agent of tb) proved to be highly adaptive (Kumar and Rao, 2011) and capable of evading the current strategies for treatment of about half million cases of multi-drug-resistant tb (MDR-tb) that were reported in 2007, including cases of extensively drug-resistant tb (XDR-tb) (WHO, 2012), and the more recently reported totally drug-resistant strains (TDR-tb) (Kumar and Rao, 2011; Velayati et al, 2009).
Several researchers so far have reported the occurrences of tb cases, a fussy report surveyed that only 35 out of 134 countries showed declination of cases of about 5% per year based on per capita rate (Lonnroth et al, 2009). This review considered the data from 1998 – 2007. Different surveillance analysis and mathematical modeling studies recommended reduction of tb incidences per capita is around 1% per year, further suggesting diminution of cases by 2015 (Adeeb et al, 2013). The world population growth has being predicted approximately 2% per year; this may be an important reason for increment of tb cases (WHO, 2012). All these preceding reports showing the presence of lacunae in the existing management approaches for tb and the inadequate effectiveness of public health systems, with particular reference to underdeveloped countries. In malice of the availability of anti-tb drugs developed over the last five decades, one-third of the world's population retains a dormant or latent form of mtb (Corbett et al, 2003).

Much research has been and is still on, on the subject with a cure not yet in view. The role of surface properties in this biological process will be established. Surface energetic approach is a very important thermodynamics tool used in determining the absolute and combined Hamaker coefficients of mtb – macrophage interaction

processes (Chukwuneke et al, 2017). The Lifshiftz derivation for van der Waals forces is used to model the interaction and the surface energies and surface free energies of adhesion obtained by this method will be used to predict the nature of surface interactions between two solid particles suspended in a liquid medium.

It is a well-known fact that surface property determination of interacting particles lead to the further understanding of the mechanism of interactions. A common area of contact is established once two particles meet each other. In such process, a certain portion of each particle gets displaced through work (Chukwuneke et al, 2016). The work responsible for the displacement of a unit area is known as surface free energy. The consecutive impact on the surface is known as surface energetic effects. To attain the equilibrium such impacts are changed in a slow pace. In this particular study similar concepts have been implemented to characterize the mtb – macrophage interactions with the sputum as the intervening medium.

In this paper, mtb is conceptualized as particle dispersed in a liquid medium "sputum" and interacting with another particle "macrophage". The bacterium attaches itself on the surface of the macrophage cell before penetrating and attacking it. If the surface of the macrophage cell is such that it will repel the bacteria, access of the bacteria into the alveoli of the cell would have been denied. Thus the initial actions actually take place on the surface of the cell and of the bacteria. It is alongside this background that this paper surveys a novel and exceptional approach using interfacial free energy approach to search for a way forward in the research on the topic of mtb – macrophage interaction mechanisms.

II. Methodology

The methodology of this paper involved sputum sample collection, mycobacterium and macrophages structural studies, mtb screening, and the study of the mechanism of interactions of the bacterium and the macrophage. Twenty samples each of infected, uninfected and *mtb*/HIV co-infected sputum were collected. Each specimen was screened to determine the infection status using GeneXpert and Ziehl-Neelsen staining method. The slides were prepared and the absorbance, ā, values of each specimen, for wavelength range of 230–950nm were measured using digital Ultraviolet Visible Spectrophotometer. The data generated and the various equations governing the relationship among the variables were used in calculating values for the reflectance, R, transmittance, T, refractive index, n, and the dielectric constant, ε. MatLab software tools were employed in the mathematical analysis of the data generated from the absorbance values.

III. Thermodynamic Mechanism of Interactions

Suppose in a certain case, the bacterium, *mtb* has been considered as a particle coming in the surrounding area of the macrophage which is further considered as another particle. In the consecutive step, the bacterium attaches itself on the surface of the macrophage under a specific condition where the macrophage is dispersed in sputum medium (see figure 1).

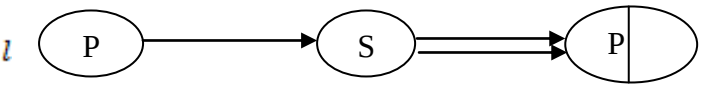

Figure 1: Conceptual representation of *mtb* – macrophage adhesion process

Expression of the thermodynamic free energy associated with adhesion process provided in figure (1) is represented as follows (Omenyi, 1978; Visser, 1981):

$$\Delta F_{pls}^{adh}(d_o) = \gamma_{ps} - \gamma_{pl} - \gamma_{sl} \qquad (1)$$

Equation (1) represents the integrated free energy of adhesion considered from infinity to the equilibrium where separation distance is d_o. In this equation, P, S represents bacteria and macrophage respectively and L signifies the sputum medium. In the right hand side of the equation, γ_{ps} expresses the interfacial free energy between P and S. γ_{pl} represents the same for P and L, whereas, γ_{sl} provides the same information between S and L.

Successful bacterial penetration in the macrophage cells will occur through the engulfment of the bacteria requiring net free energy, represented as (Omenyi, 1978; Chukwuneke et al, 2015):

$$\Delta F_{NET} = \gamma_{ps} - \gamma_{pl} < 0 \qquad (2)$$

ΔF^{adh} can be determined by several approaches, apart from the above surface free energy approach. The classical work of (Hamaker, 1937) is very appropriate.

A system containing two planes could be considered for computing the free energy of interaction (Hamaker, 1937):

$$\Delta F_{132}(d_o) = \left[-\frac{A_{132}}{12\pi d_o^2}\right] \qquad (3)$$

In this, A_{132} refers to the Hamaker coefficient for a respective system.

Considering nominal isolation distance d_0, and equation (3) as valid for such a small distance, the Hamaker coefficient should be expressed:

$$A_{132} = -12\pi d_o^2 \Delta F^{adh}(d_o) \qquad (4)$$

Following Lifshitz theory, the Hamaker coefficient A_{132} is represented as follows (Lifshitz, 1961):

$$A_{ikj} = \frac{3}{4}\pi\hbar\int_0^{\infty}\left[\frac{\varepsilon_i(i\zeta)-\varepsilon_k(i\zeta)}{\varepsilon_i(i\zeta)+\varepsilon_k(i\zeta)}\right]\left[\frac{\varepsilon_j(i\zeta)-\varepsilon_k(i\zeta)}{\varepsilon_j(i\zeta)+\varepsilon_k(i\zeta)}\right]d\zeta \qquad (5)$$

Where, $\varepsilon_j(i\zeta)$ refers to the dielectric constant of a specific material j, this is considered through the imaginary i, frequency axis, $(i\zeta)$, $\hbar$ is planck's constant.

In this context, the evaluation of equation (5) should result in equivalent value with the thermodynamic free energy of adhesion, provided in equation (1).

Thus, the Hamaker coefficient and the interfacial free energies are connected through the following equation:

$$A_{pls} = -12\pi d_o^2(\gamma_{ps} - \gamma_{pl} - \gamma_{sl}) \qquad (6)$$

This equation has been derived through combining equation (4) with equation (1). For the issue of self-interaction of a particle equation (7) should be considered:

$$A_{ij} = \frac{3}{4}\pi\hbar\int_0^{\infty}\left[\frac{\varepsilon_i(i\zeta)-\varepsilon_j(i\zeta)}{\varepsilon_i(i\zeta)+\varepsilon_j(i\zeta)}\right]^2 d\zeta \qquad (7)$$

To determine the Hamaker coefficients using the Lifshitz theorem of equation (7), there is a need to evaluate the dielectric constant ε. From the information of

light absorbance, reflection and transmittance, it could be seen that:

$\bar{a} + T + R = 1$ (8)

$T = \exp^{-\bar{a}}$ (9)

Where; $\bar{a}$ is absorbance, T is transmittance, and R is reflectance.

A value for the refractive index, *n* is obtained by employing the mathematical relation (Robinson, 1952):

$$n = \left[\frac{1 - R^{1/2}}{1 + R^{1/2}}\right] \quad (10)$$

A value for the extinction coefficient, *k* is obtained using:

$$k = \left[\frac{\alpha\lambda \times 10^{-9}}{4\pi}\right] \quad (11)$$

Where; α is the absorption coefficient defined as follows:

$$\alpha = \left[\frac{\bar{a}}{\lambda \times 10^{-9}}\right] \quad (12)$$

The dielectric constant, ε could thus be given by (Charles, 1996);

For the real part:

$$\varepsilon_1 = n_1^2 - k^2 \quad (13)$$

For the imaginary part:

$$\varepsilon_2 = 2n_2k \quad (14)$$

With these values, it is possible to determine A_{ij} using the relevant equations to determine A_{11}:

$$A_{11} = 2.5\left[\frac{\varepsilon_{10} - 1}{\varepsilon_{10} + 1}\right]^2 = 2.5\left[\frac{n_1^{\,2} - 1}{n_1^{\,2} + 1}\right]^2 \quad (15)$$

This gives a value to the Hamaker constant A_{11}, and by extension to other Hamaker constants A_{22} and A_{33}. For combination of two different materials 1 and 2 in approximation:

$$A_{12} = \sqrt{A_{11}A_{22}} \quad (16)$$

For a combination of two disimilar materials (i.e. macrophage, 1 and the bacteria, 2) with the gap between 1 and 2 filled with sputum as the medium 3, the combined Hamaker coefficient will be given by:

$$A_{132} = A_{12} + A_{33} - A_{13} - A_{23} \quad (17)$$

$$A_{131} = A_{11} + A_{33} - 2A_{13} \quad (18)$$

Therefore, the system under consideration follows equations (17) and (18):

Rewriting these equations will give:

$$A_{132} = \left(\sqrt{A_{11}} - \sqrt{A_{33}}\right)\left(\sqrt{A_{22}} - \sqrt{A_{33}}\right) \quad (19)$$

$$A_{131} = \left(\sqrt{A_{11}} - \sqrt{A_{33}}\right)^2 \quad (20)$$

Equation (20) shows that, for a three-component system involving three different materials, 1, 2 and 3, A_{132} can become negative:

$$A_{132} < 0 \quad (21)$$

When;

$$\sqrt{A_{11}} > \sqrt{A_{33}}$$

and $\sqrt{A_{22}} < \sqrt{A_{33}}$ (22)

A_{33} = Hamaker constant for sputum; A_{13} = Hamaker constant between both materials (i.e. *mtb* and sputum); A_{23} = Hamaker constant between both materials (i.e. macrophage and sputum).

Thus the Hamaker Coefficient A_{132} becomes:

$$A_{132} = \frac{3}{4}\pi\hbar\int_0^{\infty}\left[\frac{\varepsilon_1(i\zeta) - \varepsilon_3(i\zeta)}{\varepsilon_1(i\zeta) + \varepsilon_3(i\zeta)}\right]\left[\frac{\varepsilon_2(i\zeta) - \varepsilon_3(i\zeta)}{\varepsilon_2(i\zeta) + \varepsilon_3(i\zeta)}\right]d\zeta \quad (23)$$

Integrating all the values of the combined Hamaker coefficient, A_{132} gives an absolute value for the coefficient denoted by A_{132abs} and applying Lifshitz derivation for van der Waals forces as in equation (23).

IV. Results and Discussion

The absorbance values were obtained as a function of wavelength and summarized in Table 1. It could be seen that the peak absorbance values of the various sputum samples and

components vary in magnitude revealing the notable effect of the bacteria on them. The comparison between the positive and negative samples of the macrophages is imperative to this research. This is because mtb actually attacks the macrophages by attaching itself to the macrophage cells. Figure 2 reveals the disparity between the peak absorbance values of *mtb/HIV* positive and negative sputum samples respectively, and this shows an indication of how the bacteria affect the properties of sputum.

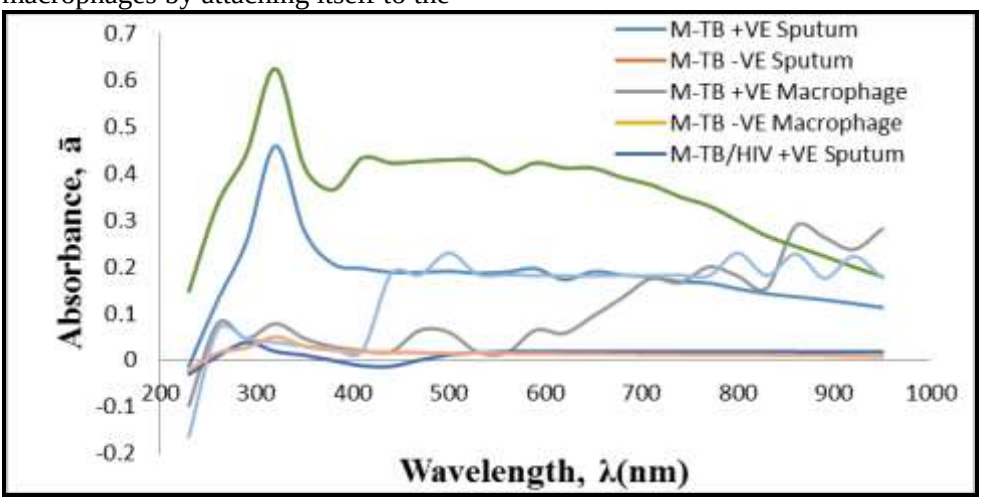

Figure 2: Variation of Average Absorbance, $\bar{a}$ with Wavelength, λ for *mtb*, *mtb/HIV* Sputum and *mtb*, *mtb/HIV* Macrophages

Table 1: Comparison between Peak Absorbance values of *mtb* Positive, *mtb/HIV* Positive and Negative Sputum Components respectively

Sample Type	Wavelength, λ(Å) Peak Values	Absorbance, ā Peak Values				
		M-TB Positive	**M-TB Negative**	**M-TB/HIV Co-infected**	**M-TB/HIV Uninfected**	**Mean Values**
Sputum	320	0.2918 – 0.7877	---	---	---	0.4588±0.1468
		---	0.2657 – 1.2501	---	---	0.6244±0.3545
	290	---	---	0.0231 – 0.0498	---	0.0379±0.0108
		---	---	---	0.2657 – 1.2501	0.6244±0.3545
Macrophage	320	0.0206 – 0.0736	---	---	---	0.0496±0.0116
		---	0.0478 – 0.1148	---	---	0.0784±0.0206
	290	---	---	0.0152 – 0.0637	---	0.0456±0.0106
		---	---	---	0.0478 – 0.1148	0.0784±0.0206

Figure 2 shows a peak absorbance value of greater than 0.60 and 0.45 for *mtb* negative and positive sputum respectively and a peak absorbance value of greater than 0.07 and 0.04 for *mtb* positive and negative macrophages respectively were recorded at wavelength of 320nm. This peak values falls within the visible range of ultraviolet radiation which is between 300 – 600nm. This is important as a reference point in the study of the infection mechanism and may be of importance in determining the critical Hamaker coefficient that favours repulsion between the bacterium and the macrophage. It could be well-known that the infected *mtb* sputum has lower absorbance values than the uninfected ones.

The absorbance values of both the *mtb* positive and negative Macrophages were increasing with increase in wavelength. This may be explained away by the fact of a higher energy level of these cells. The *mtb* infected macrophages gave higher absorbance values than the *mtb* uninfected macrophages. This is a clear indication that infection had occurred and shows the alteration in absorbance values due to *mtb* infection.

The absorbance of the *mtb/HIV* co-infectious sputum samples systematically increased as the wavelength increased until a critical wavelength of 290nm, where peak absorbance values of greater than 0.60 and 0.06 for *mtb/HIV* negative and positive sputum respectively were accomplished. The trend here shows that the uninfected sputum reveals a higher absorbance values at all wavelengths. This indicates that a shift in the energy equation of the system is tenable by some alteration to the sputum as an intervening medium in the *mtb/HIV* – macrophage interaction. It then suggests a possibility of attaining repulsion between the *mtb/HIV* and the macrophage cells by some additives to the sputum.

The absorbance values of the *mtb/HIV* positive macrophages were increasing with increase in wavelength. The *mtb/HIV* co-infected macrophages also gave higher absorbance values than the uninfected ones at wavelengths greater than 400nm. This may explain away by the fact of a higher energy level of these cells. This is a clear indicator that bacteria ingestion had occurred and shows the alteration in absorbance values due to *mtb/HIV* infection.

The trend is such that the mean absorbance peak values of mtb negative sputum samples are reduced by infection from 0.6244± 0.3545 to 0.4588± 0.1468 (see table 1) by a factor of about 26.5%. In mtb macrophage samples, the reduction is from 0.0784± 0.0206 to 0.0496± 0.0116, a factor of about 36.7%. While in mtb/HIV co-infected macrophage samples, the reduction is from 0.0784± 0.0206 to 0.0456± 0.0106 by a factor of about 41.8%. Comparing the mean absorbance peak values of mtb positive sputum samples and the mean absorbance peak values of mtb/HIV co-infected sputum samples; the results of the mean absorbance peak values reveal that the mean absorbance peak value of the mtb/HIV co-infected samples is generally reduced as compared to that of the mean absorbance peak

values of the mtb positive sputum samples (Table 1).

Equation (7) was used to obtain for each interacting system, $A_{ij}(A_{11}, A_{22}, A_{33}, A_{12}, A_{13}, A_{23})$ by approximate change of variables. MatLab computation tools were used. This involved the numerical integration of equation (7) for each wavelength from 230 to 950 for all the twenty samples in each category. Applying Lifshitz derivation for van der Waals forces as in equation (23), The absolute value for the Hamaker coefficient could be derived by obtaining the mean of all the A132, A131, and A232 values got from the Lifshitz relation.

Table 2: Values of the Hamaker Constants and Hamaker Coefficients for the Infected and Uninfected

Variable ($x10^{-21}$ Joule)	**M-TB Samples**				**M-TB/HIV Samples**			
	Infected Sputum		Uninfected Sputum		Infected Sputum		Uninfected Sputum	
	Peak Value	**Absolute Value**	**Peak Value**	**Absolute Value**	**Peak Value**	**Absolute Value**	**Peak Value**	**Absolute Value**
A_{11}	---	---	1.1328	0.9418	---	---	1.1328	0.9418
A_{22}	1.2134	0.9606	---	---	1.0267	0.9786	---	---
A_{33}	0.4205	0.2307	0.6701	0.4247	0.5962	0.2881	0.6701	0.4247
A_{132}	0.5187	0.2163	---	---	0.4253	0.1883	---	---
A_{131}	---	---	0.2241	0.1016	---	---	0.2241	0.1016
A_{232}	0.6298	0.2498	---	---	0.5014	0.2047	---	---

Table (2) show the comparison of the Hamaker constants and coefficients for the positive and negative sputum samples. A_{11} is Hamaker constant for the uninfected sputum samples. A_{22} is the Hamaker constant for the *mtb*, here represented by the infected macrophage. This is as a result of no known process of isolation of the *mtb* at the moment. This is a very close approximation for the bacteria owing to the manner of the infection

mechanism. The Hamaker constants A_{33} for the sputum show greater values for the uninfected samples which regularly indicate a higher surface energy than the infected samples. The higher absolute values of A_{132} and A_{232} as against that of A_{131}, as well as the lower value of the absolute combined Hamaker coefficient A_{131abs} for the uninfected samples is a clear suggestion of the relevance of the concept of Hamaker coefficient in the M-TB infection process. The surface energy A_{131} of the macrophages is less than the surface energy A_{232} of the bacteria (*mtb*).

Table (2) shows the comparison of the Hamaker constants and coefficients for the positive and negative sputum samples. $\tilde{A}_{11}$ is Hamaker constant for the uninfected sputum samples. $\tilde{A}_{22}$ is the Hamaker constant for the *mtb/HIV* co-infection, here represented by the infected macrophage. This is as a result of no known process of isolation of the *mtb/HIV* co-infection at the moment. Though, this is a very close approximation for the bacteria owing to the manner of the infection mechanism. The Hamaker constants $\tilde{A}_{33}$ for the sputum show greater values for the uninfected samples which regularly indicate a higher surface energy than the infected samples. The higher absolute values of $\tilde{A}_{132}$ and $\tilde{A}_{232}$ as against that of $\tilde{A}_{131}$, as well as the lower value of the absolute combined Hamaker coefficient $\tilde{A}_{131abs}$ for the uninfected samples is a clear suggestion of the relevance of the concept of Hamaker coefficient in the *mtb/HIV* co-infection process. The surface energy $\tilde{A}_{131}$ of the macrophages is less than the surface energy $\tilde{A}_{232}$ of the bacteria (*mtb*).

A_{33}, which serves as the energy of sputum as an intervening medium, is seen in *mtb* data to be reduced by infection from 0.4247×10^{-21}J to 0.23067×10^{-21}J by a factor of about 45.7% (see table 2). In *mtb/HIV* co-infection, the reduction is from 0.4247×10^{-21}J to 0.28812×10^{-21}J, a factor of about 32.2% (see table 2).

The reduction is lower in *mtb/HIV* co-infection probably because of the interaction between *HIV* and *tb*. For the combined Hamaker coefficient, the value is 0.21631×10^{-21}J for *mtb* and 0.18825×10^{-21}J for *mtb/HIV*. This result is as expected. *HIV* has the tendency to reduce the energy on the surface of a given material, in this case by about 13%, conforming adverse effects observed in HIV patients with tuberculosis. Note that the values of A_{132} are all positive showing that attraction exists between the macrophage and the *mtb* particles. The effect of the infection can only be abated if a drug, in the form of additive is added that can change the value of A_{132} to negative under that condition, mutual repulsion will occur and it will be expected that, in principle, the *tb* bacteria will not attack the macrophage

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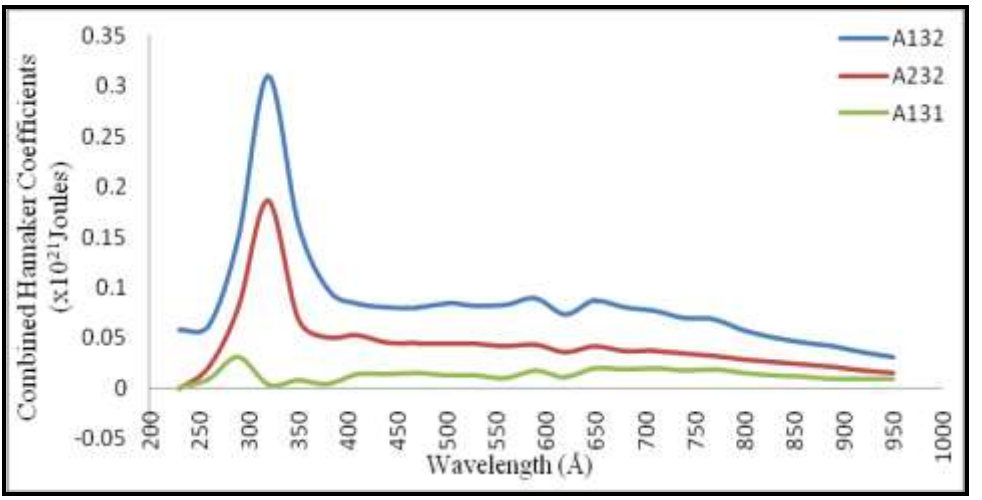

Figure 3: Variation of Average Combined Hamaker Coefficients, A_{132} A_{131} and A_{232} with Wavelength, λ (nm) for the *mtb* infected Sputum Samples

Figure (3) reveals the pattern of the average combined Hamaker coefficients, A_{132}, A_{131} and A_{232} for the sputum samples with clear peak values occurring at various wavelengths. The peak average values of A_{132} and A_{232} occur at wavelength of 320nm with values of 0.30968 x10^{-21}J and 0.18609 x10^{-21}J respectively, while the peak average value of A_{131} occurs at wavelength of 290nm with value of 0.03074 x10^{-21}J. Energy level increases in average combined Hamaker coefficients, A_{132} and A_{232} which is the infected Hamaker coefficients as against the decreased energy level of the uninfected average combined Hamaker coefficient, A_{131}. This is quite a significant phenomenon which explains away the fact of the effect of co-infection.

Table (2) reveal that the surface energy of the macrophages as computed in terms of Hamaker coefficients is less than the surface energy of the *mtb*, *mtb/HIV*. This result also shows that the surface energy of the *mtb/HIV* macrophage is less than that of *mtb* macrophage. *HIV* has the tendency to reduce the energy with the consequence of increased viral loads and decreased immune systems. *tb* is an opportunistic disease and in presence of *HIV*, the consequence is dreadful. Hence for $A_{132} > 0$, $A_{131(\text{macropage})} < A_{232(\text{mtb/HIV})}$.

The mean of all values of A_{11} and A_{22} could be obtained and substituted into equation (19) in order to derive a value for A_{33} at which A_{132} is equal to zero in agreement with the earlier stated reasons. Rearranging equation (19) and making A_{33} subject of the formula;

$$A_{33} = \left[\frac{2\sqrt{A_{11}}\sqrt{A_{22}} - A_{132}}{\sqrt{A_{11}} + \sqrt{A_{22}}}\right]^2 \qquad (24)$$

The mean of all the values of A_{11} and A_{22} respectively gave the absolute values of the Hamaker constants as: A_{11}= 0.94188x10^{-21}Joule and A_{22} = 0.96068x10^{-21}Joule (for *mtb*); $\tilde{A}_{11}$= 0.94188x10^{-21}Joule and $\tilde{A}_{22}$ = 0.97862x10^{-21}Joule (for *mtb/HIV* co-infection). Thus, inserting these values into equation (24) and rendering $A_{132} \leq 0$ will give the

critical value of A_{33C} that satisfies the condition for the combined Hamaker coefficient to be equal to or less than zero. Hence any value of A_{33} greater than the critical would be the desired value necessary to attain a negative combined Hamaker coefficient.

Thus, the critical absolute Hamaker constant A_{33C} for the sputum which renders the A_{132} negative is given as: $A_{33C} = 0.9527x10^{-21}$Joule (for *mtb*) and $\tilde{A}_{33C} = 0.9598x10^{-21}$Joule (for *mtb/HIV* co-infection). Thus for negative combined Hamaker coefficient A_{132}, $\tilde{A}_{132}$ of the infected *mtb*, *mtb/HIV* sputum to be attained respectively, the combined Hamaker constant of the sputum as the intervening medium A_{33}, $\tilde{A}_{33}$ respectively should be of the magnitude: $A_{33C} \geq 0.9527x10^{-21}$Joule and $\tilde{A}_{33C} \geq 0.9598x10^{-21}$Joule respectively. Inserting the above values of A_{33}, $\tilde{A}_{33C}$ into equation (24) would yield negative values for A_{132}, $\tilde{A}_{132}$ respectively as follows: $A_{132} = -0.22669x10^{-25}$Joule (when $A_{33} = 0.9527x10^{-21}$Joule) and and $\tilde{A}_{132} = -0.08786x10^{-25}$Joule (when $A_{33} = 0.9598x10^{-21}$Joule).

Table 3: Computation of the values of Surface Free Energy γ_{sv} of the *mtb and mtb/HIV* Infected and Uninfected Sputum and Change in free energy of adhesion, ΔF^{adh} (mJ/m^2)

Variable	Infected Sputum		Uninfected Sputum		Variable	Hamaker constant, A ($\times 10^{-14}$mJ)	ΔF^{adh} (mJ/m^2)
	Hamaker constant, A ($\times 10^{-14}$mJ)	γ_{sv} (mJ/m^2)	Hamaker constant, A ($\times 10^{-14}$mJ)	γ_{sv} (mJ/m^2)			
$\mathbf{A_{11}}$	---	---	0.94188	37.7	$\mathbf{A_{132}}$	0.21631	-17.3
$\mathbf{A_{22}}$	0.96068	38.5	---	---	$\mathbf{A_{131}}$	0.10165	-8.1
$\mathbf{A_{33}}$	0.23067	9.2	0.42470	17.0	$\mathbf{A_{232}}$	0.24986	-20.0
$\mathbf{\tilde{A}_{11}}$	---	---	0.94188	37.7	$\mathbf{\tilde{A}_{132}}$	0.18825	-15.1
$\mathbf{\tilde{A}_{22}}$	0.97862	39.2	---	---	$\mathbf{\tilde{A}_{131}}$	0.10165	-8.1
$\mathbf{\tilde{A}_{33}}$	0.28812	11.5	0.42470	17.0	$\mathbf{\tilde{A}_{232}}$	0.20474	-16.4

Since the surface energy is a measure of workdone on the surface. Table 3 shows the surface energy of the *mtb* is greater than the surface energy of the sputum. Also, the surface energy of the uninfected sputum 17.0mJ/m2 was apparently reduced to 9.2mJ/m2 when *mtb* attacked the human system. Consequently, the disease incidence lowers the surface energy of the infected sputum as shown in table 2. This in effect indicates that *mtb* has a surface energy reducing capacity.

Table 3 shows that the surface energy of the *mtb/HIV* is greater than the surface energy of the sputum. Also, the surface energy of the uninfected sputum 17.0mJ/m2 was in fact reduced to 11.5mJ/m2 when *mtb/HIV* co-infection attacked the human system. As a result, the bacteria incidence lowers the surface energy of the infected sputum. This in effect indicates that *mtb/HIV* has a surface energy reducing capacity tending to make the surface of the bacteria

(sputum) more hydrophobic. From table 3; the following deductions are made: Surface free energies of infected sputum components are lower than the uninfected, *mtb* infection has the surface energy reducing capacity (i.e. reduces the work done on the surface), It is well identified that *mtb/HIV* attacks the macrophage; the reduction (difference on infected and uninfected) in surface free energies of Sputum could be as a result of its presence in the sputum.

The surface free energies described in table 3 are used to determine the free energy of adhesion. When the change in free energy of adhesion is negative; adhesion is thermodynamically favorable. Adhesion is therefore governed by attractive van der Waal forces. When *mtb* or *mtb/HIV* co-infection affixes itself to the surface of the macrophage in a liquid medium, there is the tendency of the bacteria to be engulfed by the macrophage cells causing their destruction. Thus increased adhesion may lead to more depletion of macrophage cells. The adhesion of the bacteria on the cell will lead to penetration into the macrophage. Such a penetration will cause replication of bacteria and destruction of macrophage. What is required is for this attachment not to occur. Results obtained (Table 3) give the following deductions: The change in free energies of adhesion is penetration, in which case a condition for rendering combined Hamaker coefficient negative is required. Thus, a condition was required for repulsion to occur and that condition was based on the value of A_{33} that would render the absolute combined Hamaker coefficient A_{132abs} negative. A model for the infection mechanism was built-up employing the principle of particle – particle interaction. Mathematically, it was derived as $A_{33} \geq 0.9527x10^{-21}$Joule which satisfies this condition for negative A_{132abs}. To achieve the condition of all negative indicating that the net van der Waals forces are attractive; the change in free energies of adhesion is higher for infected *mtb/HIV* than infected *mtb*. In order words, presence of *HIV* increases the change in free energy of adhesion and hence tendency for increased attack, Hamaker coefficients are all positive suggesting van der Waals forces are attractive as negative free energy of adhesion.

V. Conclusion

The absolute Hamaker coefficient $A_{131}=0.10165x10^{-21}$Joule gives the interaction energy among the macrophage cells in the sputum while A_{232} is the interaction energy among the *mtb* particles in the sputum. A_{232} for *mtb/HIV* co-infection is less than that for *mtb* alone. Reduction in energy in the presence of HIV confirms the adverse effect when *mtb* and HIV occur simultaneously in a patient. This is so since a positive Hamaker value for any interacting system implies an attraction between the interacting bodies or particles while a negative Hamaker coefficient means repulsive van der Waals forces hence the interacting bodies would repel each other. The importance of negative Hamaker coefficient ($-0.2267x10^{-19}$mJ/m^2) indicated that separation of bacteria is realistic. The desired outcome is that the bacteria do not adhere to the macrophage cells to avoid

A_{33} above, possible additive(s) to the system (in form of drugs) to the sputum as intervening medium should be required. That, as expected, may be the much desired way out for drug resistant strains of the *mtb* bacteria.

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