Research Article

Bioavailability of Micronutrient in Functional Tablet Developed from Blend of *Baobab pulp*, Moringa Leaf Powder and Malted maize Flour for Iron Deficient Pregnant

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Abstract

In this study tablet were prepared from blend of *Baobab pulp* (BP), *Moringa oleifera* leaf powder (MLP), *Ocimum gratissimum* leaf powder (OG) with malted maize (MMF) and wheat flour (WF) for iron deficient pregnancy. These raw materials were blended in different ratio respectively according to World Health Organization/United Nations Pregnancy Fund requirement nutrient intake per day for each mineral. The main objective of the study was to evaluate the content of Phytate (PA), Iron (Fe), Zinc (Zn), Calcium (Ca), Magnesium (Mg), and bioavailability of Fe, Zn and Ca in terms of molar ratio in the tablet. Content of Fe (3.87-11.88 mg), Zn (2.23-5.8 mg), Ca (245.30-562 mg), Mg (85.60-248 mg) and Phytate (PA) (9.26-17.23 mg/100g) were significantly increased (P<0.05) as the blending ratio of BP and MLP increased. Molar ratios of PA: Fe (0.203-0.123), PA: Ca (0.018-0.022) and PA: Zn (2.925-4.089) was below the critical values 1.0, 0.24 and 15.0 respectively. Hence, Fe, Zn and Ca were bioavailable in all tablet while Fe bioavailability was more observed in T4 (55% BP, 20% MLP, 5% OG) than other treatments. Conception of 100g of this tablet per day, satisfy 38% of iron (Fe) RNI of pregnancy woman. In finally, the study indicated that T4 present good nutritional value and amount of Fe, Zn, Ca and Mg. However, T4 was found to be the best in terms of bioavailability of Fe while Zn, Ca and Mg was also bioavailable.

Keywords: Moringa leaf powder; *Baobab pulp*; Bioavailability; Iron deficiency; Pregnancy; Bio supplement

Introduction

Nutritional status of woman in reproductive age is the one of determinants factors at a good pregnancy development, until deliverance so the maternal mortality [1]. Outside, it effects on the infant mortality and morbidity. Several other micronutrients are known to be lacking in diets, particularly of the socio-economically weaker and physiologically vulnerable sections of the population in developing countries [2]. A recent estimate based on WHO criteria indicated that around 600-700 million people worldwide have marked iron deficiency anaemia, and the bulk of these people live in developing countries [3,4]. In Sub-Saharan Africa, micronutrient deficiency of pregnant woman is a major problem of public health in developing countries [5]. It's another form of malnutrition, which causes high maternal and infant mortality an overwhelming impact on health, productivity and cognitive development [6].

During pregnancy, the iron requirements of pregnant women are increased threefold to cover needs of expansion of maternal red cell mass, growth of the foetal placenta and to expand maternal erythrocyte mass [7]. In pregnant women moderate to severe deficiencies of iron, zinc and folic acid have been shown to increase risk of low birth weight, pregnancy complications and birth defects [7]. Iron deficiency also negatively influences the normal defence systems against infections [6]. While iron deficiency anaemia, especially the more severe form has been shown to be associated with increased maternal mortality, even mild iron deficiency anaemia may increase the rate of premature delivery and perinatal mortality [4,7] have derived that 82% of the pregnant women worldwide are likely to have inadequate usual intakes of zinc and calcium. Several factors have been identified that may increase a pregnant woman's risk for iron deficiency anaemia, including a diet lacking in iron-rich foods, low bioavailability as in the case of iron and zinc, gastrointestinal disease and/or medications that can decrease iron absorption, and a short interval between pregnancies [7].

Benin is one of the developing countries in West-Africa, where repercussion of micronutrient deficiencies caused negative effect on health [2]. Diet in Benin is dominated by cereals consumption at 80% of energy needs [8]. 58% of women aged of 15 to 49 years suffer anaemia [9]. Precisely 26% low form, 30% in moderate form and 2% in several form. The anaemia prevalence of pregnant women is higher than others, particular in a moderate form (44% against 28% of lactating woman). During precede 5 years, 121 death baby and 335 precocious neonatal mortalities has been noticed for 13.663 seven-month pregnancy and more. The perinatal mortality is estimated at 33% [9].

In order to reduce this problem, targeted interventions on prenatal care, and adequate pregnant women nutrition remains a challenge for developing countries like ours. Several supplement methods have been generally adopted to combat iron deficiency. This is one or more of the following four strategies: iron supplementation (i.e. giving iron tablet), iron fortification of certain foods, nutrition education by improving the bioavailability of the dietary iron [7] and intravenous route supplementation techniques [10]. The emergency supplementation periconceptional is very important. Another form of intervention could be to developed the functional bio tablet' supplement as based on a blend of fruits and vegetable product with a mineral and vitamins, which can help to decrease iron deficiency pregnant woman. The Beninese biodiversity contains some fruit and vegetable products which were rich in some nutrient [11]. These raw materials could be used for developed a bio supplement as tablet food, which improve the nutritional and health status of woman in reproduction. Natural food sources are also important potential sources of iron include certain fruits, vegetables, meat, and poultry [12].

Every parts of *Moringa oleifera* are used for different purposes, mostly providing a highly nutritious food for both humans and animals and medicinal purposes to prevent different diseases [12]. It contains vitamins (A, B, C and E), minerals (Ca, Fe, Zn, P, Cu and others), essential amino acids, essential fatty acids (both omega-3 and omega-6 fatty acids) and phytochemicals [12]. Moringa leaf powder is an excellent nutritional supplement and can be added to any dish to increase macro-and micro-nutrients content of the foods. For healthy individuals, a few spoonful of Moringa leaf powder can be added to any meal to make it more nutritious. In case of pregnant women and lactating mothers, consuming fresh or dried Moringa leaf powder or pods can improve a mother's health and reduce iron deficiency anaemia [13]. According to [14], baobab is an important source of human nutrition today in Africa. Chemical analysis of baobab parts showed the presence of proteins, amino acids, iron, vitamin C, A and E in leaves, seeds and fruit pulp [15]. Baobab pulp and leaves have a high antioxidant activity when compared to other fruits and can consequently be considered as so-called functional foods, which may have a positive impact on health in addition to their role as a food. The baobab pulp presents a high nutritional value and bioavailability of the minerals and vitamins especially high vitamin C content in baobab foods [16]. Vitamin C increase the absorption of iron. Tablet are a form of confectionary products usually dried to low moisture content [17]. Plants belonging to Lamiaceae family like Sweet Fennel (*Ocimum gratissimum* Linn.) have retained the attention of researchers, not only because of their high diversity and their distribution around the world but also for their variable use as popular medicines to treat diseases. Several studies have revealed the antimicrobial and fungicidal activities of *O. gratissimum* [11,18].

It is then benefit to propose at woman to reproduction age, a bio supplement as functional food which presents the high bioavailability of micronutrient which will cover the pregnant woman 'micronutrient needs for a successful of pregnancy. The main objective of this study was to determine the nutritional value and bioavailability of micronutrient in developed bio supplement as a functional tablet food formulated from blend of baobab pulp, moringa leaf powder, *Ocimum gratissimum* leaf powder, malted maize and wheat flour to combat iron deficient reproductive women.

Materials and Methods

The raw material is consisted of cereals as maize (*Zea mays*), Fresh *Moringa oleifera* and *Ocimum gratissimum* leaves, wheat flour and baobab pulp (*Adansonia digitata*). Raw material was obtained from local market at Ouando also located in southern Benin. The fresh *Moringa oleifera leaf* was collected in Agricultural park of Abomey-calavi.

Material Preparation

Moringa oleifera and *Ocimum gratissimum* leaves powder were prepared by sorting and cleaning the leaves from any extraneous materials by using tap water and ventilated in a shade area for two days at room temperature. It was dried in an oven at 100°C for 3 hours and powdered by using piston. The powdered leaf was sieved to remove unwanted matter by using 710 µm sieves size in a repeated manner to get fine powder. The seeds of maize were malted as described by [19]. The bio functional tablet has been prepared by mixing, in varied proportions, the *Moringa oleifera* and *Ocimum gratissimum* leaves powder, malted maize and wheat flour and baobab pulp. The mixture was placed in bio paper package.

Bio Functional Tablet Preparation

Tablet were prepared by the procedure of [20]. The *Moringa oleifera* and *Ocimum gratissimum* leaves powder, baobab pulp, malted maize and wheat flour were mixed and stirred. Baking powder (1g) was added. The dough was kneaded for 15 minutes and sheeted to a thickness of 0.6 cm and cut using a mould die of diameter 2,5 cm. Tablet were baked in a pre-heated oven at 180°C for 15 minutes, cooled and evaluated for proximate analysis and bioavailability of minerals.

Bio Functional Tablet Formulation

Five different blend proportions including control were premeditated based on simplex design (Design Expert version 6.0.8 software). Blends were prepared using mixtures of *Moringa oleifera* and *Ocimum gratissimum* leaves powder, malted maize flour, wheat flour and baobab pulp in the different ratios (Table 1). The different tablet was formulated to approximately contain the World Health Organization/United Nations Pregnancy Fund requirement nutrient intake per day for each minerals or vitamins as follows 350 kcal energy, 1.2 to 10.7g protein, 1040mg iron during pregnancy precisely 30mg/day, 600µg folate, 3.4, 4.2 and 6mg zinc respectively for first, second and third pregnancy trimester, 220mg magnesium, 1200mg calcium (last trimester), 800mg vitamin A and 55mg vitamin C [3].

Table 1: Bio functional tablet formulation

Material (%)						
Bio Tablet	**Ratio Vegetable/cereal flour**	**Moringa leaf powder (MLF)**	***Ocimum gratissimum* leaf powder (OLP)**	**Baobab pulp (BP)**	**Malted maize flour (MM)**	**Wheat flour (WF)**
T1	60/40	10	5	45	15	25
T2	70/30	15	5	50	10	20
T3	75/25	20	5	50	10	15
T4	80/20	20	5	55	10	10
T5	90/10	20	5	65	5	5

Physico-chemical Analysis

These analysis of tablet were determined using recommended methods [21]. The tablet was analysed for dry matter, pH and titratable acidity.

Nutritional Analysis

Proximate composition of protein, fat and ash contents of the samples were determined following [21] and the process described by [22]. Total available carbohydrate was calculated as 100% minus the sum of protein, fat and ash contents obtained as described above. Energy Value (EV) was calculated according to equation of Atwater and Benedict (1902). EV = (9 x Fat (%) + 4 x Proteins (%) + 4 x Carbohydrates (%)) according to the method of [23].

Bioavailability Determination

Minerals Analysis

Iron, Zinc and Calcium content of tablet were determined by using Atomic absorption spectrophotometer (AAS: Varian SpectrAA 200) by using the procedure reported by [22].

Inhibitor Factors Analysis

Phytate content of fermented infant flour was determined by using the procedure of [25] and Tannin content was determined according to the procedure of vanillin- HCl methods by [24].

Bioavailability Determination

Bioavailability of minerals (Iron, Zinc, Calcium and Magnesium) was determined by using the molar ratio of Phytate to mineral of formulated tablet according to method described by [26]. The mole of Phytate and mineral was determined by dividing the actual weight of Phytate and mineral with their atomic weight or molar weight. The calculated molar ratios were compared with critical values of Phytate: Calcium>0.24, Phytate: Iron>1 and Phytate: Zinc>15.

Statistical Analysis

The data were evaluated by analysis of variance (ANOVA) procedures in Statistica 7.1 were used to perform descriptive analysis and compare the means of triplicate measurements of parameters. Let's notify that the means were considered to be significantly different when $p<0.05$. The least significant difference test was used to separate the means when the difference was significant.

Results and Discussion

Physico-chemicals and Nutritional Value of Tablet

The physico-chemical and nutritional value of formulated tablet were showed in Table 2. The dry matter, pH and titratable acidity in the formulated tablet ranged respectively as 94.51-96.93%, 3.85-5.35 and 0.21-2.35 eq as lactic acid. The T4 and T5 Tablet have a higher dry matter and titratable acidity compared to others formulation of tablet. The five formulated tablet revealed dry matter, carbohydrates, protein, fat, and ash which were statistically different (P<0.05). These parameters contents have increased concomitantly of T1 to T5 formulation Table 2. The protein, fat and ash content have increased of 8.6-11.68, 1.8-4.05 and 4.3-7.02 respectively. Energy value of tablet varied of 374.24 to 379.89 Kcal. The energy value (372.18) of formulation T2 (15% MLP, 5% OLP, 50% BP, 10% MM and 20% WF, T3=20% MLP) was lower than that T1 (15% MLP, 5% OLP, 50% BP, 10% MM and 20% WF). Contrary the energy value (378.14) T3 tablet were higher than that T2 tablet. When the amount of MLP and BP increased in the blending ratio protein, fat and ash contents of the tablet increased dramatically [12]. Showed that the Moringa leaf powder is an excellent nutritional supplement and can be added to any dish to increase macro- and micro-nutrients content of the foods. For healthy individuals, a few spoonsful of Moringa leaf powder can be added to any meal to make it more nutritious [13]. This indicated that MLP and BP contained more protein, fat and ash content than malted maize and wheat flour. The incorporation of malted flour has permit to improve the nutritional value of tablet [22] has showed that the incorporation of malted maize flour in infant flour formulation increase the nutritional value and mineral content. In formulated tablet, T4 and T5 present a higher nutritional value compared to others tablet formulation. There is no significant difference between ash content of T4 and T5 tablet. But, the titratable acidity of T5 tablet was higher than T4 tablet. The higher titratable acidity of T5 show a higher acidity, which is not consumption. The T4 tablet which contain 55% BP, 20% MLP, 5% OLP, 10% MM and 10% WF present a good nutritional value compared at others formulation.

Table 2: Biochemical characteristics of formulated tablet

	Characteristics of tablet						
Tablet	**Dry matter**	**pH**	**Titratable acidity (eq. as lactic acid)**	**Protein**	**Fat**	**Ash**	**Energy value (Kcal)**
T1	94.51±0.05^{a}	5.20±0.05^{a}	0.21±0.05^{a}	8.6± 0.15^{a}	1.8± 0.15^{a}	3.2± 0.10^{a}	374.24±1.50^{b}
T2	95.25±0.03^{b}	5.32±0.01^{b}	0.24±0.10^{b}	10.04±0.10^{b}	2.7±0.12^{b}	5.6± 0.12^{b}	372.18±1.20^{a}
T3	96.36±0.02^{c}	5.35±0.02^{b}	0.26±0.01^{b}	11.23±0.12^{c}	3.5± 0.42^{c}	6.2± 0.12^{c}	378,14±1.51^{c}
T4	96.85±0.03^{d}	4.05±0.03^{c}	1.75±0.05^{c}	11.5± 0.15^{d}	3.8± 0.50^{d}	6.8± 0.30^{d}	379,28±1.20^{d}
T5	96.93±0.02^{e}	3.65±0.02^{d}	2.35±0.25^{d}	11,72±0.06^{e}	4.05±0.10^{e}	7.02±0.15^{d}	379,89±0.80^{e}

Means of the same letter within a column are not significantly different (P>0.05). *T1=10%MLP, 5%OLP, 45%BP, 15%MM and 25%WF, T2=15%MLP, 5%OLP, 50%BP, 10%MM and 20%WF, T3=20%MLP, 5%OLP, 50%BP, 10%MM and 15%WF, T4=20%MLP, 5%OLP, 55%BP, 10%MM and 10%WF, T5=20%MLP, 5%OLP, 65%BP, 5%MM and 5%WF*

Mineral Content of Tablet

Mineral content of formulated tablet was summarized in Table 3. The analysis result of Fe, Zn, Ca, and Mg in the formulated tablet ranged as 3.87-11.88mg, 2.23-5.8mg, 245.30-562mg and 85.60-248mg respectively. The maximum (11.88mg) and minimum (3.87mg) Iron content was recorded for the T5 tablet made from 65% BP, 20% MLP, 5% OLP, blended and first formulation as T1 tablet (45% BP, 10% MLP, 5% OLP, 15% MM and 25% WF) respectively. Statistically significant difference ($P<0.05$) was observed among the treatments. When the amount of MLP and BP increased in the blending ratio Fe contents of the tablet increased dramatically. This indicated that MLP and BP contained more Fe content than malted maize and wheat flour. The finding of this study is in close agreement with the work of [27,28] showed that the incorporation of 20% Moringa Leaf Powder (MLP) blended and control (100% wheat flour) cookies has permit to increase dramatically iron content of 2.87mg to 7.77mg. The incorporation of baobab pulp (BP) in malted infant flour (containing malted maize and sorghum flours in rate 50/50) roasted improved the taste of porridge and increased the iron content until 14.32mg of iron [22,23] showed that the incorporation of baobab pulp (BP) in infant flour developed with taro and roasted soybean flours have increased the iron content until 6.61mg of iron. The formulated tablet also has better Fe content as compared with the work of [28,29]. This showed that the blending of MLP, BP and OG presents a high mineral content tablet, which could be used as good bio supplement for cover the micronutrient deficiency of pregnancy woman. The T4 tablet which contain 20% MLP, 5% OLP, 55% BP, 10% MM and 10% WF present a good nutritional value and higher mineral content compared at others formulation. Result show that T4 tablet contains 11.5mg of iron, 5.2mg of zinc, 525mg of calcium and 234mg of magnesium. This developed tablet could be used as bio supplement for reduce the pregnancy micronutrient deficiency. The usual dose prescribed in early pregnancy is 30mg of elemental iron per day [4]. The consumption of 100g of T4 tablet (20% MLP, 5% OLP, 55% BP, 10% MM and 10% WF) per day could be estimated to satisfy 38% of Fe RNI for pregnancy women (30mg/day). Iron is necessary for the production of haemoglobin, which is an essential protein found in erythrocytes. During pregnancy, iron is also needed for the development of the foetus and placenta and to expand maternal erythrocyte mass [4]. The minimum and maximum Zn content was recorded for T1 tablet (2.23mg) and T5 tablet (5.8mg) respectively. There was an increment of Zn content in the formulated tablet as MLP and BP increased in the blending ratio, this pointed as MLP and BP contains more Zn content than malted maize and wheat flour. Similar result was observed in the finding of [30] who evaluated the mineral content of cookies developed from blend Wheat-Mungbean flour but the formulated cookies have less Zn content as compared with the work of [30]. The consumption of 100g of T4 tablet per day could be estimated to satisfy about 65%, 81% and 100% respectively for first, second and third pregnancy trimester of Zn Requirement Nutrient Intake (RNI) for pregnancy woman. The consumption of these MLP and BP blended tablet may contribute to build up normal growth and neurobehavioral development, immune and sensory function, antioxidant protection and membrane stabilisation. It also contributes to reduce the risk of inborn abnormality, low birth weight and other complications of pregnancy and delivery, such as impaired development and premature delivery. The least and the highest Ca content was recorded for T1 tablet (245.30mg) and T5 (562mg) respectively. Statistically, significant difference ($P<0.05$) was observed among the formulated tablet. This dramatic increment of Ca content in the formulated tablet was due to higher amount of Ca in MLP and BP than malted maize and wheat flour. Calcium is required for the normal development and maintenance of the skeleton. Low calcium intake has been associated with loss of bone mass (osteoporosis), resulting in bone fracture in older people, especially women [31]. Calcium reserves in bone are affected by dietary calcium intake and calcium losses from the body in different cases, for example woman has need supply Ca a last trimester during pregnancy. The Requirement Nutrient Intake (RNI) of pregnancy women is 1200mg per day (in last trimester) of Ca. She needs additional Ca supplementation in her diets [28]. Hence, this MLP, BP and OG (vegetable antibiotics) blended tablet expected as good source of Ca and contributes to supply the required amount of Ca to their body during pregnancy periods. The formulated tablet has better Ca content as compared with the work of [28,30]. According to [3] report 100g MLP and BP blended tablet could be estimated to fulfil about 20.44-46.83% of the required Ca RNI (1200mg) for pregnant woman. Consumption of 100g T4 tablet which contains 20% of Moringa LP, 5% OLP, 55% BP, 10% MM and 10% WF contribute to cover 43.75% of pregnant woman RNI of Ca. The minimum and maximum magnesium (Mg) content was recorded for T1 (85.60mg) and T5 (248mg) tablet respectively. Statistically, significant difference ($P<0.05$) was observed among the formulated tablet. The increasing of magnesium (Mg) content in the formulated tablet with respect to the level of MLP and BP ratio in the blend was due to high amount of Mg content in MLP and BP than malted maize flour and wheat flour. The MLP and BP blended tablet satisfy 38,90 to 100 of the required Mg RNI for pregnant woman by consuming 100g T4 tablet. This helps the women to supply the daily required amount of Mg for her as well as her baby in growth. It also contributes to reduce the risk of inborn abnormality, low birth weight and other complications of pregnancy and delivery, such as impaired development and premature delivery [31]. Mg is one of important minerals for bone and teeth related nutrients. Calcium provides bone stiffness and child growth [22]. Zinc represents with iron one of the most concentrated minerals in the brain. Zinc is also implicated in immunity as it reduces the incidence and severity of diarrhea in children [2]. Magnesium is needed for biochemical reactions in the body to maintain muscle, improve nerve function, maintain heart rhythm, and regulate blood sugar levels [32]. The incorporation of OG leaves powder could permit to increase the antioxidant activity of formulated tablet [33]. This is antioxidant protection is due to components of OG leaves powder. This bio efficacy may be due to the presence of some highly fungitoxic and antimicrobial components in the OG leaf powder [33]. Plants belonging to Lamiaceae family like Sweet Fennel (*Ocimum gratissimum Linn*.) were used as popular medicines to treat diseases. Several studies have revealed the antimicrobial and fungicidal activities of *O. gratissimum* [18,33].

Table 3: Mineral Composition of the formulated tablet (mg/100g)

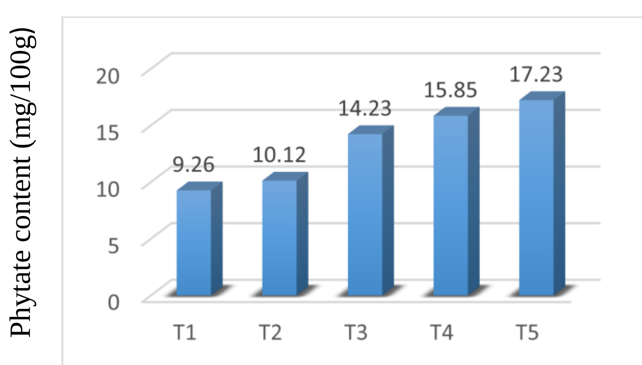
Tablets	Fe	Zn	Ca	Mg
T1	3.87[a]	2.23[a]	245.30[a]	85.60[a]
T2	5.56[b]	3.5[b]	386.75[b]	168.56[b]
T3	9.8[c]	4.7[c]	485[c]	205.52[c]
T4	11.5[d]	5,2[d]	525[d]	234[d]
T5	11.88[e]	5.8[e]	562[e]	248[e]

Means of the same letter within a column are not significantly different (P>0.05). *T1=10%MLP, 5%OLP, 45%BP, 15%MM and 25%WF, T2=15%MLP, 5%OLP, 50%BP, 10%MM and 20%WF, T3=20%MLP, 5%OLP, 50%BP, 10%MM and 15%WF, T4=20%MLP, 5%OLP, 55%BP, 10%MM and 10%WF, T5=20%MLP, 5%OLP, 65%BP, 5%MM and 5%WF*

Inhibitors Factors of Tablet

The inhibitors factors of formulated tablet such as Phytate (PA) were analysed. Their results were ranged from 9.26 to 17.23mg/100g as shown in Figure 1. Maximum Phytate content was recorded for T5 tablet (17.23mg/100g) and minimum Phytate content was recorded for T1 tablet (9.26mg/100g). Significant difference (P<0.05) was observed among the formulated tablet. When more BP and MLP was added in the blending ratio, the Phytate content was increased. This may be due to the presence of high amount of Phytate (PA) in BP and MLP leaves powder than malted maize and wheat flour. However, the formulated tablet may not be considered as high PA containing foods. According to [36] report, foods to be considered as high PA containing-foods, it must contain higher than 400 mg PA. Therefore, the PA contents of all tablet are below 400mg. The less PA (9.26 mg/100g) containing tablet among 10% MLP, 5% OLP, 45% BP, 15% MM and 25% WF (T1) tablet. This less amount of PA containing tablet may maintain the bioavailability of dietary minerals (Fe, Zn, and Ca) of the tablet; at the same time the lower level of PA may have some health promotional activities as an antioxidant and anti-carcinogens when ingested by humans it may reduce the risk of colon cancer and some other inflammatory bowel diseases [28].

Figure 1: Level of inhibitor factors (Phytate) of Formulated Tablet



Bioavailability of Minerals in Tablet

Iron Bioavailability

Minimum molar ratio of PA: Fe was recorded for the T4 (20% MLP, 5% OLP, 55% BP) tablet (0.116) and the maximum molar ratio of PA: Fe was recorded for the T1 (control) (0.203). Significant difference (P<0.05) was observed among the treatments. The results indicated (Table 4) that T1 (control) tablet showed very less Fe bioavailability compared with others, whereas T4 (20% MLP, 5% OLP, 55% BP blended) showed the highest Fe bioavailability (0.116<1). The Fe bioavailability of other blended tablet including control were good, because of their molar ratio below the critical value (1) and the binding effect of Phytate (PA) on Fe may be high; whereas the Fe bioavailability of T3 (20% MLP, 5% OLP, 50% BP) and T5 (20% MLP, 5% OLP, 65% BP) might be good because of PA: Fe molar ratio (0.123) respectively below critical value with devoid of binding effect of PA on Fe. The measured molar ratio values of PA: Fe was decreased gradually when BP and MLP amounts were increased. Contrary the bioavailability of formulated tablet was increased. So the bioavailability of Fe was less when more BP and MLP added in the blending ratio. However, the formulated tablet can be concluded as low-Phytate foods as their molar ratio of PA: Fe was very below 0.5 (PA: Fe<1 means foods with low-Phytate content) [34]. The formulated tablet (PA: Fe = 0.116-0.203) has less molar ratio value compared with cookies developed in study of [28] which presents a high molar ratio PA: Fe (PA: Fe = 0.996-1.587). Therefore, the bioavailability of Fe in these formulated tablet might be better than these cereal foods [35]. Iron has a number of key functions within the body so one can get adequate amount of Fe in the food otherwise deficiency of Fe might be appeared. Deficiency of Fe (a person which has normal haemoglobin concentrations but no Fe stores in the body) ultimately leads to Fe deficiency anaemia (low haemoglobin concentration and low iron store), the most common cause of anaemia, a condition in which the blood lacks healthy red bloods cells required to carry oxygen, and which results in morbidity and death because of the lack of proper function of cells in the body [36]. Therefore, among the formulated tablet, T4 (20% MLP, 5% OLP, 55% BP blended) is the best source of Fe bioavailability and Fe might be easily absorbed within the gastrointestinal tract and expected to contribute to minimize Fe deficiency problems of pregnant woman as well as others by using calculated amounts of tablet based on their RNI.

Table 4: Phytate - Minerals molar ratio of formulated tablet

Tablet	PA: Fe	CV > 1	PA: Zn	CV > 15	PA: Ca	CV > 0.24	Ca * PA/Zn	CV > 0.5
T1	0.203^{d}	1	4.089^{e}	15	0.022^{d}	0.24	0.015^{a}	0.5
T2	0.154^{c}	1	2.847^{a}	15	0.015^{a}	0.24	0.016^{b}	0.5
T3	0.123^{b}	1	2.981^{c}	15	0.017^{b}	0.24	0.022^{c}	0.5
T4	0.116^{a}	1	3.001^{d}	15	0.018^{c}	0.24	0.024^{d}	0.5
T5	0.123^{b}	1	2.925^{b}	15	0.018^{c}	0.24	0.025^{e}	0.5

T1=10%MLP, 5%OLP, 45%BP, 15%MM and 25%WF, T2=15%MLP, 5%OLP, 50%BP, 10%MM and 20%WF, T3=20%MLP, 5%OLP, 50%BP, 10%MM and 15%WF, T4=20%MLP, 5%OLP, 55%BP, 10%MM and 10%WF, T5=20%MLP, 5%OLP, 65%BP, 5%MM and 5%WF; PA = Phytate, M = Mean, CV = Critical Value; Means of the same letter within a column are not significantly different (P>0.05).

Zinc Bioavailability

Molar ratio of Phytate: Zinc of the tablet ranged from 2.925 to 4.089 as presented in Table 4. The minimum value (2.847) was recorded for T2 (15% MLP, 5% OLP, 50% BP blended) and the maximum value (4.089) was recorded for T1 (10% MLP, 5% OLP, 45% BP blended). Statistically, Significant difference (P<0.05) was observed among treatments. The PA: Zn molar ratio decreased as the level of BP and MLP increased in the formulation. According to [37] the foods with PA: Zn molar ratio greater than 15, between 5 and 15, less than 5 are considered as low (15%), moderate (35%) and high (55%) Zn bioavailability respectively. The molar ratios of all formulated tablet were less than 5. Hence, the bioavailability of Zn in all treatments is a high (55%) level. Therefore, the formulated tablet may be considered as high source of Zn for pregnancy woman. The bioavailability result of Zn is higher than that showed in the work of [28,30] reported on cookies made from composite flour of Wheat and Full Fat Soya and wheat flour-Moringa Leaves powder respectively. The higher of zinc bioavailability in tablet was due to higher ratio of BP, MLP and vegetable antibiotic OG leaf powder compared to malted maize and wheat flour.

Calcium Bioavailability

Minimum and maximum PA: Ca molar ratio was recorded for T2 (15% MLP, 5% OLP, 50% BP blended) (0.015) and T1 (0.022) respectively. Statistically, significant difference (P<0.05) was observed among treatments. The molar ratio of PA: Ca in all formulated tablet was very below the critical value of 0.24. The decreasing of molar ratio directs to increasing the bioavailability of Ca. This result indicated that the bioavailability of Ca may not be hindered by the PA content of the formulated tablet. Because the PA: Ca molar ratio in the tablet higher decreased as BP and MLP amount increased in the blend ratio. Therefore, the formulated tablet may be considered as high sources of Ca for pregnant woman and it may contribute for them to maintain bone formation, blood clotting and muscle contraction. The level of BP and MLP increased in the blending, the molar ratio of [Ca]*[PA]/ [Zn] increased from 0.015 (T1) to 0.025 (T5) and the effect of PA on Zn appears to increase highly. However, comparatively the [Ca]*[PA]/ [Zn] values of all formulated tablet were very below the critical value, 0.5. This indicated that the ratio of [Ca]*[PA]/ [Zn] is a higher predictor for Zn availability. The molar ratio less than 0.5 means there would not be interferences in the availability of Zn in the formulated tablet. The inhibitory effect of PA on Zn absorption by forming a Calcium-Zinc-Phytate complex in the intestine makes less and soluble the Phytate complexes formed by either ion alone [36]. Therefore, Phytate has no effect on Zn bioavailability during minerals absorption; hence, the formulated tablet may be considered as high source of Ca and Zn.

Conclusion

These formulated tablet contain good nutritional value conforms to recommendations. The T4 tablet present the better nutritional value and present a higher iron content compared others formulated tablet. This T4 tablet has an acceptable pH for consumption compared to T5 tablet. The bioavailability of Fe, Ca and Zn were very below their critical values during increasing of BP and MLP. But in 20% MLP, 5% OLP, 55% BP, 10% MM and 10% WF (T4) tablet, Fe bioavailability was in lower range than others due to its molar ratio (0.116) higher below critical value (PA: Fe<1). The T4 tablet has a higher bioavailability of iron compared at others formulated tablet. Therefore, consumption of 100g of T4 tablet have a potential to satisfy 38% of Fe of the RNI (30 mg/day), 65%, 81% and 100% respectively for first, second and third pregnancy trimester of Zn RNI and 43.75% of RNI of Ca (1200 mg/day) required by pregnant woman. In general, it might have a possibility to contribute better iron need (Fe) for pregnant woman to combat Iron deficiency problems.

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Conflicts of Interest

Authors declare that they have no conflicts of interest.

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