PACIFIC JOURNAL OF MEDICAL SCIENCES

VOLUME 12, No. 2, APRIL 2014

PACIFIC JOURNAL OF MEDICAL SCIENCES

{Formerly: Medical Sciences Bulletin}

ISSN: 2072 – 1625

Pac. J. Med. Sci. (PJMS)

www.pacjmedsci.com. Email: pacjmedsci@gmail.com.

ISSN: 2072 – 1625 Volume 12, No. 2, April 2014

A multidisciplinary journal for publication of medical and biomedical research findings on issues pertinent to improving family health and related issues of public health

PACIFIC JOURNAL OF MEDICAL SCIENCES

(Formerly Medical Sciences Bulletin)

ISSN: 2072 – 1625 Volume 12, No. 2, April 2014

A multidisciplinary journal for publication of medical and biomedical research findings on issues pertinent to improving family health and related issues of public health

INFORMATION

SUBSCRIPTIONS: Correspondences concerning subscriptions, purchase of single copies and back issues, lost copies and related business should be addressed to the Secretary, Basic Medical Sciences, School of Medicine and Health Sciences, University of Papua New Guinea, P. O. Box 5623 Boroko, N.C.D., PNG. Official website is www.pacjmedsci.com; For Instructions to Authors please visit the official website.

April 2014: **ISSN: 2072 – 1625** **VOLUME 12, No. 2**

TABLE OF CONTENTS **Page #**

EVALUATION OF TRIPLE DRUG COMBINATION (TELMISARTAN, AMLODIPINE AND HYDROCHLORTHIAZIDE) IN THE MANAGEMENT OF HYPERTENSION

*V. K. Abhichandani, MD and **A. A. Faruqui, MD

*Consultant Physician & Diabetologist, Ramanand Clinic, K. B. Commercial Center, Khanpur, Ahmedabad-380001

**Associate P, 16th Road, Near Seepz, Andheri East, Mumbai-400093

Corresponding Author: Dr. A. A. Faruqui, (MD): drfaruqui@gmail.com

ABSTRACT:

Despite many therapeutic options available only one-third of hypertensive patients achieve target Blood pressure (BP). The present study was undertaken to evaluate the efficacy and safety of triple drug, fixed dose combination of Telmisartan 40 mg + Amlodipine 5 mg + Hydrochlorothiazide 12.5mg, in the management of hypertensive patients with or without co-morbidities. A total of 60 patients were enrolled on the basis of mean seated cuff systolic blood pressure >160 mmHg and diastolic blood pressure >100 mmHg in this post-marketing surveillance (PMS) study. Patients were prescribed triple drug fixed dose combination for 120 days. In all groups (diabetic hypertensive, Dyslipidemic hypertensive and hypertension without any complication) there was statistically significant decrease ($p<0.0001$) in systolic blood pressure (SBP) from the baseline to 30th, 60th and 120th days of the treatment. Diastolic BP (DBP) was significantly decreased ($p<0.0001$) from the baseline just after 15th day of the treatment and on subsequent days of observation. 50% of diabetic hypertensive patients and 78.5% of hypertensive patients with dyslipidaemia achieved the Joint national committee VII (JNC VII) recommended goal (130/80mm Hg) at the end of study period of 120 days. Similarly in patients with hypertension without complication, 81.3% achieved the JNC VII recommended goal (140/90mm Hg) at the end of the study period. Triple drug fixed dose combination therapy of Telmisartan, Amlodipine and hydrochlorothiazide has been shown to be an effective, safe and convenient treatment strategy in controlling the blood pressure and achieving the desired blood pressure goal.

KEYWORDS: Diastolic, Systolic, Blood Pressure, Hypertension, Triple drug fixed dose combination

Submitted November 2013; Accepted February 2014

INTRODUCTION:

Hypertension (HTN) is a major public health problem in India and globally [1]. It is a major risk factor for coronary events, stroke, heart failure, peripheral vascular disease, and progression of kidney disease [2] which lead to cardiovascular mortality. In spite of the many therapeutic options available only one-third of hypertensive patients achieve correct blood pressure (BP) levels [3]. Because the etiopathogenesis of hypertension is multifactorial, most patients require more than one antihypertensive drug to achieve correct BP [4]. In patients with high or very high cardiovascular risk, such as diabetics or those with renal failure, a combination of three or more antihypertensive drugs with different mechanisms of action is required to reach the desired BP goal (BP <130/80)[5].

The use of antihypertensive combinations with complementary mechanisms of action results in greater BP reductions than those achieved by the sum of each drug in monotherapy [6]. For patients with very-high baseline BP values or those at high cardiovascular risk, European guideline recommends the combined use of a calcium channel blocker, an angiotensin II receptor blocker and a thiazide diuretic[5]. As demonstrated in various clinical studies concept of monotherapy up-titration to achieve BP target has been repetitively challenged. Such strategy is not likely to achieve the same BP-lowering effect in comparison to combination therapy [7]. In a recent meta-analysis, it was observed that the BP-lowering effect of combination drugs from two different classes was five times more than doubling the dose of a single drug [8].To achieve optimal, recommended BP targets set by Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC VII) (i.e.130/80 mmHg for diabetic hypertensive & 140/90mmHg for hypertensive without any complication), most hypertensive patients requires a combination of two or more BP-lowering drugs, and monotherapy would likely be sufficient only in a small proportion of patients (about 20%–30%) [9]

Triple-combination formulations are emerging and may additionally offer the advantage of further reducing the pill burden [10]. In addition, single-pill combination (SPC) drugs have also gained opinion as the preferred approach to combine BP-lowering drugs in recently updated European guidelines [11]

The objective of this study was to evaluate the efficacy and tolerability of fixed-dose triple-combination of Telmisartan, Amlodipine and Hydrochlorothiazide in the management of hypertension.

SUBJECTS AND METHODS:

This was a post marketing surveillance (PMS), non-randomized, open, non-comparative, mono centric study conducted in a hospital based in Ahmedabad, India. Triple drug fixed dose

combination of Telmisartan 40 mg, Amlodipine 5mg and Hydrochlorothiazide 12.5mg was administered to hypertensive patients for 4 month (120 days). Informed consent was obtained from the patients & the post marketing surveillance was in accordance with the clinical principles laid down in declaration of Helsinki.

Inclusion Criteria

Both male and female patients aged ≥ 35 years willing to give informed consent were included. Basis of selection solely based on the seated cuff SBP and DBP value and was uncontrolled on dual therapy. Patients with seated cuff SBP 140–186 mm Hg and DBP 96–120 mm Hg were included in this study.

Exclusion Criteria

Patients were excluded from entry into the study if they had a known/suspected history of hypersensitivity to any of the drugs of the fixed dose combination, hepatic encephalopathy, and known cases of hepatic or renal insufficiency, pregnant or lactating women. In addition, patients with a history of coronary disease, congestive heart failure, or a recent acute cardiovascular event (previous 3 months) or stroke (previous 6 months) were excluded.

Patient distribution

A total of 60 patients were monitored in the study and entered into the final analysis. Out of 60 patients 32 were Male and 28 were female. Female patients were in the age range of 35-68 years and male patients were in the age range of 37-73 years old. The patients were categorized into three groups based on their medical history. The three groups were diabetic hypertensive (14) (23.3%), Hypertension with dyslipidemia 14 (23.3%) and hypertension without any complication 32 (53.3%) as shown in the Table 1.

Table 1: Distribution of Patients:

	Hypertension	Hypertension with diabetes	Hypertension with Dyslipidaemia
Number (%) of Patients	32 (53.3%)	14 (23.3%)	14 (23.3%)

Evaluation of primary outcome measure: the parameters recorded at baseline and on the 15th, 30th, 60th and 120th day of the study were systolic blood pressure, diastolic blood pressure

Evaluation of secondary outcome measure: Global assessment of efficacy and safety; efficacy was evaluated at the end of the study. Investigator assessed efficacy by using a three point scale as poor, good and excellent. Poor was for those patients, whose BP did not change from baseline, good when BP changed by 15% from the baseline and excellent for those who achieved the target BP. Global assessment regarding safety was evaluated by recording any

adverse event or any complaint during the therapy in every visit.

Statistical analysis

Data analysis on patient demographics and various outcome measures were performed using graph pad prism 5 (software for statistical analysis). Comparison between the baseline values with the value on the 15th, 30th , 60th and 120th day of treatment were made, as well as comparison in between these days by applying one way analysis of variance & the Turkeys multiple comparison test. Values of $P < 0.05$ were considered as significant.

RESULTS

Diabetic Hypertensive group

The systolic blood pressure was recorded at the baseline and subsequently at 15th, 30th , 60th and 120th days of treatment. The baseline Mean ± Standard Deviation (SD) SBP was 158 ± 11.2 mmHg. The mean SBP at 15th, 30th, 60th and 120th days of the treatment were 153 ± 10.4 mmHg, 147±9.75 mmHg, 141±6.64 mmHg and 136±6.13 mmHg respectively (Table 2).

There was statistically significant decrease ($p<0.0001$) in the SBP from the baseline to 30th, 60th and 120th days of the treatment (Table No-02). Changes in SBP were 17 ± 4.56mmHg and 22±5.07 mmHg from baseline to 60th and 120th days of the treatment respectively.

Diastolic blood pressure (DBP) was recorded at the baseline and at 15th, 30th, 60th and 120th days of treatment. The mean DBP at baseline was 107 ± 6.21 mmHg and on 15th, 30th, 60th and 120th days of the treatment were 101 ± 5.13, 95.3 ± 4.61, 90.4 ± 3.25 and 85.3 ± 2.67 mmHg respectively. There was statistically significant decrease ($p < 0.0001$) in DBP from the baseline to 15th, 30th, 60th and 120th days of the treatment (Table 2). Changes in DBP were 12 ± 1.6mmHg, 16.6 ± 2.96mmHg and 21.7 ± 3.54mmHg from baseline at 30th, 60th and 120th days of the treatment respectively.

Table 2: Effect of triple drug therapy on Blood Pressure in Hypertension with diabetes

	Base line	15th Day	30th Day	60th Day	120th Day
SBP	158 ± 11.2	153 ± 10.4*	147±9.75***	141±6.64***	136±6.13***
DBP	107 ± 6.21	101 ± 5.13**	95.3±4.61***	90.4±3.25***^	85.3±2.67***^^

* $p<0.05$ vs Baseline, ** $p<0.01$ vs Baseline, *** $p<0.0001$ vs Baseline, ^ $p<0.05$ vs 30th Day, ^^ $p<0.0001$ vs 30th Day

At the end of the study period the BP in 7 of the 14 patients in the group was within the range 130/80 mmHg. This indicated that 50% of patients in the diabetic hypertensive group achieved the desired goal (130/80 mmHg) set by JNC VII.

Dyslipidemic Hypertensive group

Systolic blood pressure was recorded at the baseline and on subsequent 15th, 30th, 60th and

120th days of treatment. The baseline Mean ± Standard Deviation (SD) SBP was 160 ± 11.4 mmHg. The mean SBP at 15th, 30th , 60th and 120th days of the treatment were 155 ± 9.88, 150±9.97, 143±8.05 and 137±7.54 mmHg respectively (Table 3). There was significant decrease (p<0.0001) in systolic blood pressure from the baseline at 30th, 60th and 120th days of the treatment. Moreover there was significant decrease in systolic blood pressure between 15th day and 60th & 30th and 120th days of treatment. Changes in systolic blood pressure were 17±3.35 and 23±3.86 mmHg from baseline to 60th and 120th days of the treatment. Diastolic blood pressure at the baseline and on subsequent 15th, 30th, 60th and 120th days of treatment was recorded. The mean DBP was recorded at baseline was 107 ± 6.01 mmHg and on 15th, 30th , 60th and 120th days of the treatment were 100 ± 4.83, 95±3.74, 89.9±2.88 and 85.3±2.55 mmHg respectively (Table 3). There was significant decrease (p<0.0001) in diastolic blood pressure from the baseline at 15th, 30th, 60th and 120th days of the treatment. Changes in DBP were 12±2.27, 17.1±3.21 and 21.7±3.46 mmHg from baseline at 30th, 60th and 120th days of the treatment respectively.

Table 3: Effect of triple drug therapy on Blood Pressure in Hypertension with dyslipidaemia

	Base line	15th Day	30th Day	60th Day	120th Day
SBP	160±11.4	155±9.88	150± 9.97*	143± 8.05***	137± 7.54***^
DBP	107±6.01	100±4.83**	95±3.74***	89.9±2.88***^^	85.3±2.55***+

* p<0.05 vs Baseline, ** p<0.01 vs Baseline, *** p<0.0001 vs Baseline, ^ p<0.05 vs 30th Day, ^^ p<0.0001 vs 15th Day, + p<0.0001 vs 30th Day

At the end of the study period, the BP in 11 of the 14 patients in the group was within the range 140/90 mmHg. This indicated that 78.5% of patients of hypertension with dyslipidemia achieved the desired goal (140/90 mmHg) set by JNC VII.

Evaluation in Hypertensive without any complication

Systolic blood pressure was recorded at the baseline and on subsequent 15th, 30th, 60th and 120th days of treatment. The baseline Mean ± Standard Deviation (SD) SBP was 165 ± 12.2 mmHg. The mean SBP at 15th, 30th, 60th and 120th days of the treatment were 158 ± 11.2, 152±10.3, 146±9.59 and 140±8.13 mmHg respectively (Table 4). There was significant decrease (p<0.0001) in SBP from the baseline to 30th, 60th and 120th days of the treatment. Moreover there was significant decrease in systolic blood pressure from 15th day to 60th day & 30th to 120th days of treatment. Change in SBP was 19±2.61 and 25±4.07 mmHg from baseline

to 60th and 120th days of the treatment respectively.

Diastolic blood pressure at the baseline and on subsequent 15th, 30th, 60th and 120th days of treatment was recorded. The mean DBP was recorded at baseline was 106 ± 4.46 mmHg and on 15th, 30th , 60th and 120th days of the treatment were 97.7±15.2, 94.6±3.87, 89.4±3.08 and 84.4±3.09 mmHg respectively (Table 4). There was significant decrease ($p<0.0001$) in diastolic blood pressure from the baseline at 15th, 30th, 60th and 120th days of the treatment. Changes in DBP were 11.4±0.59, 16.6±1.38 and 21.6±1.37 mmHg from baseline at 30th, 60th and 120th days of the treatment.

Table 4: Effect of triple drug therapy on Blood Pressure in Hypertension with diabetes/dyslipidaemia

	Base line	15th Day	30th Day	60th Day	120th Day
SBP	165±12.2	158±11.2	152±10.3***	146±9.59***	140±8.13***^
DBP	106±4.46	97.7±15.2***	94.6±3.87***	89.4±3.08***	84.4±3.09***^

*** $p<0.0001$ vs Baseline, ^ $p<0.0001$ vs 30th Day

At the end of the study period 81.25% of patients in the hypertensive group achieved the desired goal (140/90 mmHg) set by JNC VII.

Global efficacy and safety evaluation

As per investigators assessment about overall efficacy of this triple drug fixed dose combination, which was assessed on the basis of investigators satisfaction in achieving the target blood pressure goal 140/90 mm Hg for hypertensive patients with or without dyslipidemia and 130/80 mmHg for diabetic hypertensives. Investigator assessed efficacy by using a three point scale as poor, good and excellent. Poor was for those patients, whose BP did not change from baseline, good when BP changed by 15% from the baseline and excellent for those who achieved the target BP.

At the end of therapy, 15.6% (5/32) hypertensive patients without any complications showed good, 81.25% (26/32) of patient showed excellent efficacy and 3.12% (1/32) showed poor response. In diabetic hypertensive patients 50% (7/14) showed excellent and 35.7% (5/14) showed good while 14.2% (2/14) showed poor efficacy. In hypertensive patients with dyslipidemia 78.5% (11/14) excellent and 21.42% (3/14) showed good efficacy (Table 5). As per investigators assessment regarding the tolerability, which was assessed on the basis of any reported side effect resulting into discontinuation of therapy or compelling the use of concomitant drug to subside the side effects. In this study all the patients tolerated the triple drug therapy well and no side effects were reported.

Table 5: Effect of triple drug combination on efficacy in various groups

	Excellent Efficacy	Good Efficacy	Poor Efficacy
Hypertension	81.25%	15.6%	3.12%
Hypertension with Diabetes	50.0%	35.7%	14.28%
Hypertension with Dyslipidaemia	78.5%	21.42%	Nil

DISCUSSION

The different available fixed dose combinations of angiotensin receptor blocker (ARBs) / hydrochlorothiazide (HCTZ) and ARBs/Amlodipine have been shown to be efficient and safe in reducing BP levels in patients in whom monotherapy was not sufficient to achieve BP control [12, 13]. The possible use of triple therapy with Amlodipine, Telmisartan and HCTZ in a single pill represents a step forward in improving the control of hypertension by making treatment simpler and thereby improving long-term adherence and treatment persistence. It is proven that delaying BP control by strategies of increasing dose, increases the risk of cardiovascular events in comparison with the initial use of combinations [14].

In different clinical studies it has been observed that the efficacy of the triple combination is superior to that of its components in monotherapy or in dual combination.

In this study we evaluated the efficacy and safety of triple drugs fixed dose combination of Telmisartan 40 + Amlodipine 5mg+ Hydrochlorthiazide 12.5 mg in the management of hypertension in three groups of patients, diabetic hypertensive, dyslipidemic hypertensive and hypertension without any complication. Results of our study are comparable and supporting the results of earlier studies using the triple fixed dose combination in the management of hypertension.

A study conducted by Duprez D et al [15] have shown that after 6 weeks of the treatment reduction in systolic/diastolic ABP (ambulatory blood pressure) were greater in the triple combination (ARB/CCB/ HCTZ) group than in the dual therapy (ARB/HCTZ) group (-22.0/-13.3 versus -17.4/-8.1 mmHg). Similarly in our results after 60th day of the treatment msBP (mean seated blood pressure) reduction was -19/-16.6 mmHg which is comparable to this study. Another study conducted by Destro M et al, [16] after 8 weeks of the treatment reduction in msBP was -30.5/-13.8 mmHg while in our study reduction in msBP was -19/-16 and 25/-21.6 mmHg respectively at 60th day of treatment & after the completion of study. It is clear that in the present study reduction in DBP was better than the earlier study but the reduction in SBP was less. Moreover regarding achieving the target BP goal, a study conducted by Kereiakes DJ et al [17]

observed that after 16 weeks of study 79.8% of participants reached BP goal which is comparable to present study in which 81.25% patients achieved the target blood pressure goal after the completion of the study (120 days).

Regarding the efficacy of triple drug fixed dose combination in the diabetic hypertensives, there was significant reduction in SeBP and results of the present study is comparable to the previous study conducted by Kereiakes et al [18]. After 12 weeks of the treatment SeBP (seated BP) reductions was -37.9/-22.0 mm Hg while in the present study after 120th days of the treatment it was -22/21.7 mmHg.. Moreover more number of diabetic hypertensives achieved the desired goal in this study (50%) compared to the study conducted by Kereiakes et al. (41.1 %). In dyslipidemic hypertensive's there was significant decrease ($p<0.0001$) in SBP & DBP from the baseline. In this group 78.5% of patient achieved the desired target goal (140/90 mmHg) at the end of therapy. This result is comparable to the study conducted by Roth EM et al [19] used the triple drug fixed dose combination in obese hypertensive patients where 12 weeks of treatment lead to 62% of patients achieving the target BP.

CONCLUSION:

The triple drug fixed dose combination therapy of Telmisartan, Amlodipine & hydrochlorothiazide has been shown to be effective with excellent tolerability.

ACKNOWLEDGEMENT:

The authors are thankful to Mr. Md. Wasim Siddique, M. Pharm (Pharmacology) for doing the statistical analysis & proofing the manuscript.

REFERENCES:

1. Rajeev Gupta & Soneil Guptha, Strategies for initial management of hypertension Indian. J Med Res, Nov 2010;132:531-542
2. Samir G Mallat, Houssam S Itani, Bassem Y Tanios, Current perspectives on combination therapy in the management of hypertension. Integrated Blood Pressure Control 2013;6: 69–78.
3. Ong KL, Cheung BM, Man YB, Lau CP, Lam HS. Prevalence, awareness, treatment, and control of hypertension among United States. Hypertension. 2007;49:69–75.
4. Monica Doménech Antonio Coca, Role of triple fixed combination valsartan, amlodipine and hydrochlorothiazide in controlling blood pressure.Patient Preference and Adherence 2010:4; 105–113.
5. Mancia G, De Backer G, Dominiczak A, Cifkova R, Fagard R, Germano G, Grassi G, Heagerty AM. Guidelines for the Management of arterial hypertension: The Task Force for the management of arterial hypertension of the European Society Hypertension (ESH) and the European Society of Cardiology (ESC). J Hypertens. 2007;25:1105–1187.
6. Doménech M, Coca A. Role of triple fixed combination valsartan, amlodipine and hydrochlorothiazide in controlling blood pressure. Patient Prefer Adherence. 2010 May 13;4:105-13.
7. Law MR, Wald NJ, Morris JK, Jordan RE. Value of low dose combination treatment with blood pressure lowering drugs: analysis of 354 randomised trials. BMJ. 2003;326(7404):1427

8. Wald DS, Law M, Morris JK, Bestwick JP, Wald NJ. Combinatory therapy versus monotherapy in reducing blood pressure: meta-analysis on 11,000 participants from 42 trials. Am J Med. 2009;122(3):290–300.
9. Morgan TO, Anderson AI, MacInnis RJ. ACE inhibitors, beta-blockers, calcium blockers, and diuretics for the control of systolic hypertension. Am J Hypertens. 2001;14(3):241–247.
10. De la Sierra A, Barrios V. Blood pressure control with angiotensin receptor blocker-based three-drug combinations: key trials. Adv Ther. 2012;29(5):401–415.
11. Mancia G, Laurent S, Agabiti-Rosei E, Ambrosioni E, Burnier M, Caulfield MJ, Cifkova R, Clément D. Reappraisal of European guidelines on hypertension management: a European Society of Hypertension Task Force document. J Hypertens. 2009;27(11):2121–2158.
12. Alleman Y, Fraile B, Lambert M, Barbier M, Ferber P, Izzo JL Jr. Efficacy of the combination of amlodipine and valsartan in patients with uncontrolled hypertension with previous monotherapy: The Exforge in previous failure after single therapy (EX-fast) study. J Clin Hypertens.2008;10:185–194.
13. PoolJL,GlazerR,Weinberger M, Alvarado M, Huang J, Graff. A. Comparison of valsartan/hydrochlorothiazide combination therapy at doses up to 320/25 versus monotherapy: a double blind placebo controlled study followed by long-term combination therapy in hypertensive adults. Clin Ther. 2007;29:61–73.
14. Nasser SA, Lai Z, O'Connor S, Liu X, FlacK JM. Does earlier attainment of blood pressure goal translate into fewer cardiovascular events? Curr Hypertens Rev. 2008;10:398–404.
15. Duprez D, Ferdinand K, Purkayastha D, Samuel R, Wright R. Ambulatory blood pressure response to triple therapy with an angiotensin-receptor blocker (ARB), calcium-channel blocker (CCB), and HCTZ versus dual therapy with an ARB and HCTZ. Vasc Health Risk Management 2011;7:701-8
16. Destro M, Crikelair N, Yen J, Glazer R. Triple combination therapy with amlodipine, valsartan, and hydrochlorothiazide vs dual combination therapy with amlodipine and hydrochlorothiazide for stage 2 hypertensive patients. Vasc Health Risk Manag. 2010 Sep 7;6:821-7.
17. Kereiakes DJ, Chrysant SG, Izzo JL. Long-term efficacy and safety of triple-combination therapy with olmesartan medoxomil and amlodipine besylate and hydrochlorothiazide for hypertension. J Clin Hypertens (Greenwich). 2012 Mar;14(3):149-57.
18. Kereiakes DJ, ChrysantSG,I zzoJL, LeeJ, Littlejohn T, Melino M, Fernandez V, Heyrman R; Olmesartan / amlodipine/ hydrochlorothiazide in participants with hypertension and diabetes, chronic kidney disease, or chronic cardiovascular disease: a sub-analysis of the multicenter, randomized, double-blind, parallel-group TRINITY study. Cardiovascular Diabetology 2012; 11(134):1-
19. Roth EM, Oparil S, Melino M. Olmesartan/amlodipine/hydrochlorothiazide in obese participants with hypertension: a TRINITY subanalysis. J Clin Hypertens (Greenwich). 2013 Aug;15(8):584-92.

HETEROTOPIC OSSIFICATION AFTER RECONSTRUCTIVE ACETABULAR SURGERY IN SINGLE CENTRE EXPERIENCE KHARTOUM NORTH TEACHING HOSPITAL, SUDAN

Hussam I. Elkhawad[*] MRCSEd, MD, Osman B. Mohammed[**] FRCSI, Ahmed A. Abdalla[***], MD, MRCSEd

*Orthopedic surgeon, Ministry of Health, Sudan **University of Medical Sciences & Technology, Dept. of Orthopaedics, Khartoum, Sudan, *** Gezira University, Dept. of Surgery, Wad Medani, Sudan.

Corresponding author:
Mr Ahmed Abd Elrahman Abdalla, Tel +249-919102071, e-mail: hantoub22@yahoo.com, PO Box 20

ABSTRACT:

Acetabular reconstructive surgery is a good alternative method to treat acetabular fractures. The aims of this study were to assess the incidence, clinical pattern, and the relationship between various factors and heterotopic ossification (HO). This was a retrospective study involving all cases of acetabular fractures that underwent reconstructive surgery at Khartoum North Teaching Hospital in Khartoum state during the study period, (December 2006 to December 2011). A total of 132 patients with acetabular fractures were reviewed. The complete preoperative and post-operative data, together with information on regular follow up visits at 3 months, 6months and 2 years for each patient were collected. The SPSS was used for data processing. The male to female ratio was 4.3:1. Of the 132 cases 13 (9.8%) of them developed heterotopic ossification as a complication of the surgery. This consisted of 12 (92.3%) male and 1 (7.7%) female patients. The age range of the patients was 26-50 years. Road Traffic Accidents (RTA) /occupant constitute the most common mode of trauma 46.2%. AO (**A**rbeitsgemeinschaftfür **O**steosynthesefragen foundation fracture classification) class A1 and B1 were the commonest types associated with HO. The mean hospital stay was 14 days. Male patients over 25 years of age with posterior hip dislocation, class A1 or A2, treated by posterior approach, were the highest at risk of developing Heterotopic Ossification.

***KEYWORDS**: Acetabular fractures, Heterotopic ossification*

Submitted: February 2014; Accepted: March 2014

INTRODUCTION:

Acetabular fractures, especially displaced ones, constitute serious intra-articular injuries and are often accompanied by posterior hip dislocation or other musculoskeletal injuries that may significantly affect the treatment protocol as well as the outcome [1]. Displacement of the fracture to more than 2.0mm is known to increase the danger of post-traumatic arthritis and lead to a poor functional outcome. Surgical treatment of displaced acetabular fractures is considered the treatment of choice today, because it ensures the best possible anatomical reconstruction of the joint surface, thus increasing the chances of a satisfactory functional result [2]

Heterotopic ossification was originally described in 1692 by Guy Patin, the Doyen of the Faculté de médicine de Paris [3]. Patin described a condition he observes in children and called myositis ossificans progressive. The next development in history of heterotopic ossification came in 1918 because of military injuries sustained during World War I Dejerine and Ceillier described a condition they referred to as par-aosteoarthropathy, which they observed in patients with paraplegia caused by gunshots wounds to the spinal cord [4]. The historical term of heterotopic ossification has been superseded. Ectopic ossification and myositis ossificans are used interchangeably with the term heterotopic ossification. The condition may affect the bones or the joints. Three types of heterotopic ossification have been described: myositis ossificans prograssiva (Münchmeyer disease), is an autosomal dominant (a rare pediatric metabolic disease whereby skeletal muscles ossifies); Neurogenic heterotopic ossification (this occurs as result of burns or neurologic injury); traumatic heterotopic ossification (this follows injury to tissue surrounding the bones and joint [4].

Alternatively, pathologic bone formation surrounding the bone and joint can be defined histologically. Heterotopic ossification is the formation of mature lamellar bone in nonosseous tissue, whereas myositis ossificans is a specific type of heterotopic ossification that occurs in inflammatory muscle. Both of these processes are examples of ectopic ossification, and they may coexist although they are distinct from periarticular calcification, which is deposition of pyrophosphates within the soft tissues surrounding the joints.

There is no registry in Sudan about the cases of post-operative heterotopic ossification. The condition can be easily miss-diagnosed especially in the early presentation as painful lump that have large spectrum of differential diagnosis [5].

The aims of this study were to assess the incidence, clinical pattern, and the relationship between various factors and heterotopic ossification

.

PATIENTS AND METHODS:

This was a retrospective, descriptive hospital based study involving all cases of acetabular fractures that underwent reconstructive surgery at Khartoum North Teaching Hospital in Khartoum state during the study period (December 2006 to December 2011).

Acetabular reconstructive surgery was started during the last 5-6 years in Sudan in Khartoum North Hospital as an alternative method to the old methods which were mainly conservative.

A total of 132 Patients with acetabular fractures presented to the hospital. Proper history, thorough examinations including assessment of x-rays were done on each patient. The data for each patient was recorded regardless of whether it was acetabular fracture alone or combined with hip dislocation. Appropriate treatment decision was then made for each patient. Each patient then underwent surgery with different approaches.

Regular follow-up was done for each of them by the same means and information related to functional and psychosocial recovery and x-rays of the area in a regular interval at 3 months, 6 months and 2 years collected. Duration of hospital stay preoperative and postoperative was also recorded. A total of 13 patients were found to have heterotopic ossification and they were each reviewed. AO fracture classification and its relation to the incidence observed and other factors such as age and hospital stay and others were noted. The data used in the present study were taken from the patients records retrospectively, processed and analyzed using SPSS version 17. Statistical significance difference was considered when $p<0.05$.

RESULTS:

In this study there was 107 male (81.1%) and 25 female (18.9%) patients, which is equivalent to 4.3:1 male to female ratio. The age range of all the patients was 8-70 years; the mean age was 36.2 ± 12.6 years (Mean ± SD). Of the 132 patients 13 (9.8%) of them developed heterotopic ossification as a complication of the surgery. Gender distribution of the 13 patients indicated 12 (92.3%) males and 1 (7.7%) female. Their age range was 26-50 years and their mean age was 36.4 ± 8.7 years. 30.8 % of the patients were workers. Figure 1 shows the distribution of the 13 patients according to their mode of injury. The commonest (46.2%) mode of injury in the patients with HO was RTA/Occupants

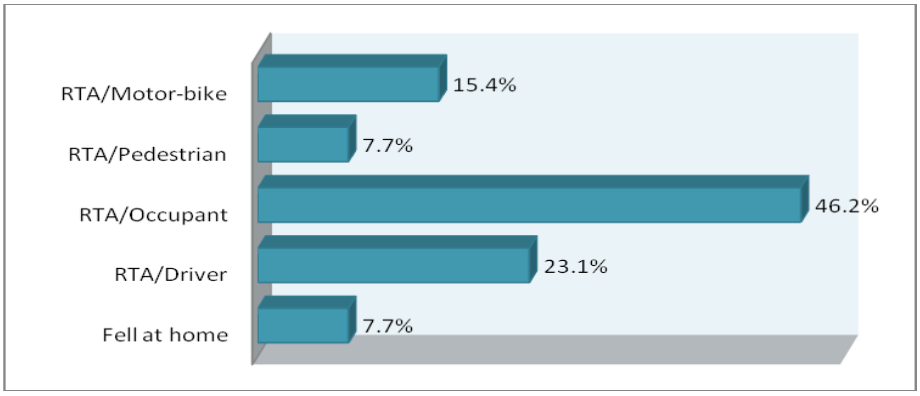


Fig 1: Mode of injury in patients with HO (n=13)

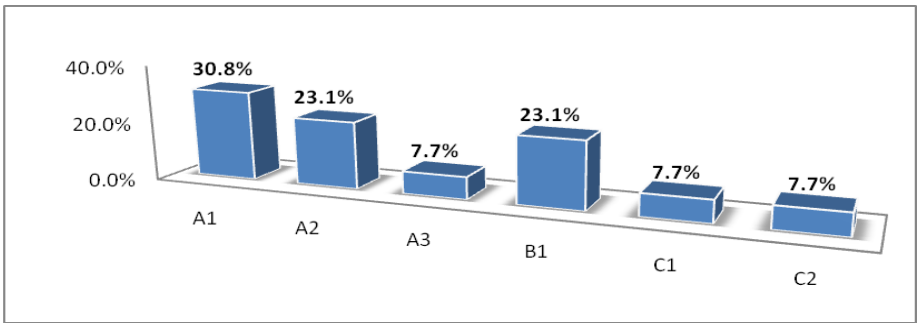


Fig 2: AO classification for HO pts (n=13)

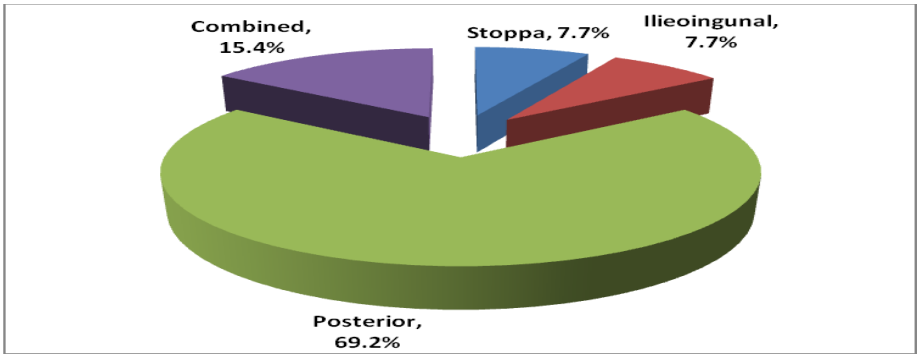


Fig 3: Surgical approach used (n=13)

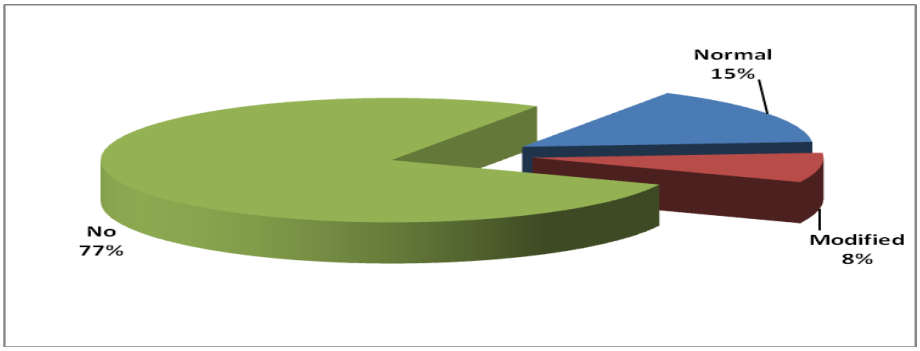


Fig 4: Functional outcome – work assessment (n=13)

There were 10 (76.9%) patients with only acetabular fractures and 3 (23.1%) patients with combination of acetabulum and pelvic fracture. Six out of the 13 pts who were HO positive had 11 types of associated injuries the highest was posterior hip dislocation in 4 pts (36.4%), upper limb injuries in 3 pts (27.3%), lower limb injuries in 2 pts (18.2%), 1 pt (9.1%) with chest injury and 1 pt (9.1%) with abdominal injury. Figure 2 shows the AO classification of the heterotopic ossification of the 13 patients.

All patients were fixed by plates and screws (13 patients 100%), the surgical approach in 9 pts (69.2%) was posterior Kocher-Langenbeck, 1 patient (7.7%) was treated by Stoppa, 1 patient (7.7%) by ilieoingunal and 2 patients (15.4%) had a combination of 2 approaches. The result is presented in fig 3.

By the time the HO became positive on the X-ray we found that 10 pts (76.9%) were positive at 3rd month of follow up, 2 pts (15.4%) at 1 year and 1 pt (7.7%) at 2nd year after the time of surgery. At the 3ed months of the follow up 7 pts (53.8%) had no pain, 6 pts (46.2 %) had mild pain and no patient had severe pain. Regarding sitting assessment 11 pts (84.6%) could sit normally and 2 pts (15.4%) had to modify the way they sit. We also found that 2 pts (15.4%) could walk normally without walking aids, 11 pts had a modified gait and using walking aids and there was no pt still in bed or using wheelchair at the 3rd month of follow up.

Four patients (30.8%) had normal sexual life, 2 patients (15.4%) had a modified sexual life, 3 patients (23.1%) did not return to any sexual activity and we had 4 patients (30.8%) still single at the 3rd month of follow up. Figure 4 shows that 2 patients (15.4%) return to the work they use to do before the injury, one patient (7.7%) modify his work and 10 patients (76.9%) still could not get back at the 3rd month of follow up. The mean hospital stay was 13.94 days, but it ranged from 3 to 64 days).

DISCUSSION:

Acetabular fractures are uncommon fractures; the average orthopedic surgeon will never see a large number of cases [6]. The aim of surgery is to preserve a painless hip with good mobility [2].

In this study the acetabular fractures were seen in younger age with mean age of 36.2 ±12.6 years. This can be compared to the results reported by Matta in 1996 [2]. So the acetabular fracture can occur at any age.

The incidence of heterotopic ossification in our study was 9.8%, which is an acceptable one when compared to the literature as it ranges from 3-69 % [7, 8, 9, 10] and in a series of 262 patients reported by Matta [2] where no prophylaxis against heterotopic ossification was administered a rate of heterotopic ossification was as high as 82%. Giannoudis et al [11] in large Meta-analysis of 23 articles found the incidence was 25.6%.

There is no enough international data about the pre-operative delay and hospital stay and incidence of heterotopic ossification. In this study we did not find a difference in pre-operative delay or hospital stay in pts with or without heterotopic ossification. Gupta et al [12] in his result claimed that the delay is major risk factor because the surgery need more dissection and stripping and 3 of his 5 pts with HO positive presented late (more than 2 weeks). No specific correlation with age was found in literature [13], but some authors indicated that it is more common in the over 30 year's age group [14]. Our finding indicates that it occurs mainly among males between 25- 50 years. The male gender has a higher risk of heterotopic ossification, generally [13, 15] around hip, it is also higher in male after arthroplasty [16, 17]. In our study we find 92.3% of our patients were males so we consider gender as risk factor.

The mode of injury in patients with heterotopic ossification was no differ from any patients with acetabular fracture which high energy trauma caused by RTAs. The mode of injury in patients with heterotopic ossification was RTA related injury in 92.4% and the mode of injury in acetabular fracture in general as described by Matta [2] was 82% due to RTA related injury.

In our study we noticed the higher incidence of heterotopic ossification in patients who had acetabular fractures only without involvement of pelvic bones and also higher in the right side; although there is no clear explanation for that either in our study or in the literature.

Many authors tried to link the presence of heterotopic ossification to associated injuries, Ghalambor N et al [13] in his report of Heterotopic ossification following operative treatment of acetabular fracture, an analysis of risk factors, mentioned the chest and abdominal injury as risk factor, also Webb et al [18] agreed on that and included head injury as risk factor. In our study 18.2% patients positive for HO had associated chest and abdominal injury. Nothing mentioned about association with upper or lower limb injury but Ghalambor N et al [13] indicated that the Injury Severity Score affects the incidence of heterotopic ossification; our results support this observation.

Regarding the association of HO with posterior hip dislocation we found that it about 36.4% of patients. According to Koval et al [8] there was 2 % incidence of HO after posterior hip dislocation, they also indicated that the incidence of HO was directly related to the amount of initial soft tissue damage. Timothy et al [19] found 44% incidence of posterior dislocation but they reported that there was no significant association between the dislocation and grade of HO. The authors however did not indicate the association between the dislocation and presence of HO regardless of the grade. In addition they indicated that approaches that do not violate the gluteal muscles are believed to be associated with a lower rate of heterotopic ossification. Our data tends to indicate that posterior hip dislocation which

result from high energy trauma, violate the gluteal muscles and need posterior approach is one of the risk factors for HO.

In patients with HO we found the common fracture pattern was A1 (31.8%) which correspond to posterior wall fracture in Letournel classification [20] and 23.1% as type A2 which correspond to posterior columns; thus over 50% patients whom HO positive had posterior element fracture can be regarded as risk factor. However, Timothy et al [19] disagreed and found no correlation between fracture type and HO. In addition Kaempffe et al [21] reported 58% incidence and also did not find a correlation with fracture type, but 26 of 28 (92.9%) of their patients with trochanteric osteotomy developed HO. The posterior approach was used in 69.2 % of pts who developed HO and the combined approach was used in 15.4% of HO positive patients.

In a meta-analysis, Giannoudis [11] found the highest risk was the iliofemoral approach followed by the Kocher- Langenbeck approach. Matityahu et al [22] also agreed on that and he also had 14 % incidence with the combined approach. The iliofemoral approach was not used for any pt in our present study. There were 76.9% cases diagnosed by the 3rd month of follow up. Timothy et al [19] also found strong correlation between the duration of follow up and incidence of HO.

All our patients had mild form of HO except one pt (still there was no ankylosis or restriction of movement), most authors suggest that the poor outcome correlate with the severity of HO [19, 21]. Fang-Yao et al [23] reported 75% good to excellent outcome in patients with heterotopic ossification.

The mean hospital stay of 13.94 days, which range from 3 to 64 days in our study, was similar to that reported by Matta [2]. He found that the mean hospital stay was 19 days (ranging 3-137) and keeps with a previous local study (24).

CONCLUSIONS:

Incidence of HO in our present study is within the range reported by others. Male pts over 25 years of age with posterior hip dislocation, class A1 or A2, treated by posterior approach, should be regarded as risky patients especially if they have past history of Heterotopic Ossification.

Competing Interests: The authors reported no conflict of interest and no funding was received for this project.

REFERENCES:

1. Letournel E., Judet R. Fractures of the acetabulum. New York, Springer, 1993
2. Matta J.M. Fractures of the acetabulum: Accuracy of reduction and clinical results in patients managed operatively within three weeks after injury. J Bone Joint Surg. 1996; 78A, 1632-45

3. Hubert Wanyura, ZygmuntStopa, Filip Brzozowski. Limited mouth opening caused by myositis ossificanstraumatica of the temporal muscle – a case report. J Stoma 2011, 64, 9: 656-666

4. Shafer DM, Bay C, Caruso DM, Foster KN. The use of eidronate disodium in the prevention of heterotopic ossification in burn patients. Burns. May 2008; 34(3):355-60

5. Samuel L. Stover, MD, Heterotopic Ossification After Spinal Cord Injury, Chapter 11 in Management of Spinal Cord Injuries, edited by R. F. Bloch, MD, PhD; M. Basbaum, MSW and Wilkins, 1986, pp 284-301

6. Hollis G. Pofter, Kenneth D. Montgomery, Carl W. Heise, David L. Helfet. MR Imaging of Acetabular Fractures: Value in Detecting Femoral Head Injury, Intraarticular Fragments, and Sciatic Nerve Injury. AJR 1993; 163:881.-886.

7. Neal B, Gray H, MacMahon S, Dunn L .Incidence of heterotopic bone formation after major hip surgery. Institute for International Health, University of Sydney, New South Wales, Australia. ANZ J Surg. 2002 Nov; 72(11):808-21

8. Koval, kennth J. Zucherman, Joseph D Handbook of fracture 3rd edition p300, p303

9. N. Briffa, R. Pearce, Hill, Bircher Outcomes of acetabular fracture fixation with ten years' follow-up. J Bone Joint Surg [Br] 2011; 93-B: 229-36.

10. Pavelka T, Houcek P. [Complications associated with the surgical treatment of acetabular fractures]. ActaChirOrthopTraumatolCech. 2009 Jun; 76(3):186-93

11. P. V. Giannoudis, Grotz, C. Papakostidis, H. Dinopoulos Operative treatment of displaced fractures of the acetabulum A META-ANALYSIS J Bone Joint Surg [Br] 2005; 87-B: 2-9.

12. Ravi K. Gupta, Harmeet Singh, Bias Dev, Rajeev Kansay, Parmanand Gupta & Sudhir Garg. Results of operative treatment of acetabular fractures from the Third World—how local factors affect the outcome International Orthopaedics (SICOT) (2009) 33:347–352

13. Ghalambor N, Matta JM, Bernstein L. Heterotopic ossification following operative treatment of acetabular fracture. An analysis of risk factorsClinOrthopRelat Res. 1994 Aug ;(305):96-105

14. Auri Bruno-Petrina, MD, PhD; Chief Editor: Denise I Campagnolo, MD, MS posttraumatic Heterotopic Ossification http://emedicine.medscape.com

15. By Andreas F. Mavrogenis, MD; Panayotis N. Soucacos, MD, FACS; Panayiotis J. Papagelopoulos, MD, DSc Heterotopic Ossification Revisited www.orthosupersite.com

16. Ritter MA, Vaughan RB. Ectopic ossification after total hip arthroplasty Predisposing factors, frequency, and effect on results. J Bone Joint Surg Am 1977; 59: 345–351

17. Ahrengart L. Periarticular heterotopic ossification after total hip arthroplasty. Risk factors and consequences. ClinOrthop 1991; 263: 49–58.

18. Webb LX, Bosse MJ, Mayo KA, Lange RH, Miller ME, Swiontkowski MF. Results in patients with craniocerebral trauma and an operatively managed acetabular fracture. J Orthop Trauma. 1990; 4:376-82.

19. TIMOTHY, KENT J. LOWRY, AND JEFFREY O. ANGLEN, Indomethacin Compared with Localized Irradiation for the Prevention of Heterotopic Ossification Following Surgical Treatment of Acetabular Fractures THE JOURNAL OF BONE & JOINT SURGERY · JBJS.ORG VOLUME 83-A DEC 2001
20. Letournel E. Acetabulum fractures: classification and management. Clin Ortho 1980; 151:81-106.
21. Kaempffe, F. A., Bone, L. B., & Border, J. R. (1991). Open reduction and internal fixation of acetabular fractures: heterotopic ossification and other complications of treatment. Journal of Orthopaedic Trauma, 5(4), 439-445. Retrieved from http://.ncbi.nlm.nih.gov/pubmed/1762005
22. Matityahu, Amir, Bruck, Nathan, miclau, Theodor. Heterotopic Ossification and acetabular Fractures Current Opinion in Orthopaedics; Feb 2006 -volume 17-issue1-pp 34-37
23. Fang-Yao Chiua, Chuan-Mu Chena, b, Wai-HeeLoa Surgical treatment of displaced acetabular fractures 72 cases followed for 10. Injury, Int. J. Care Injured 31 (2000) 181±185.
24. Osman Bakri Mohammed and Saadeldin A. Idris. Patterns of pelvic and acetabular injury among Sudanese patients. SJMS 2009; 4(4):375-84.

CASE REPORT

FOCAL FIBROUS HYPERPLASIA: A CASE REPORT

Asaf Aboobakker, *Laxmikanth Chatra, Prashanth Shenai, Veena KM, Prasanna Kumar Rao, Rachana V Prabhu, TashikaKushraj and Prathima Shetty

Department of Oral Medicine and Radiology, Yenepoya Dental College, Yenepoya University, Mangalore, Karnataka, India

*Corresponding author: Professor Laxmikanth Chatra
Department of Oral Medicine and Radiology Yenepoya Dental College Yenepoya University Deralakatte Post, Deralakatte, Mangalore PIN– 575018, Karnataka, India ydcomr@gmail.com

Running Title: Fibrous Hyperplasia

ABSTRACT:

Fibromas are the benign tumours that are most commonly encountered among the oral soft tissue lesions. They are mostly seen as a protective mechanism of the mucosa towards chronic irritation. They present clinically as a round or ovoid, soft to firm in consistency, exophytic growth, mostly pale pink in colour with smooth surface. These lesions are asymptomatic and do not require any treatment until bothersome to the patient. The clinical features, histopathological features and treatment of an irritational fibroma occurring on the hard palate of a 32 year old female are presented.

KEYWORDS: *Irritational Fibroma, Hard palate, Chronic irritation.*

Submitted: December 2013: Accepted February 2014

INTRODUCTION:

Fibroma commonly known as Traumatic fibroma, Irritational fibroma or Focal fibrous hyperplasia is a sub mucosal response to some chronic irritation or trauma inflicted from the prostheses or adjacent tooth [1]. It was first reported as fibrous polyp and polypus in 1846 [1]. It is usually caused by irritants such as calculi, overhanging margins, restorations, foreign bodies, chronic biting, margins of caries, sharp spicules of bones and over extended borders of appliances [2]. This article discusses a case report of Fibroma located on the hard palate. The clinical features, histopathological features, various excisional modalities are also discussed. The ethical clearance for the publication of this case report was obtained from the university ethics committee.

CASE REPORT:

A 32 year old female patient visited the dental OPD with the chief complaint of spacing between anterior teeth since one year. She gave the history that she noticed the space increasing since one year and Aesthetic concern. No associated symptoms were reported. She was in apparent good health and had undergone uneventful extraction of mandibular left second molar ten years back. She did not have any deleterious habits and cleaned her teeth once daily with tooth brush and tooth paste. She was predominantly a non – vegetarian.

On general physical examination, she was conscious and co – operative, moderately built and nourished, well oriented in time, place and person. All her vital signs were within the normal limits. There were no signs of pallor, icterus, cyanosis, clubbing and oedema. On extra oral examination, she had incompetent lips, with convex profile. No gross asymmetry of the face was seen. Her ears and nose showed no abnormality except for her squint eyes. There was no abnormality detected in the Temperomandibular Joint (TMJ) and lymph node examination. On intra oral examination of the soft tissues, the buccal mucosa, labial mucosa, tongue, floor of the mouth, showed no abnormalities except for palate which showed a growth. Examination of the gingival status revealed her oral hygiene status to be fair with moderate stains and calculus deposits. On hard tissue examination maxillary right and left third molar was decayed, fractured restoration in relation to mandibular right first molar and missing mandibular left first and second molar were evident.

On local examination, on inspection a solitary, nodular swelling of size 0.5 cm in diameter was seen on the right side of the hard palate in relation tomaxillary right second premolar region (Fig.1). The surface of the swelling appears smooth and colour same as that of adjacent normal mucosa. Margins appear to be smooth. The lesion appears pedunculated and raised above the mucosal surface. No ulcer, sinus, discharge evident. Inspectory finding regarding the site, size and location of the lesion were confirmed on palpation. The lesion was non-tender, soft to firm in consistency, non-fluctuant, non-compressible, non-reducible, non-pulsatile, and non-translucent. The adjacent mucosa was normal.

Based on the clinical features a provisional diagnosis of Midline Diastema between maxillary central incisors, chronic generalized gingivitis, dental caries in relation to maxillary right and left third molar, fractured restoration if mandibular right first molar, partially edentulous area in relation to mandibular left first and second molar, irritational fibroma on the hard palate was made. Excision of the lesion was performed and the tissue was sent for histopathological examination.

A histopathological report of atrophic parakeratinized stratified squamous epithelium

with short blunt rete ridges. Underlying connective tissue found to be densely collagenous showing wavy bundles of collagen fibres and a few Red Blood Cells (RBC) filled blood vessels (Fig.2). Based on the clinical features and histopathological features a final diagnosis of fibroma was made. The patient was further referred for Oral prophylaxis, Restoration of 18, 28 and 46; orthodontic correction for midline diastema and prosthetic rehabilitation of the missing teeth. Patient was recalled and review was done.

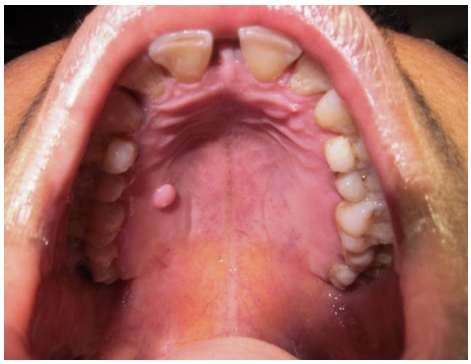


Figure 1: Location and size of the lesion.

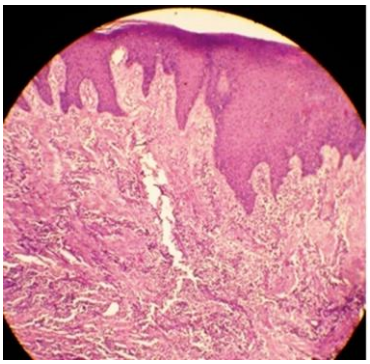


Figure 2: Microscopic findings of the excised mass

DISCUSSION:

Fibroma, oral fibroma or fibromatosis fibroma is the most commonly occurring soft tissue tumour in the oral mucosa [3]. It is of connective tissue origin. Although, the clinical appearance and pathogenesis of this entity is described better by the term focal fibrous hyperplasia, it is not commonly used [3].Ina study conducted on 300 benign tumors on the oral cavity160 (53.3%) were diagnosed as Fibroma [4]. Of the 160 cases of Fibroma, 105 (65.6%) were females and 55 (34.4%) were males. It is a reactive hyperplasia of the connective tissue rather than a neoplasm.

Clinically, this entity is seen to have a higher incidence in females than males in the third to sixth decade of their life [5]. In our case the patient was female in her 4th decade. It is seen more commonly occurring on the buccal mucosa along the occlusal line followed by labial mucosa, gingiva and palate. Biting of the cheek is considered to be one of the reasons for the occurrence of fibroma along the occlusal line [6]. But in the present case it was seen on the hard palate which is rare.

Fibroma lesion presents as a round to ovoid asymptomatic nodule which has a smooth texture and usually pale pink in colour. It may also appear white due to hyperkeratosis due to continued irritation. It is firm in consistency and usually pedunculated, although sessile cases have also been reported. They present as

asymptomatic mass unless traumatized [4]. In the present case, the clinical features were similar except for the hyperkeratosis.

Histopathologicaly, fibroma show masses of fibrous connective tissue which is lined by stratified squamous epithelium. Underlying connective tissue is usually dense and collagenized and these collagen bundles are seen arranged in radiating, circular or haphazard fashion [7]. Hyperkeratosis may be seen associated from secondary trauma. The connective tissue also shows few blood vessels and few inflammatory cells mostly lymphocytes and plasma cells which are usually seen beneath the epithelial cells [8]. Similar features were also seen in our case.

Mostly these lesions do not require any treatment unless it becomes symptomatic due to trauma or interferes with the occlusion. Excision of the lesion is the treatment of choice [9]. Recently, Soft tissue lasers are also being used for the excision of the fibromas [10]. In a study, excision was done using Nd: YAP laser and Photo-biomodulation using In-Ga-AIP which showed safe, quick procedure and better healing than conventional surgery [10]. However, in this case conventional excision of the lesion was done using scalpel. Recurrence of the lesion is rare [8].

CONCLUSION:

Although Fibroma is a common entity among the benign tumors of the oral cavity, occurrence in the hard palate is rare. In addition, these lesions tend to mimic the clinically pyogenic granuloma and Peripheral ossifying fibroma especially when occurring on the gingiva. Thus, proper diagnosis, management and excision of these lesions along with histopathological examination are of utmost importance due to the occurrence and similar presentations of neoplastic growths though the incidence is rare.

REFERENCES:

1. Singh A, Vengal M, Patil N, Sachdeva SK. Traumatic fibroma- a saga of reaction against irritation. Dental Impact 2012; 4: 49-52.
2. Alam MN, Chandrasekaran SC, Valiathan M. Fibroma of the Gingiva: A Case Report of a 20 year old lesion. International Journal of Contemporary Dentistry 2010; 1: 107-9.
3. Mathur LK, Bhalodi AP, Manohar B, Bhatia A, Rai N, Mathur A. Focal Fibrous Hyperplasia: A Case Report. International Journal of Dental Clinics 2010; 2: 56-7.
4. Halim DS, Pohchi A, Yi PE. The Prevalence of Fibroma in Oral MucousaAmong Patient Attending USM Dental Clinic Year 2006 – 2010. The Indonesian J Dent Res 2010;1:61-6.
5. Seyedmajidi M, Hamzehpoor M, Bagherimoghaddam S. Localized lesions of Oral Cavity: A Clincopathological study of 107 cases. Res. J. Med. Sci. 2011; 5: 67-72.
6. Madhusudan AS, Gupta S, Sowmya GV. Focal Fibrous Hyperplasia: Report of two

Cases. International Journal of Dental Clinics 2011; 3: 111-2.

7. Toida M, Murakami T, Kato K, Kusunoki Y, Yasuda S, Fujitsuka H. Irritation Fibroma of the Oral Mucousa: A Clinicopathological study of 129 lesions in 124 cases. Oral Med Pathol 2001; 6: 91 – 4.
8. Kolte AP, Kolte RA, Shrirao TS. Focal Fibrous Overgrowths: A Case Series and review of literature. Contemp Clin Dent 2010; 1: 271-4.
9. Reddy V, Saxena S, Saxena S, Reddy M. Reactive hyperplastic lesions of the oral cavity: A ten year observational study on North Indian Population. J Clin Exp Dent. 2012; 4: e1 36-40.
10. Pedron IG, Ramalho KM, Moreira LA, Freitas PM. Association of Two Lasers in the Treatment of Traumatic Fibroma: Excision with Nd: YAP laser and Photobiomodulation Using InGaAIP: A Case report. J Oral Laser Applications 2009; 9: 49-53.

ACUTE AND CHRONIC EFFECTS OF BETEL NUT QUID CHEWING ON THE CARDIOVASCULAR SYSTEM AND ITS ROLE AS A CARDIOVASCULAR RISK FACTOR: A REVIEW

Rodney Itaki, MBBS, B. Med. Sc.

Division of Pathology, School of Medicine and Health Sciences, University of Papua New Guinea

Email: itaki7@gmail.com

ABSTRACT:

Betel nut quid chewing is a common cultural practice in the Asia-Pacific region. Much is known about betel quid and its association to pre-malignant and malignant lesions in the oral cavity but little is known about its link to other poor health comes. There is a growing body of evidence to suggest that betel nut quid chewing may play a role in the development of cardiovascular abnormalities in pre-disposed individuals. This review examines some of the current available literature suggesting betel nut quid chewing as cardiovascular risk factor.

KEYWORDS*: betel nut quid chewing, cardiovascular risk.*

Submitted January 2014; Accepted March 2014

INTRODUCTION:

Betel nut (areca catechu) quid chewing is a common habit in the Asia-Pacific Region, including Papua New Guinea (PNG) although the method of chewing varies between countries [1, 2, 3]. The habit can be addictive and its popularity is thought to be due to the mild euphoric effect and mental alertness it produces [4, 5, 6]. In PNG, betel nut (areca nut) is chewed with piper betle inflorescence dipped in lime powder without addition of tobacco or spices, as practiced in Asian countries [3, 7]. The betel quid that forms is kept in the mouth and chewed. Lime (calcium hydroxide) used for chewing betel nut is made from heated sea shells or white corals [3].

The link between betel nut chewing and oral pre-malignant and malignant lesions is well established [7] and betel quid chewing is regarded as carcinogenic by the World Health Organization [7, 8]. Betel quid chewing has also been linked to aggravation of asthma [9, 10, 11], reduced tendon reflexes [3], poor pregnancy outcome [12, 13], adverse effect on

Vitamin D metabolism [14], manganese toxicity [15] and abnormal psychological and EEG profile [16]. Betel quid chewing has even been proposed to be a possible treatment for schizophrenia [17, 18, 19] although the data to support this is scanty. Betel nut has been used as a traditional herbal medicine. However, a study from Cambodia has raised the possibility of transmission of infectious diseases through its use [20]. The possible role of betel quid chewing in the transmission of tuberculosis has been discussed in various forums in PNG but no study has been done to support this claim.

There is growing epidemiological evidence that betel nut chewing is associated with ischaemic heart disease [21, 22, 23, 24], hypertension [2, 25], metabolic syndrome [26, 27, 28] and poor glycaemic control in diabetes [29]. Earlier study done by Kevau et al [30] and anecdotal evidence from Port Moresby General Hospital suggests that betel quid chewing may have an effect on the cardiovascular system. Betel quid chewing is increasingly being recognized as cardiovascular risk factor in the Asia-Pacific region where this habit is commonly practiced [21, 22, 24, 30]. This paper reviews current literature on the acute and chronic effects of betel quid chewing on the cardiovascular system and its potential roles as a cardiovascular risk factor.

Pharmacological action of betel nut quid:

Betel nut contains numerous compounds but the four main active ingredients are alkaloids; arecoline, arecaidine, guvacoline and guvacine [16]. The main alkaloid producing pharmacological effects is thought to be arecoline, a naturally occurring acetylcholine agonist acting on muscarinic and nicotinic receptors [16, 31]. Acetylcholine is a neurotransmitter in the autonomic nervous system; acetylcholinesterase converts it to acetate and choline [16, 31, 32]. Apart from the autonomic nervous system this enzyme is also present on red blood cell membranes where it is used to monitor cholinesterase-inhibiting properties of pesticides and toxicity among agricultural workers [32, 33]. Laboratory experimental evidence suggests that arecoline may be broken down by acetylcholinestarase and carboxylestarase [34]. Pharmacological effects of betel nut chewing are thought to be due to parasympathetic stimulation which includes euphoria, central nervous system stimulation, vertigo, excessive salivation, miosis and tremor [16, 31].

Effect of betel quid chewing on heart rate:

Betel quid chewing causes tachycardia [35]. This effect is transient and lasts for 17 minutes regardless of the whether the chewer is a novice, occasional chewer or a habitual chewer [35]. It has also been suggested that the rise in heart rate is dose dependent [36]. Consumption of small amount of betel quid

results in an increase in heart rate, suggesting sympathetic stimulation while chewing large amounts reduces the RR interval variation suggesting parasympathetic stimulation [36]. Nicotinic receptors are present on adrenal medulla [31] and stimulation of nicotinic receptors by arecoline may release catecholamines into the circulation thus causing tachycardia [2,19]. Betel quid chewing has been shown to increase basal secretions of catecholamine [2] and it is possible that the tachycardia observed is due to this mechanism. In the presence of lime, arecoline is hydrolyzed to arecaidine which has sympathetic effects via inhibition of gamma-aminobutyric acid (GABA) uptake [4]. Piper betle inflorescence also has been shown to release catecholamines in-vitro [4, 19] which may also explain the transient rise in heart rate.

Acute transient rise in heart rate induced by chewing betel quid may be a risk factor for cardiac arrhythmias in predisposed patients [37, 38]. Epidemiological data from Taiwan also suggest this possibility [37]. Betel nut chewing has also been implicated in acute myocardial ischaemia [30, 39]. These earlier observations are now supported by epidemiological data showing association between betel nut chewing and cardiovascular death [21, 22, 23, 24, 40].

Effect betel quid chewing on blood pressure:

Blood pressure response is variable after chewing betel quid. Chu observed that only the systolic pressure increases in first time chewers [35]. In another study hypertension was observed in chronic betel quid chewers [2]. More studies are needed to explain these differences in the actions of betel nut chewing.

Betel quid chewing and ischaemic heart disease:

One of the first reported cases of sudden death from myocardial infarction linked to betel quid chewing was by Hung and Deng in 1998 [39]. It has been proposed that arecoline may play a role in coronary artery spasm due to parasympathetic effects on abnormal endothelium [39]. It has also been proposed that coronary artery ischemia may be induced by arecoline similar to the action of acetylcholine [41]. More recently it has been shown that long term betel nut chewing increases coronary artery disease in Taiwanese men as an independent risk factor [22]. Acetylcholine has been shown to cause vasodilation in coronary arteries with normal endothelium but vasoconstriction in atheroslerosed coronary arteries [41, 42, 43]. It appears therefore that the vasodilator effect of acetylcholine on coronary arteries is dependent on an intact normal endothelium. Arecoline from betel nut may be acting via a similar mechanism in normal coronary arteries but paradoxically induce vasoconstriction in

arteroslerosed coronary arteries [30]. This may be the underlying pathological mechanism explaining sudden death after chewing betel nut in high risk individuals [39]. Detailed laboratory based studies are needed to test this hypothesis.

Piper betle inflorescence has been shown in experiments to increase catecholamines [4, 19] but it is not clear if the sympathetic effect of betel quid chewing has a direct cause-effect relationship to clinical or sub-clinical ischaemic heart disease. Betel quid chewing has been associated with clinical and subclinical ischaemic heart disease in Taiwan [24] but proposed mechanisms has not been elucidated.

Betel quid chewing and its association with metabolic syndrome and diabetes:

Betel quid chewing has been strongly associated with central obesity [37] and the habit also has a negative effect on blood glucose control in diabetics [29]. It is unclear whether betel nut quid accelerates the development of ischaemic heart disease in diabetics. Using computational modeling arecoline was shown to inhibit endocytosis of low density lipoprotein (LDL) cholesterol by blocking LDL receptor function and also so prevent LDL cholesterol uptake by liver by interfering with high density lipoprotein (HDL) receptor [44]. The aforementioned mechanisms may possibly contribute to artherosclerosis [44]. Further, in-vitro studies have also provided evidence that arecoline induced fat cell dysfunction, which may contribute to Hyperlipidemia, hyperglycemia and insulin resistance; hence contributing towards development of metabolic syndrome [28].

Experimental models to study betel quid effect on cardiovascular system:

Most of the data currently available showing association between betel quid chewing and cardiovascular disease has been through population based cohort studies [40]. There is a need for laboratory based studies to determine the mechanism involved. One way is to use a rat model to observe dose dependent responses [40]. Using tissue perfusion systems to see effects on isolated arteries to compare responses from normal and abnormal endothelium is another way. Frog models can also be used to see dose dependant heart rate responses. Observation of blood pressure responses in-vitro will be technically challenging. Although there is mounting epidemiological data linking betel quid chewing and negative cardiovascular effects, the underlying mechanisms are yet to be elucidated.

CONCLUSIONS:

Betel quid chewing is a possible cardiovascular risk factor in susceptible individuals. Although there is mounting data from population based

cohort studies to support this link, there is lack of in-vitro studies to determine the underlying mechanisms.

REFERENCES:

1. Senn M, Baiwog F, Winmai J, Mueller I, Rogerson S, Senn N. Betel nut chewing during pregnancy, Madang Province, PNG. Drug Alcohol Depend 2009;105(1-2):126-131.
2. Heck J.E, Marcotte E.L, Argos M, Parvez F, Ahmed A, Islam T, Sawar G, Hasan R, Ahsan H, Chen Y. Betel quid chewing in rural Bangladesh: prevalence, predictors and relationship to blood pressure. Int J Epi 2012; 41:462-471.
3. Harrison O, Linjim W, Adeniyi K.O. The effect of betel nut chewing on the Achilles tendon reflex time. PNG Med J 2006;49(1-2):47-51.
4. Chu N.S. Effects of betel nut chewing on the central and autonomic nervous systems. J Bio Med Sci 2001;8 (3):229-236.
5. Gupta P.C, Warnakulasuriya S. Global epidemiology of areca nut usage. Addic Biol 2002;7:77-83.
6. Gupta P.C, Ray C.S. Epidemiology of betel quid usage. Ann Acad Med Singapore 2004;33 (suppl):31S-36S.
7. Sharan R.N, Mehrotra R, Choudhury Y, Asotra K. Association of betel nut with carcinogenesis: revisit with a clinical perspective. PLoS One 2012;7(8):e42759.
8. World Health Organisation. Betel quid and areca nut chewing and some areca nut derived nitrosamines. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans 2004;85:1-301.
9. Kiyingi K.S. Betel nut chewing may aggravate asthma. PNG Med J 1991;34(2):117-121.
10. Taylor RF, al-Jarad N, John LM, Conroy DM, Barnes NC. Betel nut chewing and asthma. Lancet 1992;339 (8802):1134-1136.
11. Kiyingi S, Saweri A. Betel nut chewing causes bronchoconstriction in some asthma patients. PNG Med J 1994;37(2):90-99.
12. Garcia-Algar O, Vall O, Alameda F, Puig C, Pellegrini M, Pacifici R, Pichini S. Prenatal exposure to arecoline (areca nut alkaloid) and birth outcomes. Arch Dis Child Fetal Neonatal Ed. 2005;90:F276-F277. doi:10.1136/adc.2004.061325.
13. Senn M, Baiwog F, Winmai J, Mueller I, Rogerson S, Senn N. Betel nut during pregnancy, Madang Pregnancy, Papua New Guinea. Drug Alcohol Depend 2009;105 (1-2):126-131.
14. Ogunkolade W.B, Boucher B.J, Bustin S.A, Burrin J.M, Noonan K, Mannan N, Hitman G.A. Vitamin D metabolism in peripheral blood mononuclear cells is influenced by chewing betel nut (Areca catechu) and vitamin D status. J Clini Endo Metab 2006;91 (7);2612-2617.
15. Al-Rmali S.W, Jenkins R.O, Haris P.I. Betel nut chewing as source of manganese exposure: total daily intake of manganese in a Bangladeshi population. BMC Pub H 2011;11:85.
16. Osborne P.G, Chou T.S, Shen T.W. Characterization of the psychological, physiological and EEG profile of acute betel quid intoxication in Naïve subjects. PLos One 2011;6(8):e23874.

17. Burton G. Burton- Bradley. Betel nut chewing in retrospect. PNG Med J 1978 21(3);236-241.
18. Roger J. Sullivan,John S. Allen, Caleb Otto, Josepha Tiobech, Karen Niro. Effects of chewing betel nut (Areca catechu) on the symptoms of people with schizophrenia in Palau, Micronesia. British Journal of Psychiatry 2000;177:174-178.
19. Boucher B.J, Mannan N. Metabolic effects of the consumption of areca catechu. Addic Biol 2002;7:103-110.
20. Singh P.N, Natto Z, Yel D, Job J, Knutsen S. Betel quid use in relation to infectious disease outcomes in Cambodia. Int J Infect Dis 2012;16(4):e262-e267.
21. Lin W.Y, Chiu T.Y, Lee L.T, Lin C.C, Huang C.Y, Huang K.C. Betel nut chewing is associated with increased risk of cardiovascular disease and all-cause mortality in Taiwanese men. Am J Clin Nutr 2008;87:1204-1211.
22. Tsai W.C, Wu M.T, Wang G.J, Lee K.T, Lee C.H, Lu Y.H, Yen H.W, Chu C.S, Chen T.Y, Lin H.T, Su. H.M, Hsu P.C, Cheng H.K, Duh T.H, Ko C.Y, Sheu S.H, Lai W.T. Chewing areca nut increases the risk of coronary artery disease in Taiwanese men: a case-control study. BMC Pub H 2012;12:162.
23. Yen A.M.F, Chan L.S, Chiu Y.H, Boucher B.J, Chen T.H.H. A prospective community-population-registry-based cohort study of the association between betel quid chewing and cardiovascular disease in men in Taiwan (KCIS no.19). Am J Clin Nutr 2008;87:70-78.
24. Tseng C.H. Betel nut chewing and subclinical ischaemic heart disease in diabetic patients. Cardio Res Pract 2011;doi:10.4061/2011/451489.
25. Shafiqe K, Mirza S.S, Vart P, Memon A.R, Arain M.I, Tareen M.F, Haq U.Z. Areca nut chewing and systemic inflammation: evidence of a common pathway for systemic diseases. J Inflam 2012;9:2.
26. Lin W.H, Pi-Sunyer F.X, Liu C.S, Li T.C, Li C.I, Huang C.Y, Lin C.C. Betel nut chewing is strongly associated with general and central obesity in Chinese middle aged adults. Obesity 2009;17(6):1247-1254.
27. Shafique K, Zafar M, Ahmed Z, Khan N.A, Mughal M.A, Imtiaz F. Areca nut chewing and metabolic syndrome: evidence of a harmful relationship. Nutr J 2013;12:67.
28. Hsu H.F, Tsou T.C, Chao H.R, Shy C.G, Kuo Y.T, Tsai F.Y, Yeh S.C, Ko Y.C. Effects of arecoline on adipogenesis, lipolysis, and glucose uptake of adipocytes-A possible role of betel quid chewing in metabolic syndrome. Toxicol Appl Pharmacol 2010;245(3):370-377.
29. Amos L.B, Margis D. Betel nut chewing: a contributing factor to the poor glycaemic control in diabetic patients attending Port Moresby General Hospital, Papua New Guinea. PNG Med J 2005;48(3-4):174-182.
30. Kevau I.H, Miam B, Jothimanikam J, Urae G, Itaki R, Mari, R, Wagiebu J, Sengupta A. Betel nut causes paradoxical vasoconstriction in patients with coronary artery disease – an exciting new discovering in Papua New Guinea with important clinical implications. Cardiac Society of Australia and New Zealand, 47th conference presentation, 1998.

31. Joan H. Brown, Palmer Talyor. In: Goodman and Gilman's The Pharmacological Basis of Therapeutics. 11th ed. New York: MacGraw-Hill, 2006.
32. Suarez-Lopez J.R, Jacobs Jr D.R, Himes J.H, Alexander B.H, Lazovich D, Gunnar M. Lower acetylcholine esterase activity among children living with flower plantation workers. Environ Res 2012;114:53-59.
33. Magnotti R.A, Eberly J.P, Quarm D.E.A, McConnell R,S. Measurement of acetylcholinesterase in erythrocytes in the field. Clin Chem 1987;33(10):1731-1735.
34. Giri S, Idle J.R, Chen C, Zabriskie M, Krausz K.W, Gonzalez F.J. A metabolomics approach to the metabolism of the areca nut alkaloids arecoline and arecaidine in the mouse. Chem Res Toxicol 2006; 19 (6): 818-827.
35. Chu N.S. Cardiovascular responses to betel nut chewing. J Formos Med Assoc 1993;92(9):835-837.
36. Chu N.S. Effect of betel nut chewing on RR interval variation. J Formos Med Assoc 1995;94(3):106-110.
37. Chiang W.T, Yang C.C, Deng J.F, Bullard M. Cardiac arrhythmia and betel nut chewing-is there a causal effect. Vet Hum Toxicol 1998;40(5):287-289.
38. Tsai W.H, Chen C.Y, Kuo H.F, Wu M.T, Tang W.H, Chu C.S, Lin T.H, Su H.M, Hsu P.C, Jhuo S.T, Lin M.Y, Lee K.T, Sheu S.H, Lai W.T. Areca nut chewing and risk of atrial fibrillation in Taiwanese men: A nationwide ecological study. Int J Med Sci 2013;10(7):804-811.
39. Huang D.Z, Deng J.F. Acute myocardial infarction temporally related to betel nut chewing. Vet Hum Toxicol 1998;40(1):25-28.
40. Iqbal M.P, Mehboobali N, Haider G, Pervez S, Azam I. Effects of betel nut on cardiovascular risk factors in a rat model. BMC Cardio Dis 2012;12:94.
41. Ludmer P.L, Selwyn A.P, Shook T.L, Wayne RR, Mudge G.H, Alexander R.W, Ganz P. Paradoxical vasoconstriction induced by acetylcholine in atherosclerotic coronary arteries. New Engl J Med 1986;315(14):1046-1051.
42. Vita J.A, Treasure T.B, Nabel E.G, McLenachan J.M, Fish R.D, Yeung A.C, Vekshtein V.I, Selwyn P, Ganz P. Coronary vasomotor response to acetylcholine relates to risk factors for coronary artery disease. Circulation 1990;81:491-497.
43. Egashira K, Suzuki S, Hirooka Y, Kai H, Sugimachi M, Imaizumi T, Tekeshita A. Impaired endothelium dependent vasodilation of large epicardial and resistance coronary arteries in patients with essential hypertension; Different responses to acetylcholine and substance P. Hypertension 1995;25:201-206.
44. Choudhurry M.D, Chetia P, Choudhurry K.D, Talukdar A, Datta-choudhari M. Atherogenic effect of arecoline: A computational study. Bioinformation 2012;8(5):229-239.

ENDOCRINE AND METABOLIC CONSEQUENCES OF BEING BORN SMALL- OR LARGE-FOR-GESTATIONAL AGE: A REVIEW

Alphonsus N. Onyiriuka; FMCPaed

Endocrine and Metabolic Unit, Department of Child Health, University of Benin Teaching Hospital, Benin City, Nigeria.

E-mail: alpndiony@yahoo.com; didiruka@gmail.com.

ABSTRACT:

In this review article, the genetics of size at birth, prenatal metabolic programming and the endocrine and metabolic consequences of abnormal size at birth are discussed. In addition, the relevance of fetal origin of adult disease in developing countries and the public health implication as well as future perspectives are also discussed. Being born either small- or large-for-gestational age affects such children and adults in several ways. These include increased risk of type 2 diabetes mellitus, metabolic syndrome, oxidative stress, persistent reduction in growth, cardiovascular disease, osteoporosis and premature pubarche as well as adrenarche. Individuals with abnormal size at birth who experienced rapid growth in the first three years of life have the greatest risk for future metabolic abnormalities. The mechanisms involved in prenatal (fetal) metabolic programming in infants with abnormal size at birth are just beginning to be explored. Both the "thrifty genes" and the "thrifty phenotype" could result in adverse health consequences later in life on exposure to plentiful nutrition. The most important epigenetic reactions affecting genetic transcription are acetylation and methylation. However, the major challenge at this point in time is to link such alterations with modifications in gene expression and ultimately, with metabolic abnormalities encountered in adult life. Thus, developmental origins of health and disease (DOHaD) represent a relatively new frontier of research and with time, some of the discrepancies may be resolved.

KEYWORDS: *Endocrine effects, metabolic effects, birth size, large-for-gestational age, small-for-gestational age*

Submitted January 2014; Accepted March 2014

INTRODUCTION:

Size at birth is defined by measurement of weight, length or both and is influenced by the quality of nutrition during fetal life [1,2]. Several decades ago, concerns over abnormal size at birth centered on survival and health of the offspring in the immediate neonatal period. Today, the emphasis has shifted to long-term consequences of such abnormal fetal growth, and ultimately size at birth. In man, diseases in adulthood are increasingly associated with growth patterns in early life, implicating early nutrition as an underlying mechanism [3].

Genetics of size at birth:

Birth size is influenced by genetic and environmental factors [1]. Reports from epidemiological studies indicate that genetic factors account for 38 to 80 percent of birth weight variance with environmental influences accounting for the remainder [4,5]. The relative contributions of genetic and environmental factors vary, not only from individual to individual, but also between populations [1]. Fetal genes have a greater influence on birth size variance (18% - 69.4%) than parental genes (3 % - 20%) [4,5]. Several animal knockout experiments have identified the important role played by insulin-like growth factor 1 (IGF-I), IGF-II, insulin and their respective receptors in regulating fetal growth and ultimately, size at birth [6-8].

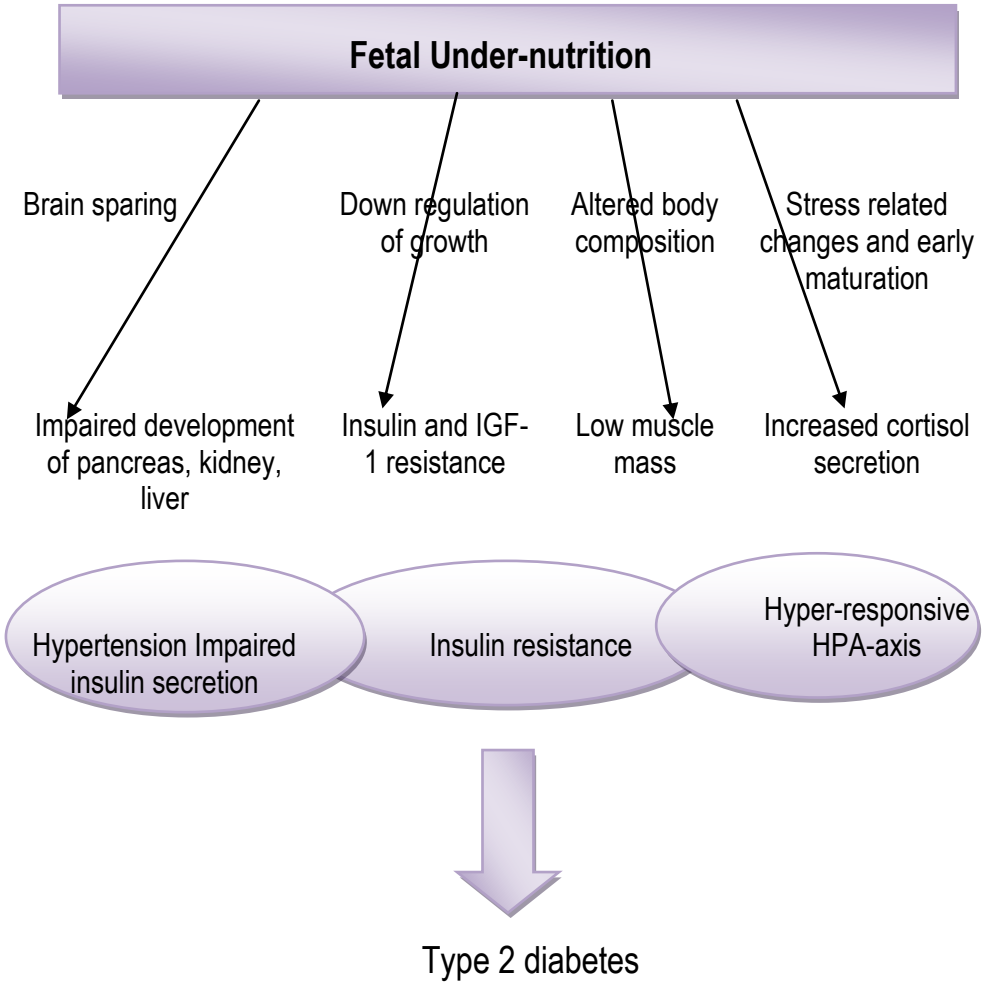
Prenatal (fetal) metabolic programming:

The term “programming” refers to a permanent change in the structure and function of an organism as a result of a transient stimulus or insult occurring at a critical or sensitive period of development [9]. The mechanisms by which programming could occur at a tissue or cellular level may be categorized into structural changes or changes in cellular homeostatic process. An example of a structural change which has been described in low birth weight infants is reduction in pancreatic beta cell mass, which could increase disease risk later, if it persists [10]. Growth-restricted human fetuses have an altered endocrine profile. For instance, they have low circulating insulin and insulin-like growth factor (IGF) concentrations [11]; an example of changes in cellular homeostatic process. Programming is well described in endocrine systems. Some examples include (i) female rats given testosterone in the first 4 days of life fail to develop normal pattern of female sexual behaviour [11]; (ii) vaginal adenocarcinoma in young women has been linked to a transient intrauterine exposure to diethylstilbesterol and shown to have a long latent period between exposure and disease [12]. Thus, both represent

programming stimuli in fetal life. The hypothalamus has been implicated as the key site that is programmed by transient changes in prenatal endocrine status [13].

The existence of prenatal metabolic programming is captured in the concept represented by the terms “thrifty phenotype” coined by Hales and Barker [14] and “thrifty genotype” coined by Neel [15]. Neel suggested that type 2 diabetes is caused by ‘thrifty’ genes which were selected for during man’s phylogenetic evolution when food supply was precarious, but became diabetogenic in a modern setting of plentiful nutrition. In contrast, the thrifty phenotype hypothesis (also called the small-baby-syndrome) describes the metabolic adaptations adopted as a survival strategy by an under-nourished fetus. In later life, such a phenotype must be thrifty and help affected individuals to cope better with conditions of food shortage. However, under conditions of abundant food intake and decreased energy expenditure, this advantage turns into a disadvantage, leading to metabolic syndrome, type 2 diabetes and cardiovascular diseases. Thus, both the thrifty genes and the thrifty phenotype could become detrimental on exposure to plentiful nutrition [12,16]. This idea is widely accepted and is a source of concern for societies undergoing a transition from sparse to better nutrition [17]. Risk factors of thrifty phenotype include advanced maternal age and placental insufficiency [18]. The gestational age at which the nutritional deprivation occurred influence the metabolic consequences. For instance, if fetal exposure to nutritional deprivation occurs during early pregnancy it will affect lipid metabolism (associated with higher LDL/HDL cholesterol concentrations and (in women) higher BMI and waist circumference), but if it occurs in late pregnancy, it will affect glucose metabolism (associated with glucose intolerance, insulin resistance and some increase in type 2 diabetes) [19]. In most cases, programming is beneficial for health and survival of the organism. However, the problem of “mismatch” occurs when individuals developmentally adapted to one environment are exposed to another, resulting in detrimental effects on health [20]. The effects of fetal under-nutrition on structure and metabolism, which may lead to later disease is illustrated in figure 1.

Fig 1: The effects of fetal under-nutrition on structure and metabolism, which may lead to later disease. Source: Fall CHD [11]



Today, there is emerging evidence that epigenetic mechanisms are involved in such "programming" of offspring to either maintain health or develop disease in adults [21]. A clear-cut "cause and effect" can be seen in rodent model of intrauterine growth restriction (IUGR). When fetal rats are growth-retarded by diminishing uterine blood flow, they develop diabetes as adults due to reduced β-cell mass [22]. This occurs because the expression of

pancreatic and duodenal homeobox 1 (PDX 1); a transcription factor involved in pancreatic islet development) is compromised as a result of specific alteration in DNA methylation and histone acetylation. Epigenetic factors, by different types of reactions, could mediate the interplay between genes and the environment, resulting in activation or repression of genetic transcription or even silencing the genetic transcription [23]. The most important epigenetic reactions affecting genetic transcription are acetylation and methylation. These reactions occur mainly in the tail of histones which are part of the protein component in chromosomes [24]. Thus, epigenetic processes such as DNA methylation and histone modification allow the developmental environment to modulate gene transcription and many of these changes may then be stable throughout the individual's life time. A report by Einstein et al [25] identified epigenetic alterations that could provide a mechanism linking IUGR with type 2 diabetes later in life. In that animal model study, they identified 56 candidate loci and performed detailed analyses on a subset. They found consistent differences in loci near genes controlling growth such as those involved in cell cycle. Of particular interest was the observed reduction in DNA methylation in the hepatocyte nuclear factor-4-α (HNF4 A) promoter. It is has been established that mutation in HNF4 A is associated with the pathogenesis of maturity onset diabetes of the young, type 1. Several studies using different methods, have identified regions of differential DNA methylation in the placenta of IUGR infants [26-28], supporting this concept in human. Epigenetic modifications are frequently tissue specific, so findings in the placenta may not apply to muscle, adipose tissue, or the pancreatic β-cell. Despite these limitations, this is an area of research that has diagnostic, prognostic and therapeutic implications [29].

A review of the literature suggests that impaired intrauterine growth and development are linked to several diseases in adulthood. Some examples include coronary heart disease, hypertension, and type 2 diabetes [30]. There is also emerging evidence that mitochondrial dysfunction and oxidative stress play important role in the pathogenesis of the Fetal Origin of Adult Disease (FOAD) [30,31]. It is believed that oxidative stress is the primary link between adverse fetal growth and later risks of the metabolic syndrome and type 2 diabetes [30].The fetus is entirely dependent on the nutrients from the mother and adapts to an inadequate nutrient supply in a number of ways: (i) prioritization of brain growth and metabolism at the expense of other tissues such as abdominal viscera; (ii) reduced secretion of and sensitivity to the fetal

growth hormone, insulin and IGF-I; and (iii) up-regulation of the hypothalamic-pituitary-adrenal axis [32]. This preferential distribution of nutrients to the brain impairs the growth of the liver and may underlie permanent abnormalities in the regulation of cholesterol and clotting factors [28]. The FOAD hypothesis proposes that although the events occur in response to a transient phenomenon (fetal under-nutrition), the resultant adaptation become permanent or "programmed" because they occur during a critical period of early development [30]. Tissues and systems for which there is evidence of programming in humans are summarized in Table 1.

Table 1: Tissues and systems for which there is evidence of programming in humans [32]

Tissue or system	Examples of programming
Endocrine system	Hypothalamic-pituitary-adrenal axis Glucose-insulin metabolism Growth hormone-IGF-1 axis
Reproductive system	Age at menarche Polycystic ovary syndrome
Skeletal muscle	Insulin resistance Glycolysis during exercise
Bone	Bone mineral content
Liver	Cholesterol metabolism Fibrinogen and factor VII synthesis
Kidney	Renin-angiothensin system
Cardiovascular system	Vascular compliance Endothelial function
Immune system	Thyroid authoantibodies IgE concentrations
Respiratory system	Lung volume
Central nervous system	Schizophrenia

IGF-I = insulin-like growth factor I; Ig = immunoglobulin

Infants who are born large-for-gestational age (LGA) are at risk of long-term metabolic complications. The effects of fetal "over nutrition" may, therefore, contribute to the rising prevalence rates of type 2 diabetes [12]. Plagemann et al [34-36], in three separate

studies, using animal models suggested that over-nutrition in prenatal period can lead to alteration in DNA methylation patterns within the promoter regions of genes whose products are involved in the hypothalamic regulation of appetite, body weight and metabolism. In the promoter region of proopiomelanocortin (POMC), the most important anorexigenic neurohormone, neonatally overfed rats developed hypermethylation of activating transcription factor binding sites in parallel with hypomethylation at an inhibitory transcription binding site. The promoter region of the hypothalamic insulin receptor gene promoter was found to be hypermethylated. The findings of these studies suggest that perinatal programming of long-term increased obesity and diabetes risk due to neonatal (by extension fetal) over-nutrition may occur via altered methylation patterns of the promoter regions of central nervous body weight-regulating neuropeptides and receptors [34-36]. St Jeor et al [37], reported that a higher birth weight is associated with a higher BMI and an increase in prevalence of obesity in adulthood. Data from the study also suggest rapid weight gain during infancy is associated with obesity later in childhood, perhaps reflecting a combination of genetically determined catch-up growth and postnatal environmental factors [37].

Postnatal growth pattern:

The pattern of growth in the postnatal period is also an important issue. Data from longitudinal studies have shown that after infancy, in individuals born small, crossing BMI centiles upwards during childhood or adolescence is strongly associated with adult disease [37,38]. The phenomenon of adiposity rebound (the point in early childhood when BMI starts to rise, having fallen during infancy) plays an important role in subsequent glucose metabolism. Early adiposity rebound was a strong risk factor for impaired glucose tolerance and development of type 2 diabetes [38,39]. Rapid postnatal catch-up growth contributes actively to insulin resistance, at least during the first years of life [40]. There are a number of possible explanations for the observation that weight gain in childhood in individuals born small might be associated with disease. The process of catch-up may be disadvantageous in itself. Studies, using animal models have shown that compensatory growth can lead to adverse short- and long-term effects, operating through a variety of mechanisms [41]. Such mechanisms include (i) LBW babies undergo catch-up growth in the postnatal period and the rapidity of such growth may simply be a reflection of the severity of the growth retardation; (ii) rapid weight gain may be disadvantageous in itself because of excess demand on tissues that are not capable

compensatory hyperplasia, such as the pancreas; (iii) rapid weight gain may lead to altered body composition; good nutrition in childhood may enhance the development of fat, which maintains the capacity for growth throughout life, but may not restore muscle tissue, which may lose the capacity for cell division early in life; (iv) it is possible that hormones driving catch-up growth have adverse cardiovascular and metabolic effects [42].

Endocrine and Metabolic Consequences:

Several longitudinal studies have demonstrated that being born small-for-gestational age (SGA) is detrimental with adverse metabolic effects, such as insulin resistance and dyslipidaemia as well as hypertension [43]. Together, these contribute to the "multimetabolic" syndrome (Syndrome X); an association that is independent of obesity in later life and a family history of metabolic problems [44]. In the literature, there is increasing evidence that has linked some diseases in adulthood to being born SGA. Such diseases result from fetal programming of certain metabolic and endocrine systems, which may affect health in childhood and adolescence [45]. There is also emerging evidence that epigenetic mechanisms are involved in such "programming" of offspring to either maintain health or develop disease in adulthood [21]. In addition, genetic variations that affect the insulin axis might influence both birth weight and subsequent development of type 2 diabetes and could also explain transgenerational effects [46,47]. The endocrine and metabolic effects of being born SGA and LGA are summarized in Tables 2 and 3 respectively.

Endocrine and Metabolic Effects of Being Born Small-For-Gestational Age:

Alterations in endocrine axes/functions:

Hypothalamic-pituitary-adrenal (HPA) axis:

Animal studies have shown that the early environment can permanently modify the HPA axis [60,61]. In most species, maternal stress during pregnancy leads to hyper-responsiveness of the HPA axis in the offspring, with increased peak and prolonged duration of glucocorticoid secretion [12]. Different types and timings of maternal stress have different long-term effects, and there may be sexual dimorphism; for example, female rat fetuses are more susceptible to HPA effects induced by maternal stress [12]. The placenta forms a partial barrier to glucocorticoids because of the enzyme 11β-hydroxysteroid dehydrogenase (11β-HSD), which converts cortisol and corticosterone to inactive metabolites. It has been observed that levels and activity of 11β-HSD and thus fetal protection from glucocorticoid exposure, are reduced in animal models of intrauterine growth restriction and

maternal under-nutrition [60,61] and in human pregnancies complicated by intrauterine growth restriction [62]. Subtle programmed abnormalities of the HPA axis may play a role in the development of hypertension, insulin resistance, and type-2 diabetes in humans and their link with low birth weight [61]. Growth-restricted fetuses have higher circulating cortisol concentrations [63]. An association between LBW and a syndrome of exaggerated adrenarche, hyperinsulinaemia, precocious puberty, and ovarian hyperandrogenism in girls has been reported [64]. The mechanisms are unknown but the authors postulated that it might indicate either programming by the prenatal environment or genetic disorder of serine kinase activity, leading to abnormal phosphorylation of hormone receptors.

Table 2: Summary of endocrine and metabolic effects of being born small-for-gestational age (SGA)

Organ/System involved	**Biochemical abnormality or clinical disorder**
Growth, puberty and body composition [47-51]	Decrease in growth hormone (GH) action and secretion; Short stature; Accelerated gain in fat mass
Adrenal function [46,52]	Increase in cortisol secretion; Increase in dehydroepiandrosterone sulphate; Exaggerated adrenarche
Gonadal function / Genitalia [47,53]	Polycystic ovary syndrome; Anovulation; Small ovaries and uterus; Male subfertility; Cryptoorchidism; Precocious pubarche
Bone metabolism [54]	Reduction in bone mineral density; increase in risk of osteoporosis and fracture in adulthood
Changes in other hormones [31,55]	Decrease in adiponectin and follistatin in children. Increase in Prostaglandin factor-2α
Metabolic; Resetting of insulin-like growth factor and insulin systems [49-51]	Insulin resistance; Hyperinsulinism; Decrease in insulin-like growth factor-binding protein-1; Hypercholesterolaemia; Increase in prevalence of metabolic syndrome; Ketotic hypoglycaemia (accelerated starvation) in childhood
Increased risk of adult disease [30,56]	Stroke; Heart failure; Type 2 diabetes; Obesity; Hypertension

Table 3: Summary of endocrine and metabolic effects of being born large-for-gestational age (LGA)

Organ/System involved	Biochemical abnormality or clinical disorder
Growth, puberty and body composition [57]	At the age of 9-10 years and 23-25 years, individuals born LGA remain heavier and taller than individuals born AGA. Young adults born LGA present higher BMI, waist circumference and blood pressure than controls
Metabolic [31,37,58]	Insulin resistance Increase in risk of obesity Increase in risk of type 2 diabetes Increase in prevalence of metabolic syndrome Increase in risk of oxidative stress Increase in risk of hypertension
Changes in other hormones [31,59]	Increase in levels of plasma IGF-1 Increase in levels of Prostaglandin factor-2α

AGA = appropriate for gestational age

Growth hormone (GH) and insulin-like growth factors (IGF):

Cord blood IGF-I concentrations are low in growth-restricted fetuses and newborns, suggesting a possible growth hormone resistance [12]. On the other hand, insulin-like growth factor binding protein-1 (IGFBP-I) concentrations are increased in such fetuses and newborns. GH deficiency is associated with osteoporosis [12]. Data from follow-up studies of children with IUGR and subsequent short stature indicate that they have persistent abnormalities of the GH-IGF axis with low-amplitude GH peaks and high baseline GH secretion [50,65]. In addition, they have low serum IGF-I, IGF-II , and IGFBP-3 concentrations compared with non-IUGR children of normal stature [50,66] but higher concentrations than non-IUGR short children, suggesting that they may be resistant for both IGF and GH. Infants affected by IUGR have low concentrations of insulin and IGF-I at birth and normalization of these parameters occur in the postnatal period [67]. It is thought that tissues chronically depleted of insulin and IGF-I throughout fetal life and then suddenly exposed to increased concentrations of the two hormones shortly after birth may counteract the actions of insulin by developing insulin resistance. Thus, in this proposed scenario, insulin resistance is serving as a metabolic

defense mechanism to protect the organism against hypoglycaemia [68].

Puberty and reproductive function:

Report of some studies indicate that girls born SGA have earlier menarche [69,70], particularly if they experienced accelerated growth in infancy and accelerated BMI gain from birth to 7 years of age [71]. Data from Nordic countries indicate that the risk of cryptoorchidism and hypospadias was higher in babies born SGA than babies born AGA [72,73].

Body composition and obesity:

In a study involving Danish conscripts, the prevalence of obesity, defined as a BMI of 30kg/m^2 or more, rose from 3.5% in those with birth weight of 2.5 kg or less to 11.4 % in those with birth weight greater than 4.5kg [74]. However, there is good evidence that the positive correlation between birth weight and adult BMI reflects increased lean and muscle mass rather than adiposity [13]. In a small study involving 22 young Korean men, no association was observed between birth weight and visceral fat area measured by computed tomography [75].

Bone metabolism and osteoporosis:

Fetal growth has been linked to osteoporosis later in life [76]. Being born SGA has been associated with reduced bone mineral density and an increased risk of osteoporosis and fracture in adulthood, particularly in those born short [54]. It has been postulated that fetal programming of bone mass may be mediated through the effects of environmental stressors during intrauterine or early postnatal life on the sensitivity of the growth plate to growth hormone and cortisol [45]. The consequences of such fetal programming might be to reduce peak skeletal size and perhaps, mineralization, thus predisposing to an accelerated rate of bone loss during later life.

Metabolic abnormalities:

Obesity, type-2 diabetes and the metabolic syndrome:

Several epidemiological studies have demonstrated the long-term impact of being born SGA on metabolic health in adulthood. For instance, increased risk of type 2 diabetes and metabolic syndrome have been demonstrated in such individuals, both during childhood and adulthood [77-79]. Leunissen et al [80] demonstrated that rapid weight gain during the first 3 months of life in individuals born SGA was associated with reduced insulin sensitivity, lower HDL-cholesterol, and higher serum triglycerides.

Endocrine and Metabolic Effects of being Born Large-For-Gestational Age:

Endocrine function:

Growth, puberty, body composition and polycystic ovarian syndrome:

At the age of 9-10 years and 23-25 years, individuals born LGA were found to remain heavier and taller than individuals born AGA [56]. In the same study, the authors also found that young adults born LGA present higher BMI, waist circumference and blood pressure than controls [57]. Some studies have shown that 30% of children born LGA had early adiposity rebound and is associated with a larger body size in childhood [81]. There is evidence that prenatal factors play a role in the aetiology of polycystic ovarian syndrome (PCOS). Cresswell et al [82] observed that some women with high birthweight and whose mothers were above average weight in pregnancy were at increased risk of developing PCOS. The authors linked it to an ovarian defect, either of genetic origin or resulting from some effect of maternal obesity, leading to hypersecretion of androgens.

Hormone-related cancer:

Higher birth weight is associated with an increased risk of breast cancer (relative risk 1.5 - 1.7 for birth weights > 4000g compared with normal birth weights 2500-2999 g) [83].

Metabolic abnormalities:

Infants who are born LGA are at risk of potentially long-term metabolic complications.

Glucose homeostasis and lipid metabolism:

Exposure to fetal over-nutrition, resulting in LGA at birth is associated with increased risk of type 2 diabetes and obesity (additional to any inherited predisposition) [82]. This is often referred to as the "fuel-mediated teratogenesis hypothesis". Children who are LGA at birth are at increased risk of insulin resistance and oxidative stress [31,85]. There is a strong positive correlation between oxidative stress and obesity in childhood [86]. In the beginning of adult life, subjects born LGA, especially those who did not experience a catch-down of weight during childhood appear to be at increased risk of higher BMI, central adiposity and higher blood pressure [57]. Boney et al [58] have demonstrated that children who were LGA at birth and were exposed to an intrauterine environment of either diabetes or maternal obesity were at increased risk of metabolic syndrome during childhood.

The Relevance of FOAD in Nigeria and Other Developing Countries:

Worldwide, there is a linear and graded trend in cardiovascular mortality in relation to birth weight [87]. This observation suggests that majority of the world's population experience sub-optimal fetal growth, particularly in developing countries; also referred to as "resource-limited countries". In Nigeria and other developing countries, the prevalence of low birth weight range from 17% to

38% [88]. In addition, some developing countries are witnessing a rapid increase in incidence of obesity among children and adults because of economic development, nutrition transition and changing lifestyle [89,90]. Thus, the combination of high prevalence of fetal growth retardation and rising incidence of obesity, create a potential greater risk for adult cardiovascular disease and type 2 diabetes mellitus, ultimately reducing life expectancy. Studies have shown that within the same country, obesity-related health burden is disproportionately experienced by children from low income and ethnic minority families and this serves to perpetuate health disparities between rich and poor families [91,92].

The Public Health Implications and Future Perspective:

The knowledge that fetal growth retardation is linked to endocrine and metabolic disease later in life, suggests that such diseases could be prevented by improving fetal growth and development via improved maternal health and nutrition. Data from the study by Boney et al [57] provided strong evidence that a mother's nutrition programs the metabolism and growth of her offspring. Perinatal epigenetic analysis may have utility in identifying individual vulnerability to later obesity and metabolic disease. The epigenetic changes induced by maternal/fetal environment are not necessarily immutable, but they can be reversed during critical developmental windows. For example, the programming of diabetes in IUGR rats can be avoided by injection of a glucagon-like peptide analogue at birth with restoration of β-cell mass [23]. Thus, with a clear understanding of how maternal conditions affect the pathway that lead to diabetes, obesity and the metabolic syndrome, we may one day be able to "vaccinate" children to reverse potentially detrimental epigenetic alterations and thereby prevent the manifestation of adult disease [28].

Future research should focus on exploring the relationship between prenatal, natal and postnatal growth pattern on one hand and nuero-endocrine and metabolic effects in later life on the other hand. In addition, studies focusing on the mechanisms by which metabolism, body composition and growth may be permanently affected in individuals born either SGA or LGA is necessary. There are ongoing research projects in various institutions in India addressing these subjects [31].

CONCLUSIONS:

Individuals born SGA are at increased risk of development of endocrine dysfunction and metabolic abnormalities in later life. This detrimental effect manifests as insulin resistance, gonadal and somatotropic axes abnormalities and premature adrenarche. SGA birth has been linked to the escalating prevalence of type 2

diabetes in the paediatric age group. Individuals born LGA are at increased risk of metabolic syndrome and obesity later in life. The rising incidence of type 2 diabetes in childhood and adolescence has been attributed to the rising incidence of obesity in various populations. Thus, the persistent rise in incidence obesity will result in a perpetual increase in incidence of type 2 diabetes with the attendant adverse health consequences in subsequent generations. Thus, abnormal size at birth represents a significant public health concern. This public health importance of being born either SGA or LGA, underlie the relevance of careful follow-up of children born either SGA or LGA to detect the development of metabolic abnormalities during childhood. Health intervention strategies directed at mothers and aimed at reducing the frequency of delivery of babies with abnormal size at birth will be beneficial, not only to the individual's health but also the health of future generations.

REFERENCES:

1. Johnston L. The genetics of birth size: small for gestational age. MIMS Advances: Short children born small for gestational age. London, Haymarket Medical Publications Ltd, 2004:10-12.
2. Usha K, Sarita B. Placental insufficiency and fetal growth restriction. J Obstet Gynecol India 2011;61(5):505-511.
3. Barker DJP. Mothers, babies and health in later life. Edinburgh, Brace and Co 1998.
4. Dunger DB, Petry CJ, Ong KK. Genetics of size at birth. Diabetes Care 2007;30(Suppl 2):S150-S155.
5. Magnus P. Causes of variation in birth weight: a study of offspring of twins. Clin Genet 1984;25:15-24.
6. Barker J, Lin JP, Robertson EJ, Efstratiads A. Role of insulin-like growth factors in embryonic and postnatal growth. Cell 1993;75:73-82.
7. Woods KA, Camacho-Hubner C, Savage MO, Clark AJ. Intrauterine growth retardation and postnatal growth failure associated with deletion of insulin-like growth factor I gene. N Engl J Med 1996;335:1363-1367.
8. Abuzzahab MJ, Schneider A, Goddard A, Grigoreseu F, Lautier C, Keller E, Kiess W, Klammt J, Kratzsch J, Osgood D, Pfaffle R, Raile K, Seidl B, Smith RJ, Chernausck SD. IGF-I receptor mutation resulting in intrauterine and postnatal growth retardation. N Engl J Med 2003;349:2211-2222.
9. Lucas A. Programming by early nutrition in man. Ciba Foundation Symposium 1991;156:38-50.
10. Waterland RA, Garza C. Potential mechanisms of metabolic imprinting that lead to chronic disease. Am J Clin Nutr 1999;69:179-197.
11. Nieto-Diaz A, Villar J, Materras-Weinig R, Valenzuela-Ruiz P. Intrauterine growth retardation at term: association between anthropometric and endocrine parameters. Acta O & G Scand 1996;75:127-131.
12. Fall CHD. Endocrine programming and the fetal and early-life origins of adult disease. In: Brooks CGD, Clayton PE, Brown RS eds. Clinical Pediatric Endocrinology. 5th edition, Oxford, Blackwell Pub Ltd 2005:396-409.
13. Hales CN, Baker DJP. The thrifty phenotype hypothesis. Br Med Bull 2001;60:5-20.
14. Neel JV. The "thrifty genotype" in 1998. Nutr Rev 1998;57(Part II):S2-S9.

15. Mericq MV. Low birth weight and endocrine dysfunction in postnatal life. In: Lifshitz F ed. Pediatric Endocrinology, 5th edition, Vol 2, New York, Informa Healthcare USA, Inc, 2007:251-260.
16. Robinson R. The fetal origin of adult disease: No longer just a hypothesis and may be critically important in South Asia. BMJ 2001;322(7283):375-376.
17. Aiken CE, Ozanne SE. Transgenerational developmental programming. Hum Reprod Update 2013;20:63.
18. Ravelli ACJ, van der Meulen JHP, Michels RPJ, Osmond C, Barker DJ, Hales CN, Blecker OP. Glucose intolerance in adults after prenatal exposure to famine. Lancet 1998;351:173-177.
19. Sebert S, Sharkey D, Budge H, Symonds ME. The early programming of metabolic health: Is epigenetic setting the missing link? Am J Clin Nutr 2011;94:1953S-1958S.
20. Bateson P, Barker D, Clutton-Brock T , Deb D, D'Udine B, Foley RA, Gluckman P, Godfrey K, Kirwood T, Lahr MM, McNamara J, Melcalfe NB, Monaghan P, Spencer AG, Sultan SE. Developmental plasticity and human health. Nature 2004;430(6998):419.
21. Park JH, Stoffer DA, Nicholls RD, Simmonds RA. Development of type 2 diabetes following intrauterine growth retardation in rats is associated with progressive epigenetic silencing of Pdx 1. J Clin Invest 2008;118:2316-2324.
22. Pinney SE, Jaeckle Santos LJ, Han Y, Stoffers DA, Simmons RA. Extendin-4 increase histone acetylase activity and reverse epigenetic modifications that silence Pdx 1 in intrauterine growth retarded rat. Diabetologia 2011;54:2606-2614.
23. Negrato CA, Gomes MB. Low birth weight: causes and consequences. Diabetol Metab Syndrome 2013;5:49-56.
24. Einstein F, Thompson RF, Bhagat TD, Fazzari MJ, Verma A, Barzilai N, Greally JM. Cytosine methylation dysregulation in neonates following intrauterine growth restriction. PLoS One 5:e8887.
25. Lambertini L, Lee TL, Chan WY, Lee MJ, Diplas A, Wetmur J, Chen J. Differential methylation of imprinted genes in growth-retarded placentas. Reprod Sci 2011;18:1111-1117.
26. Yuen RK, Penaherreva MS, von Dadelszen P, McFadden DE, Robinson WP. DNA methylation profiling of human placentas reveal promoter hypomethylation of multiple genes in early-onset pre-eclampsia. Eur J Hum Genet 2010;18:1006-1012.
27. Banister CE, Koestler DC, Maccani MA, Padbury JF, Houseman EA, Masil CJ. Infant growth restriction is associated with distinct pattern of DNA methylation in human placenta. Epigenetics 2011;6:920-927.
28. Chenausek SD. Update: Consequences of abnormal fetal growth. J Clin Endocrinol Metab 2012;97(3):689-695.
29. Gera T, Choudhury P, Dubey AP. Fetal origin of adult diseases. In: Dutta AK, Sachdva A eds. Recent Advances in Pediatrics. New Delhi, Jaypee Brothers Medical Publishers Ltd, 2007:757-761.
30. Chiavaroli V, Giannini C, D'Adamo E, de Giorgis T, Chiarelli F, Mohn A. Insulin resistance and oxidative stress in children born small and large for gestational age. Pediatrics 2009;124(2):695-702.
31. Khanna SB, Swasti KD, Dwivedee K. Fetal origin of adult disease. JK Science 2007;9(4):206-210.
32. Godfrey KM, Barker DJP. Fetal nutrition and adult diseases. Am J Clin Nutr 2000;71(suppl):1344S-1352S.
33. Plagemann A, Harder T, Brunn M, Harder A, Roekpe K, Wittrock-Staar M, Ziska T, Schellong K, Rodekamp E, Melchior K, Dudenhausen JW. Hypothalamic proopiomelanocortin promoter methylation becomes altered by early over-feeding: an

epigenetic model of obesity and metabolic syndrome. J Physiol 2009;587:4963-4976.

34. Plagemann A, Roekpe K, Harder T, Brunn M, Harder A, Wittrock-Staar M, Ziska T, Schellong K, Rodekamp E, Melchior K, Dudenhausen JW. Epigenetic malprogramming of the insulin receptor promoter due to developmental overfeeding. J Perinat Med 2010;38:393-400.
35. Plagemann A, Harder T, Schellong K, Schulz S, Stupin JH. Early postnatal life as a critical time window for determination of long-term metabolic health. Best Pract Res Clin Endocrinol Metab 2012;26(5):641-653.
36. St Jeor ST, Hayman LL, Daniels SR, Gillman MW, Howard G, Law CM, Lewis CE, Poehlman E, American Heart Association. Prevention Conference VII: Obesity, a worldwide epidemic related to heart disease and stroke: group II: Age-dependent risk factors for obesity and co-morbidities. Circulation 2004; 110: 471- 475.
37. Erickson JG, Forsen T, Tuomilehto J, Osmond C, Barker DJP. Early adiposity rebound in childhood and risk of type 2 diabetes in adult life. Diabetologia 2003; 46: 190 - 194.
38. Bhargava SK, Sachdev HPS, Fall CHD, Osmond C, Lakshmy R, Barker DJP, Dey Biswas SK, Ramji S, Prabhakaran D, Reddy KS. Relation of serial changes in childhood body mass index to impaired glucose tolerance in young adulthood. N Engl J Med 2004;350:865-875.
39. Cettour-Rose P, Samec S, Russell AP, Summermatter S, Mainieri D, Carrillo-Theander C, Montani JP, Seydoux J, Rohner-Jeanrenaud F, Dulloo AG. Redistribution of glucose from skeletal muscle to adipose tissue during catch-up fat: a link between catch-up growth and later metabolic syndrome. Diabetes 2005;54(3):751-756.
40. Metcalfe NB, Monaghan P. Compensation for a bad start: grow now, pay later? Trends Ecol Evol 2001;16:254-260.
41. Lever AF, Harrap SB. Essential hypertension: a disorder of growth with origins in childhood? J Hypertens 1992;10:101-120.
42. Jaquet D, Gaboriau A, Czennichow P, Levy-Marchal C. Insulin resistance early in adulthood in subjects born with intrauterine growth retardation. J Clin Endocrinol Metab 2000;85:1041-1046.
43. Morgan AR, Thompson JM, Murphy R, Black PN, Lam WJ, Ferguson LR, Mitchell EA. Obesity and diabetes genes are associated with being born small for gestational age: results from the Auckland Birthweight Collaborative Study. BMC Med Genet 2010;11:125.
44. Zhao J, Li M, Bradfield JP, Wang K, Zhang H, Sleiman P, Kim CE, Annaiah K, Glaberson W, Glessner JT, Otieno FG, Thomas KA, Garris M, Hou C, Frackelton EC, Chiavacci RM, Berkowtiz RI, Hakornarson H, Grant SF. Examination of type 2 diabetes loci implicates CDKAL 1 as a birthweight gene. Diabetes 2009; 58: 2414-2418.
45. Gregory JW. Metabolic consequences of being born small for gestational age (SGA). MIMS Advances: Short children born small for gestational age. London, Haymarket Medical Publications Ltd, 2004:13-15.
46. de Zegher F, Lopez-Bermojo A, Ibenez L. Adipose tissue expandability and the early origins of PCOS. Trends Endocrinol Metab 2009;20:418-423.
47. Saenger P, Czernichow P, Hughes I, Reiter EO. Small for gestational age: short stature and beyond. Endocr Rev 2007;28:219-251.
48. Hernandez MI, Merciq V. Impact of being born small for gestational age on onset and progression of puberty. Best Pract Res Clin Endocrinol Metab 2008;22:463-476.

49. Boguszewski M, Bjarnason R, Jansson C, Rosberg S, Albertson-Wikland K. Hormonal status of children born small for gestational age. Acta Paed 1997;(Suppl 423):189-192.
50. Cutfield WS, Hofman PL, Wickers M, Breier B, Blum WF, Robinson EM. IGFs and binding proteins in short children with intrauterine growth retardation. J Clin Endocrinol Metab. 2002;87:235-239.
51. Iniguez G, Ong K, Bazaes R, Avila A,Salazar T, Dunger D, Merciq V. Longitudinal changes in insulin-like growth factor-1, insulin sensitivity, and secretion from birth to age three years in small for gestation age children. J Clin Endocrinol Metab 2006;911:4645-4649.
52. Walker BR, Irving RJ, Andrew R, Belton NR. Contrasting effects of intrauterine growth retardation and premature delivery on adult cortisol secretion and metabolism in man. Clin Endocrinol 2002;57:351-355.
53. Ibanez L, Potau N, de Zegher F. Precocious pubarche, dyslipidaemia, and low IGF binding protein-1 in girls: relation to reduced prenatal growth. Ped Res 1999;46:320-322.
54. Cooper C, Eriksson JG, Forsen T, Osmond J, Tuomilehto J, Barker DJ. Maternal height, childhood growth and risk of fracture in later life: a longitudinal study. Osteoporosis Int 2001;12:623-629.
55. Ibanez L, Lopez-Bermjo A, Diaz M, Jaramillo A, Marin S, de Zegher F. Growth hormone therapy in short children born small for gestational age: effects on abdominal fat partitioning and circulating follastatin and high molecular weight adeponectin. J Clin Endocrinol Metab 2010;95:2234-2239.
56. Barker DJ. Adult consequences of fetal growth restriction. Clin Obstet Gynecol 2006;49:270-273.
57. Espineira AR, Fernandes-Rosa FL, Bueno AC, de Souza RM, Moreira AC, de Castro M, Barbieeri MA, Bettiol H, Antonini SR. Postnatal growth and cardiometabolic profile in young adult born large for gestational age. Clin Endocrinol 2011;75(3):335-341.
58. Boney CM, Verma A, Tucker R, Vohr BR. Metabolic syndrome in childhood: Association with birth weight, maternal obesity, and gestational diabetes. Pediatrics 2005;115(3):e290-e296.
59. te Velde SJ, van Rossum EF, Voorhoeve PG, Twisk JWR, van de Waal HAD, Stehouwer CDA, van Mechelen W, Lamberts SWJ, Kemper HCG. An IGF-I promoter polymorphism modifies the relationship between birth weight and risk factors for cardiovascular disease and diabetes at age 36. BMC Endocr Disorders 2005;5:5-14.
60. Matthew SG. Early programming of the hypothalamo-pituitary-adrenal axis. Trends Endocrinol Metab 2002;13:373-380.
61. Phillips DIW. Programming of adrenocortical function and the fetal origin of adult disease. J Endocrinol Invest 2001;24:742-746.
62. Secki JR, Cleasby M, Nyirenda MJ. Glucocorticoids, 11β hydroxysteroid dehydrogenase, and fetal programming. Kidney Int 2000;57:1412-1417.
63. Economides DL, Nicholaides KH, Linton EA, Perry LA, Chard T. Plasma cortisol and adrenocorticotrophin in appropriate and small for gestational age fetuses. Fetal Ther 1988;3:158-164.
64. Ibanez I, Dimartino-Nardi J, Potua N, Saenger P. Premature adrenarche – normal variant or forerunner of adult disease? Endocr Rev 2000;21:671-696.
65. Albertsson-Wikland K, Boguszewski M, Karlberg J. Children born small-for-gestational age: postnatal growth and hormonal status. Horm Res 1998;49(Suppl 2):7-13.
66. Clanfarani S, Geremia C, Sctt CD, Germani D. Growth, IGF-I system, and cortisol in children with intrauterine growth retardation: is catch-up growth affected by

reprogramming of the hypothalamic-pituitary-adrenal axis? Pediatr Res 2002;51:94-99.
67. Leger J, Noel M, Limal M, Czernichow P. Growth factors and intrauterine growth retardation II. Serum growth hormone, insulin-like growth factors (IGF-I) and IGF-binding protein 3 levels in children with intrauterine growth retardation compared with normal control subjects: prospective study from birth to two years of age. Pediatr Res 1996;40(1):101-107.
68. Clanfarani S, Germani D, Branca F. Low birthweight and adult insulin resistance: the "catch-up growth hypothesis." Arch Dis Child 1999;81(1):F71-F73.
69. Voordouw JJ, van Weissenbruch MM, Delamarre-van de Waal HA. Intrauterine growth retardation and puberty in girls. Twin Res 2001;4:299-306.
70. Luo ZC, Cheung YB, He Q, Albertsson-Wikland K, Karlberg J. Growth in early life and its relation to pubertal growth. Epidemiology 2003;14(1):65-73.
71. dos Santos Silva I, De Stavola BL, Mann V, Kuh D, Hardy R, Wasworth MEJ. Prenatal factors, childhood growth trajectories and age at menarche. Int J Epidemiol 2002;31:405-412.
72. Boisen KA, Main KM, Rajpert-de Meyts E, Skakkebaek NE. Are male reproductive disorders a common entity? The testicular dysgenesis syndrome. Ann NY Acad Sci 2001;948:90-99.
73. Boisen KA, Chellakooty M, Schmidt IM, Kai CM, Damgaard IN, Soumi AM, Toppari J, Skakkebaek NE, Main KM. Hypospadias in a cohort of 1072 Danish newborn boys: prevalence and relationship to placental weight, anthropometrical measurements at birth, and reproductive hormone levels at three months of age. J Clin Endocrinol Metab 2005;90(7):4041-4046.
74. Sorensen HT, Sabroe S, Rothman KJ, Gillman M, Fischer P, Sorensen TIA. Relation between weight and length at birth and body mass index in young adulthood: cohort study. BMJ 1997;315:1137.
75. Choi CS, Kim C, Lee WJ, Park JY, Hong SK, Lee MG, Park SW, Lee KU. Association between birth weight and insulin sensitivity in healthy young men in Korean: role of visceral adiposity. Diabetes Res Clin Pract 2000;49:53-59.
76. Javaid MK, Cooper C. Prenatal and childhood influences on osteoporosis. Best Pract Res Clin Endocrinol Metab 2002;16:349-367.
77. Reinehr T, Kleber M, Toschke AM. Small for gestational age status is associated with metabolic syndrome in overweight children. Eur J Endocrinol 2009;160:579-584.
78. Brufani C, Grossi A, Fintini D, Tozzi A, Nocerino V, Patera PI, Ubertini G, Porzio O, Barbetti F, Cappa M. Obese children with low birth weight demonstrate impaired β cell function during oral glucose tolerance test. J Clin Endocrinol Metab 2009;94:4448-4452.
79. Meas T, Deghmoun S, Alberti C, Carreira E, Armoogum P, Chvenne D, Levy-Marchas C. Independent effects of weight gain and fetal programming on metabolic complications in adults born small for gestational age. Diabetologia 2010;53:907-913.
80. Leunissen RW, Kerkhof GF, Stijnen T, Hokken-Koelega A. Timing and tempo of first-year rapid growth in relation to cardiovascular and metabolic risk profile in early adulthood. JAMA 2009;301:2234-2242.
81. Whitaker RC, Pepe MS, Wright JA, Seidel KD, Dietz WH. Early adiposity rebound and the risk of adult obesity. Pediatric 1998;101(1011): Available at www.peditrics.org/cgi/content/full/101/3/e5.
82. Cresswell JL, Barker DJP, Osmond C, Egger P, Phillips DIW, Fraser RB. Fetal growth, length of gestation, and polycystic ovaries in adult life. Lancet 1997;350:1131-1135.

83. Potischman N, Troisi R. In-utero and early – life exposures to risk of breast cancer. Cancer Causes Control 1999;10:561-573.
84. Plagemann A, Harder T. Fuel-mediated teratogenesis and breast feeding. Diabetes Care 2011;34:779-780.
85. Evagelidou EN, Kiortsis DN, Bairaktari ET, Giapros VI, Cholevas VK, Tzallas CS, Andronikou SK. Lipid profile, glucose homeostasis, blood pressure and obesity-anthropometric markers in macrosomic offspring of non-diabetic mothers. Diabetes Care 2006;29(6):1197-1201.
86. Ustundag B, Gungor S, Aygun AD, Turgut M, Yilmaz E. Oxidative status and serum leptin levels in obese prepubertal children. Cell Biochem Funct 2007; 25(5):479-483.
87. Barker DJ, Osmond C, Golding J, Kuh D, Wadsworth ME. Growth in utero, blood pressure in childhood and adult life and mortality from cardiovascular disease. Br Med J 1989;298:564-567.
88. Ghosh S. Low birth weight babies. In: Udani PM ed. Textbook of Pediatrics With Special Reference to Problems of Child Health in Developing Countries. New Delhi, Jaypee Brothers Medical Pub Ltd, 1998:324-348.
89. Monteiro CA, Benicio MHD'A, Conde WL, Popkin BM. Shifting obesity trends in Brazil. Eur J Clin Nutr 2000; 54:342-346.
90. Popkin BM. The shift in stages of the nutrition transition in the developing world differs from past experiences. Public Health Nutr 2002; 5:205-214.
91. Anderson S, Whitaker R. Prevalence of obesity among US preschool children in different racial and ethnic groups. Arch Pediatr Adolesc Med 2009; 163(4):344-348.
92. Wang Y, Beydoun M. The obesity epidemic in the United States – gender, age, socioeconomic, racial/ethnic, and geographic characteristics: a systematic review and meta-regression analysis. Epidemiological Review 2007;29:6-28.

"SILENT" CARRIERS OF HIV AND THE EPIDEMIOLOGY OF AIDS: A REVIEW

CLEMENT E. ANYIWO

MD, M. Sc; Zeugnis Immun; FMCPath, FWACP, FICS

American International Institute of Graduate Studies, San Antonio, Texas USA

caemenike@gmail.com

ABSTRACT:

Notwithstanding that 30 years have elapsed since the discovery of the causative agent there is still no cure or vaccine for AIDS despite relentless efforts by researchers. This is complicated by the emergence of "silent" HIV carriers which are fueling the pandemic. "Silent" carriers are defined as seronegative, yet infected individuals who have never seroconverted. This should not be confused with late stage of AIDS, when infected individuals are severely immunocompromised, and the so-called "long window" period when antibodies are not detectable. Previous attempts to detect "silent" carriers have used a nucleic acid-dependent viral load test, Polymerase Chain Reaction (PCR) and Antigen Capture Assay (p24). The PCR that detects viral load as early as 2-4 weeks is costly and complicated; it is prone to false positives because of high sensitivity. The Antigen Capture Assay also has limitations of low sensitivity and false negative results. It is for these reasons that a new test called "stimmunology technique" -a SMARTube pre-analytical test is being advocated. This novel in-vitro enhancement technique enables accelerated pre-seroconversion confirmed diagnosis within a few days. The test is cheap, simple to perform and does not change the current algorithm for HIV antibody testing and diagnosis. This technique corroborates what WHO says; that the opportunity to control the epidemic lies in detecting the infection in its early stages and focusing prevention and treatment on them.

***KEYWORDS**: Impact, HIV, Silent carriers, AIDS Epidemiology*

Submitted: February 2014; Accepted: March 2014

INTRODUCTION:

During my postdoctoral fellowship at the university college hospital in London, a fellow researcher on HIV/AIDS jokingly said it is about time we changed the meaning of the acronym "AIDS" to be "Acquired Inhuman Deficiency Syndrome". Everybody laughed. But it is no longer a laughing matter when you consider that AIDS has defied consistent efforts of researchers to produce either a cure or vaccine and the disease has robbed the world of some 25 million lives since its advent over 30 years ago and still decimating the sanctity of mankind! Such a disease is truly inhuman.

However, in his article titled "Berlin patient spreads hope for AIDS cure" in Washington Times in July 2012, Cory Brown [1] reported about the man believed to be the only patient completely cured of AIDS virus. He is Timothy Brown, known in medical circles as "The Berlin Patient". His physician, a German haematologist, Gero Hutter treated him with a revolutionary therapy - a stem cell bone marrow transplant from a donor who had natural immunity to HIV and leukaemia for which he was also diagnosed. Allers and colleagues had earlier reported this evidence of the cure of HIV infection [2].

There are several conflicting history and theories about the origin of AIDS. It would appear that scientists now believe that HIV probably transferred to humans in Africa between 1884 and 1924 [3]. Studies have shown that HIV first arose in Africa and spread from primates to humans early in the 20th century, most probably when humans came into contact with infected blood during a chimpanzee hunt [4] (The Hunter's theory). By testing stored blood samples, scientists have found direct evidence of a human being infected as far back as 1959 with a virus now classified as HIV [5]. In the 1970s African doctors saw a rise in opportunistic infections and wasting, while western scientists and practicing physicians were not yet aware of the growing epidemic. The origin of AIDS and HIV has puzzled scientists ever since the illness first came to light in the 1980s. At this juncture one can only rehearse what an erstwhile president of Zambia - Dr. Kenneth Kaunda said, "……..that we should get more concerned with where AIDS is going rather than where it is coming from".

On June 5, 1981 the United States Center for Disease Control and prevention (CDC) published an article in their Morbidity and Mortality Weekly Report describing cases of rare lung infection *Pneumocystis carinii* pneumonia in 5 young previously healthy gay men in Los Angeles [6]. It was then termed "gay plague". By the end of that

year there were a cumulative total of 270 reported cases from physicians across the United States of severe immune deficiency among homosexuals and 121 of those individuals have already died. We are now aware that the disease has spread to every corner of the globe and is the leading cause of infectious death worldwide. According to the WHO over 35 million people have been infected worldwide, 25 million dead since the pandemic, 2 million dying every year, 250,000 being children and that over 50 percent are women [7]. There are over 6000 infections daily and most of them are occurring in Sub-Saharan Africa [7]. Thus, Africa has been designated as the center point of the AIDS pandemic recording some 25 million of AIDS population. The AIDS epidemic not only affects the health of individuals, it impacts households, communities and the development and economic growth of nations [7]. The immune-pathogenesis of HIV infection is presented in the schematic diagram in Fig. 1.

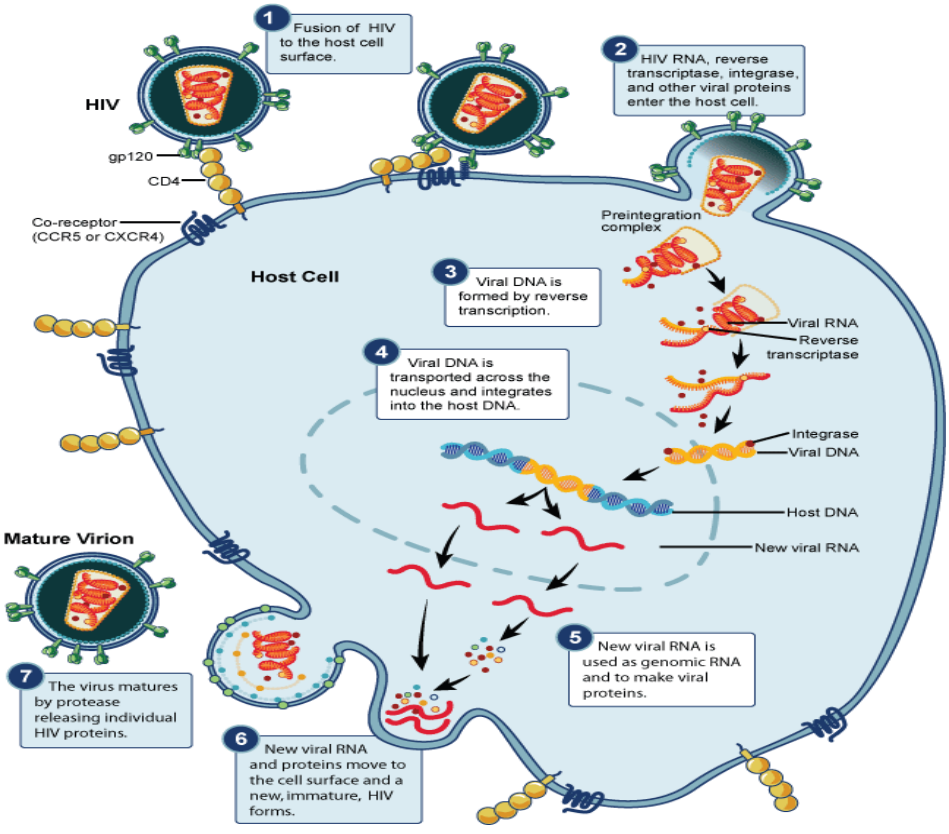


Fig. 1: Schematic diagram of the immune-pathogenesis of HIV infection (Adapted from National Institute of Allergy and Infectious Disease (NIAID) [8]

Transmission is through exposure to sexual bodily fluids, cerebrospinal fluid, sharing needles, tattooing or piercing with contaminated equipment from an infected individual, from mother to her unborn child during pregnancy or delivery [9]. Transmission through blood transfusion is rare as well as through anti-haemophilic factor VIII since blood is screened and factor VIII heated especially in developed countries [9].

The HIV targets CD4 immune cells and the nervous system. During the course of infection, patients experience gradual decline in CD4 cells which immunocompromises them to the development of life threatening opportunistic infections, (viral, fungal and protozoal), Tuberculosis, extreme weight loss, cancers (such as Kaposi sarcoma) and dementia [10]. However, patients consistently taking anti-retroviral medication are likely to control the infection and not progress to full blown AIDS [11].

The predominant virus is HIV-1 and the relatively uncommon type is HIV-2 which is concentrated in West Africa and rarely found elsewhere. The strains of HIV-1 remained groups M, N, and O until Plantier et al. identified a new strain in a Cameroonian woman in 2009 [12], which they designated group P. Within group M of HIV-1 there is a total of 9 genetic subtypes or clades identified and are designated A, B, C, D, F, G, H, J, K, within which are Circulating Recombinant Forms (CRFs) which developed as a result of viral sexual reproduction. Examples of such are CRF A/B and CRF A/E because it could have resulted from a hybridization between subtype A and some other “parent”, subtype E, although no one has yet isolated subtype E in pure form [13]. In addition, 10 genetic subtypes of HIV-2 have been detected worldwide [14].

The relevance of HIV subtypes is reflected on seroepidemiology, transmissibility, therapy and vaccine production [15]. Differences in transmission and disease progression occur between these subtypes and CRFs. For example, subtype D is shown to be more virulent and accelerates development of AIDS more than subtype A, because it is more effective in binding to immune cells. Subtype B is found to be spread mostly by homosexual contact and intravenous drug use while subtype C and CRF A/E fuel heterosexual epidemic [12] Are more subtypes likely to occur? It is almost certain that new HIV subtypes as well as CRFs will be described in the future as virus recombination continues to occur [16]. The current subtypes and CRFs may also continue to spread to new geographical areas as the global epidemic continues [13].

The search for a cure for HIV began as soon as the virus was identified in 1983 [17]. HIV is probably the most studied virus in the history of

scientific research. Scientists have detailed knowledge of the virus replicative cycle, genes and proteins and also understand their functions. One of the problems with finding a cure is that the virus can hide in cells throughout the body and also hide in areas that are difficult for drug to reach such as the brain. These infected cells that persist in the body are being studied to determine how they can be stimulated to produce virus and /or be targeted for clearance from the body with new drugs that are being developed [16]. Fortunately nearly all subtypes identified so far are responsive to anti-retroviral drug. However, some studies have found variation in the ways different subtypes and CRFs respond to anti-retrovirals [12]. Unfortunately HIV cannot be cured, with the exception of the case of the Berlin Patient mentioned earlier. Nevertheless the Highly Active Antiretroviral Therapy (HAART), which is a poly-therapy consisting of a combination of several antiretroviral drugs, does delay or potentially completely stops disease progression, thus allowing an infected individual to live longer healthy life. This is because antiretroviral drugs can lower the viral load and raise the CD4 count [10]. However, it is a myth that people on anti-retroviral medication cannot transmit HIV to their sexual partners. Since there is no effective vaccine for HIV currently, post exposure prophylaxis (PrEP) is being advocated for HIV prevention in high risk individuals, especially those with multiple sexual partners and those at occupational risk. Currently gene therapy is been viewed as having the potential to engineer HIV control by introducing cells resistant to the virus [18]. A clinical trial using gene-editing techniques successfully targeted and destroyed a gene in the immune system of 12 people living with HIV, increasing their resistance to the virus [18]. However, because of the invasive nature of stem cell therapy, it is not viable for the majority of people living with HIV since the body is likely to attack the donor cells as a sequel of transplantation [18].

The search for effective vaccine for HIV is still in progress and remains an active area of research. When presenting lectures on HIV/AIDS, as a United Nations Peer Educator on HIV/AIDS, I emphasize to participants that, currently one of the most effective alternatives to “vaccine” is public enlightenment. If there was a microbicide for HIV infection, as we have in bacterial diseases, it would have been applied per vaginam or per rectum to prevent the virus being passed on sexually and may kill the HIV or inhibit its replication. Development of HIV/AIDS vaccine is affected by a range of factors including continuous genetic variability of HIV, through mutation and recombination, which poses a

scientific challenge [19]. The vaccine scientists have to discover aspects of the virus that are consistent enough to provoke an effective immune response against multiple HIV variants and have the full understanding of the correlates of immunity. Since some potential vaccines work only against particular subtypes, one can suggest that prevailing subtypes in any geographical area should always be included in the vaccine. Such was the case in Thailand when an experimental vaccine was modified following a report of molecular epidemiologists that the predominant subtype B had been replaced with another subtype E in a population of injecting drug users among whom the trial was to be conducted [15].

The other problem is the wide variety of human populations who need protection and differ in their genetic make-up, including human leukocyte antigen diversity (HLA) and their routes of exposure to HIV. To add to these problems is the occurrence of HIV super infection which indicates that an immune response triggered by a vaccine to prevent infection by one strain may not protect against all other strains of HIV. This is one of the problems being addressed by the Council of Global HIV Vaccine Enterprises as well as Homsy and colleagues [20-21].

Perhaps a saving grace, while scientists are still battling with the problems of developing an effective HIV vaccine, is the Passive Immunotherapy (PIT) developed by Abraham Karpas [22]. He noted that infected individuals develop an early vigorous antibody response to infection which often lasts for several years. This led him to suggest that blood plasma collected from these “healthy” HIV infected individuals which contains antibodies that can neutralize and kill viruses including HIV could then be given to AIDS patients. Immunity to the virus in form of antibodies, is passively transferred from the “healthy” HIV individuals to the sick AIDS patients, hence the term passive immunization [22]. Results published as far back as 1988 suggest that passive immunotherapy is currently the best form of treatment for people with HIV disease, and therefore raises the prospect of reducing the spread of HIV in the population [22].

“Silent” HIV Carriers:

Perhaps there is no better way to introduce this section on HIV silent carries than paraphrase, what AVERT stated: that the spread of AIDS could quite conceivably have been induced by a combination of many different events [13]. Whether through injections, travels, wars, colonial practices or genetic engineering, the realities of the 20th century have undoubtedly had a major role to play. Nevertheless, a more pressing concern for scientists today should not

be how AIDS epidemic originated, but how those infected with HIV can be treated; how the future spread can be prevented and how the world can change to ensure that a similar pandemic never occurs again [13].

It is on this premise that we have to look at those seronegative, yet infected individuals who have never seroconverted and yet spreading the disease. These individuals are called “Silent” HIV Carriers. These individuals should not be confused with those at late stage of AIDS, when they are so immunocompromised that they are unable to mount an immune response to produce antibodies or those in the “long window” period or those with genetic “defect” to produce HIV-specific antibodies.

Research has shown that the widely assumed “latent period” (Window period) of HIV infection was utterly fictitious. Rather a “titanic” struggle erupts, perhaps from the time the virus first gains a foothold. In the struggle the body’s CD4+ T immune cells respond furiously, die daily in billions, are prodigiously replaced, but ultimately overwhelmed by fulminating HIV replication [23]. This may mean that even in the window period antibodies can still be produced if the cells are stimulated.

Why the concern about silent carriers [24]?

- They can transmit HIV and fuel the pandemic, thus negating all the efforts so far in containing the spread.
- Each unknowing HIV carrier can infect at least 50 or more individuals each year according to the hypothesis proposed by Frerichs, who extrapolates that about 12-14 million silent carriers roam the world.
- False negative individuals tend not to be retested 3 months later, which is the recommended practice; thus they may contribute to the spread of HIV.

As an example, a patient was assumed to be a silent carrier because she was consistently HIV seronegative; although she was seronegative until she died, she was not a silent carrier. Because we did not know what to call it we used the term AIDS-related illness [25] in line with what Onwubalili and colleagues had earlier described in a group of patients who also died from AIDS-related illnesses [26]. We then advised surgeons to be aware of such patients and endeavour to screen their patients before surgery [27]. These finding made me more cautious, during my United Nations years, working as HIV/AIDS Specialist and as community-based physician. It is possible that some of the high risk seronegative patients that presented could have been HIV silent carriers.

Knowledge of existing HIV strains in a particular geographic area is as important as knowledge of endemic diseases that can mimic HIV infection or affect HIV tests, such as chronic parasitaemia, for example due to malaria [28].

Three groups of basic HIV tests are:

- Antibody tests: Enzyme-Linked Immunosorbent Assay (ELISA) and Western Blot (WB) which is a confirmatory test.
- Antigen test also known as p24.
- Nucleic Acid Test: Polymerase Chain Reaction (PCR) an RNA or DNA-dependent Viral Load Assay.

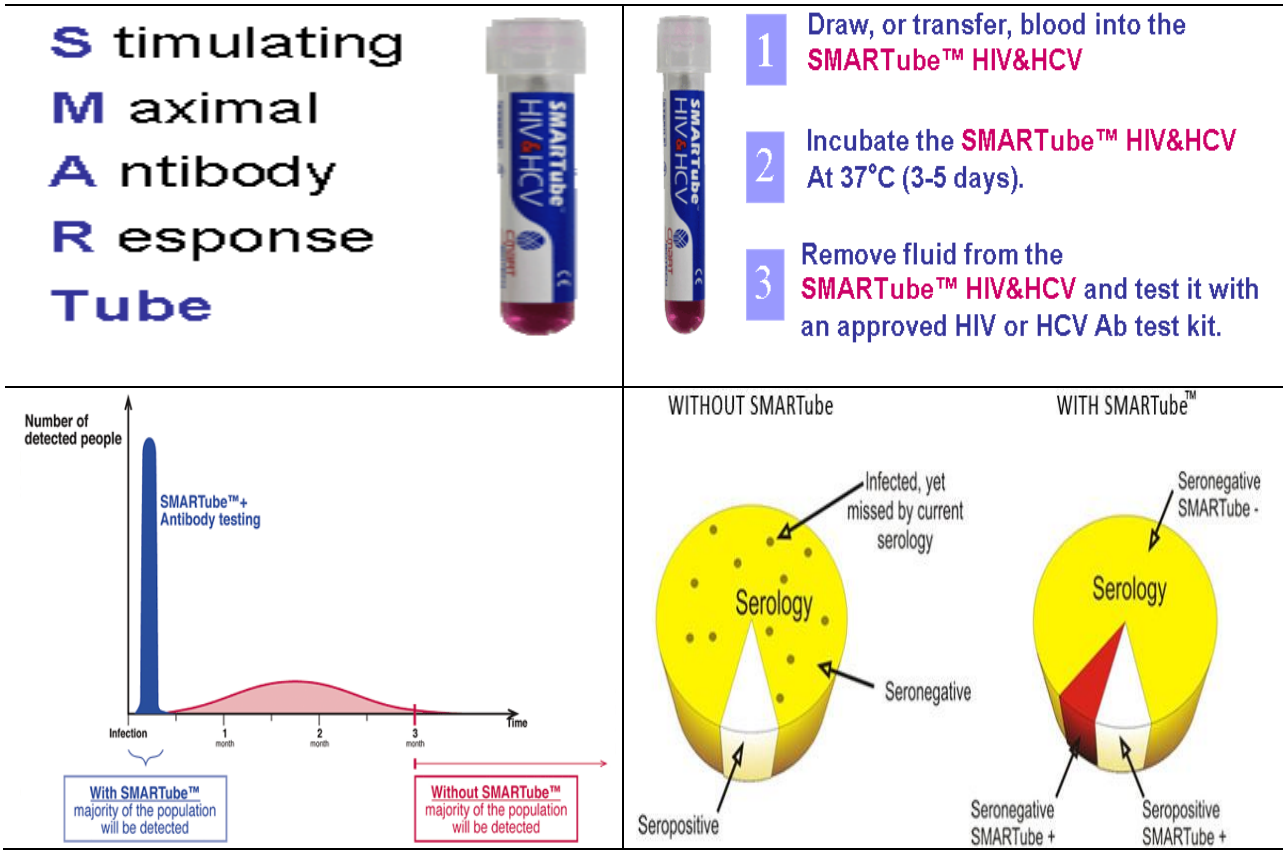
If both ELISA and WB are positive the chances are that in more than 99% of cases the patient is infected. It is critical that pregnant women, who seem to have a long window period, be tested because medications are very effective in reducing transmission from mother to child. A pregnant woman testing false-negative for HIV will not be offered anti-retroviral treatment which could have saved her baby. HIV tests may miss some infections, resulting in false negative results. This often occurs soon after infection when antibodies are just starting to form and are at a level too low to be detected. Other tests that have been used in HIV serology include the Particle Agglutination Test for HIV antibody developed by Fujirebio Inc. of Japan, the Karpas Cell Test and the HLA Testing developed in London Hospital Medical College [29]. Earlier attempts to detect HIV silent carriers have employed the PCR as well as the antigen capture test called p24. These tests have the disadvantages of either cost or technical complexity in performing them as in PCR or low sensitivity as well as false negative results which both of them share. These tests detect infections only when they have lasted several weeks. In the case of PCR false positive results have been reported [30] or failure to detect HIV [31] in a study involving 92 female prostitutes and their heterosexual partners of infected individuals.

The Stimmunology Test was developed to help resolve the problems of false negative results that tend to complicate the epidemiology of AIDS [32]. Stimmunology technique is an antibody enhancement pre-analytical procedure using immune potentiating agents. Stimmunology technique uses the Stimulating Maximal Antibody Response Tube- (SMART) Test. An overview of the procedure is illustrated in the diagram shown in Fig. 2 (adopted from Biotech Inc.).

SMARTube is not a diagnostic test; it is an in-vitro pre-test enhancement of antibody output, by stimulated B lymphocytes that were primed by HIV in the infected person, leading to early

detection of HIV infection even prior to seroconversion. Some of the advantages of Stimmunology are: Promotes early diagnosis (3-5 days); relatively cheap; Simple to perform; Simultaneous detection of both HIV and Hepatitis virus (HCV); Overcomes non-responsiveness due to immune-suppression; Does not change the current algorithm of antibody tests and diagnostics that are in use.

Fig. 2: The Stimmunology technique: Schematic representation of the procedure using the **S**timulating **M**aximal **A**ntibody **R**esponse **T**ube- (SMART) Test (Adopted from Biotech Inc. [33])



Although observation about the potential impact of HIV silent carriers was made over 20 years ago, the initial research to highlight the possible impact of silent retroviral infection carriers on the epidemiology of AIDS were conducted in nonhuman primates and later in humans [34-47].

The major results obtained from these studies indicated that when whole blood from seronegative subjects was stimulated most of them produced HIV-specific antibodies. One of such studies was conducted by Mumo et al. [41]. They reported that pre-treatment of whole blood sample in SMARTube containing immune potentiating agents promoted the synthesis and release of antibodies against HIV-1 prior to their detection in corresponding plasma samples in a group of donors who would have been otherwise classified as HIV-1 seronegative (Figure 2). This is critical for saving lives not only through a safer blood supply but also by detection of HIV infection among pregnant women who seem to have a very long window period. More so, since a pregnant woman testing negative for HIV, as correctly stated by Jehuda-Cohen, may not be given any anti-retroviral therapy that could have saved her baby [41].

In their study, using polyclonal B-cell activation test (PBA-T) and PCR among wives of seropositive HIV carriers in Ethiopian community in Israel, Jehuda-Cohen and colleagues reported that 8 of the 12 silent carriers studied were discordant for serum antibodies, yet 5 of the 8 seronegatives (66%) were found to be PBA-T and PCR positive for HIV [42]. Aiuti and coworkers, investigating 65 HIV silent carriers showed that 12 (18%) of them had HIV-1 proviral sequences by PCR. They concluded that prolonged seronegative individuals can transmit HIV through their body fluids [43].

DISCUSSION:

Medical science has taught us that anything hidden in the body is a threat to humanity. When cancer cells begin to grow in the body, they are not easily noticeable until you screen and identify them, after which management can commence. In the same way, most individuals do not know that they have essential hypertension, whose symptoms are silent. They may only be informed that their blood pressure is high during a regular medical check-up. Silent HIV infection is not an exception.

Several studies have indicated unambiguously that silent HIV carriers fuel the epidemic of AIDS. Ralph Frerichs, [24] in his extrapolation, contends that about 12-14 million silent HIV carriers roam the world. He indicated that “anonymous testing and absolute confidentiality,

as promoted by many public health officials, are self-defeating, making winners of the virus and losers of the people".

The impact of HIV silent carriers on the epidemiology of AIDS cannot be overemphasized. During the 6th International AIDS Society conference in Rome, Jehuda-Cohen [48] discussed the results of an independent multicenter study in five countries involving 5000 high-risk and 3000 low-risk silent carriers. The researchers were able to show that there were no false positive results in the low-risk population thus providing improved specificity, and that the use of SMARTube can identify early HIV infection, as shown by increased detection over regular plasma from infected person, thus improving sensitivity.

The study also showed that epidemiologically, the SMARTube Stimulation Index (ratio of antibodies in SMART plasma/plasma) indicates a low false recent rate, resulting in excellent specificity which is essential for determination of incidence (rate in which the epidemic is spreading). This they contended is the key to public health efforts in carrying out epidemiological studies to determine where high rate of new infections are occurring and for monitoring the efficacy of preventive programmes.

In conclusion, it was stated that both WHO and CDC are working towards a wide acceptance of new approaches, such as SMARTube, to combat the spread of HIV such as early and complete detection of HIV infections to facilitate the concept of "test and treat" which they support as a major advance in helping to curb the spread of HIV infection. It is therefore being advocated that strategies for preventing potential transmission from HIV-1 infected but seronegative individuals be addressed. Common sense dictates that failure to recognize and implement such strategies condones significant threat to the spread of HIV-1 among the world's most affected population. HIV antibody testing is the gold standard for HIV diagnosis. While it is quite effective for general use, conventional antibody-based testing for HIV infection does not allow for a thorough evaluation of a population's epidemic state since it excludes individuals within the "window period" which is estimated at 3 months for 95% of infected individuals [49, 50].

Thus, it is common practice that people at risk for HIV infection, who tested negative, are usually requested to come back for re-test 2-3 months later. Majority of them do not come back because they do not appreciate the need to do so. We

have to shift the paradigm of laboratory research from mere laboratory curiosities to result-oriented, problem-solving and cost-beneficial exercise. It is for these reasons that I recommend the simple cost-effective and reliable stimulating antibody assay - Stimmunology SMARTube assay for use in resource-poor setting to increase blood supply safety and quantity. Incorporating Stimmunology into basic blood bank testing and diagnostic protocols can also decrease preventable disease transmission.

" Life affords no higher pleasure than that of surmounting difficulties, passing from one step of success to another, forming new wishes and seeing them gratified" - Samuel Johnson

REFERENCES:

1. Brown, C. Berlin Patient spreads hope for AIDS cure. Washington Times. July 2012
2. Allers K, Hutter G, Hofman J, Lodden Kemper C, Rieger K, Thiel E, and Scneider T. Evidence for the cure of HIV infection by CCR5 Delta stem cell transplantation. Blood 117 (10) 2791-2799, 2011
3. Worobey M, GemmelM, Teuwen DE, Haselkorn T, Kunstman K, Bunce M, Muyembe JJ, KobongoJ-M M, Kalengayi RM, Marck EV, Gilbert TP and Wolinsky SM. Direct Evidence of Extensive Diversity of HIV-1 in Kinshasa by 1960. Nature 455: 661-664, 2008
4. Wolfe ND, Switzer WM, Carr JK, Bhullar VB, Shanmugan V, Tamoufe U, Prosser AT, Torimiro JN, Wright A, Mpoudi-Ngole E, McCutchan FE, Birx DL, Folks TM, Burke DS, and Heneine W. Naturally acquired simian retrovirus infections in Central African Hunters. The Lancet 20 (363) 932-937,2004
5. Zhu T, Korber BT, Nahmias AJ, Hooper E, Sharp PM and Ho DD. An African HIV-1 sequence from 1959 and implications for the origin of the epidemic. Nature, 391: 594-597, 1998
6. Center for Diseases Control and Prevention: A timeline of AIDS,1981
7. World Health Organisation. HIV/AIDS: Data and Statisitics.2012
8. Understanding how HIV causes AIDS. (http://www3.niaid.nih/topics/HIV/AIDS);
9. Chapel H, Haeney M, Misbah S and Snowden N. Immunodeficiency. In: Essentials of Clinical Immunology. London: Blackwell Scientific Publications, 4th Ed. 1999 : 68-75
10. Idoko JA. Management of opportunistic infections and malignancies in HIV/AIDS patients. Nig. J. Med 7: 21-24, 1988
11. Anyiwo CE and Ifeanyichukwu M. Anti-retroviral treatment in Nigeria: A review. J. Infect Chemother 6: 196-199, 2000
12. Plantier JC, Leoz M,Dickerson JE, Oliveira FD, Cordonnier F, Lemee V, Damond JE, Robertson DL and Simon F. A new HIV derived from gorillas. Nature Medicine 15: 871-872, 2009
13. AIDS Virus Educational and Research Trust (AVERT) HIV and AIDS Information 2014
14. Kanki PS, Sankale TL, and M'Boup S. Biology of HIV-2 In: AIDS in Africa. 2nd Ed. NY Academic Pub 2002 74-103
15. Anyiwo CE, Imai M, Igo JD, Ogunbanjo BO, Iwamoto A and Babona DV. Genotyping of Human Immunodeficiency Virus isolates in Papua New Guinea. Nig. Qt. J. Hosp Med 20, (40) 181-185, 2010

16. Burke DS. Recombination of HIV: An important viral evolutionary strategy. Emerg. Inf. Dis 3 (3) 253-257 , 1997
17. Associated Press. AIDS specialists release 'road map' for HIV cure. July 2012
18. Nature News. Gene-editing method tackles HIV in first clinical trial, March 2014
19. Essex M. Retroviral vaccines: Challenges for the developing world. AIDS Res Hum Retroviruses 12 : 361-363, 1996
20. The Council of Global HIV Vaccine Enterprise. The 2010 Scientific strategic plan of Global HIV Vaccine Enterprise. Nature Medicine 16:9, 981-988, 2010
21. Homsy J, Steimer K, and Kaslow R. Towards an AIDS vaccine: Challenges and Prospects. Immunology Today 718 : 193-196, 1989
22. Karpas A, Hill F, and Yoyle M. Effects of passive immunization in patients with acquired immunodeficiency syndrome-related complex and acquired immunodeficiency syndrome. Proc. Natl. Acad.Sci. USA.85: 9234-9237, 1988
23. Health Horizons. Infernal HIV: Do we finally have a handle on it? 25: 4-6, 1995
24. Frerichs JC HIV winners and losers. Epidemiology 6 (3) 329-331, 1995
25. Anyiwo CE and Azubike CO. Death from AIDS-like illness after surgery. Nig. J Surg Sci 4: 68-70, 1994
26. Onwubalili JK, Nwosu CM and Onuigbo WIB. Deaths from AIDS-like illnesses in West Africans. East African Med Journal 65:867-874, 1988
27. Anyiwo CE and Ikedife D. AIDS: Safe surgical practice. Nigerian Journal Surg Sci 3:82-85, 1993
28. Anyiwo CE. AIDS and HIV Testing in tropical developing countries. A monograph. Cambridgeshire, UK. Tropical Health Technology 1988
29. Anyiwo CE, Navarette C, Festenstein H, Bainbridge DR, Razak K, Colvin BT, Lowdell M, Macey M, Pinching AJ, Holbert M, Robinson D, Goh BT and Foster G. Expression of Class II human leukocyte antigen (HLA) on peripheral blood CD4 and CD8 cells of healthy subjects and HIV infected patients. Nigerian Journal Immun 2: 20-24, 1989
30. Jehuda-Cohen T. The False-positive polymerase chain reaction and the ostrich. J. Infect Dis 172:1420-1421, 1995
31. Luque F, Leal M, Pineda JA, Torres Y,Aguado I, Olivera M, Hernandez-Quero J, Sanchez-Quijano A, Rey C and Lissen E. Failure to detect silent HIV infection by PCR in subjects at risk for heterosexually transmitted HIV-1 infection. Eur J Clin Microbiol Inf Dis 663-667,1993
32. Mumo J, Vansover A and Jehuda-Cohen T. Detecting seronegative-early HIV infections among adult versus student Kenyan blood donors, by Stimmunology. Exp Biol Med 234(8) 931-939
33. SMARTube.(http://smartube-bio.com)
34. Jehuda-Cohen T, Powell JD, Villinger Y, Lockwood E ,McClure HM and Ahmed-Ansari A. Transmission of retroviral infection by seronegative blood in nonhuman primates. J Infect Dis 163(6): 1991; 1223
35. Jehuda-Cohen T, Powell JD, Villinger F, Mayne AE, Sell KW and Ansari AA. Evidence for simian immunodeficiency virus-specific IgM and IgG response in peripheral blood mononuclear cells of serum enzyme-linked immunosorbent assay-negative nonhuman primates. J Acquir Immune Defic Syndr. 7: 539-550,1994
36. Jehuda-Cohen T, Powell JD, Sell KW, Villinger F. Lockwood E, McClure HM and Ahmed-Ansari A. Transmission of retroviral infection by transfusion of seronegative blood in nonhuman primates. Journal Infect Diseases 163(6) 1223 - 1228, 1991

37. Jehuda-Cohen T, Villinger F, Powell JD, McClure HM, Sell KW and Ansari, AA. Presence of SIV antibodies in the sera of infants born to seronegative monkeys. Journal of Acquir Immune Defic Syndr 4 (2): 1991; 204 - 5
38. Jehuda-Cohen T, Slade BA, and Powell JD. Polyclonal B cell activation reveals antibodies against Human Immunodeficiency Virus Type 1 in HIV-1 Seronegative individuals. Proceedings of the National Academy of Sciences of the United States 87: 3972-3976, 1990
39. Brookmeyer R, Quinn T, Shepherd M, Mehendale S, Rodrigues J and Bollinger R.The AIDS epidemic in India: A new method for estimating current human immunodeficiency virus incidence rates. Am J Epidemiol 142: 709-713, 1995
40. Jehuda-Cohen T, Mumo JM, Bwayo JJ, and Pezzella M. Detection of HIV infection during window-period using polyclonal B-cell activation test. AIDS 11:124-125,1997
41. Mumo J, Vansover A and Jehuda-Cohen T. In-vitro antibody production enables HIV infection detection in window period-Key to safer blood. Exp Biol Med 234 (8) vi 2009
42. Jehuda-Cohen T, Vonsover A, Miltchen R and Bentwich Z. Silent HIV infection among wives of seropositive HIV carriers in Ethiopian community in Israel. Scan J Immun supp 11: 81-83, 1992
43. Aiuti F, Ensoli F, Fiorelli V, Mezzaroma I, Pinter E, Guerra E and Varani AR. Silent HIV infection.Vaccine11(5)538-541, 1993
44. Pezzela M, Rossi MA and Miceli M. Persistence of HIV-1 silent infection in seronegative subjects at high risk. J Med Virology 35: 14-18,1991
45. Varia D, Francois-Gerard CL and Doppagne A. Diagnosis by PCR of HIV-1 infection in seronegative individuals at risk. AIDS Res Hum Retro 6: 173-174,1990
46. Loch M, Mach B. Identification of HIV-1 infected seronegative individuals by a direct diagnostic test based on hybridization of amplified viral DNA. Lancet ii: 418-420, 1990
47. Potts M, Anderson R and Boily MC. Slowing the spread of HIV in developing countries. Lancet ii: 338:608-613, 1989
48. Jehuda-Cohen T, Saleh AR and Constantine NT. Novel in-vitro enhancement enables accelerated HIV pre-seroconversion diagnosis. Presentation at VI International AIDS Society Conference, Rome July,2011
49. Litvak E, Siegal JE, Pauker SG, Lallemant M, Fineberg HV and Weinstein MC. Whose blood is safer? The effect of the stage of the epidemic on screening for HIV. Med Decis Making 17: 455-463,1997
50. Shearer GM and Clerici M. Protective immunity against HIV infection: Has nature done the experiment for us? Immunol Today 17:21-24,1996.

STATUS OF MEDICINAL AND AROMATIC PLANTS IN PAPUA NEW GUINEA: A REVIEW

PREM P. RAI

Department of Pharmacy, School of Medicine and Health Sciences University of Papua New Guinea,
Email: raipp@yahoo.com.

ABSTRACT:

Medicinal and aromatic plants (MAPs) have a long history of use rooted in local cultures and traditions due to a range of their pharmaceutical, cosmetic, perfumery, dietary and nutritional applications. The medicinal and aromatic plants play a significant role in ensuring health security of millions of people globally. Although a very large segment of the population in Papua New Guinea (PNG) meets its healthcare needs through herbal medicines and products, very little has been done to develop this sector. It is argued that exploitation of MAPs as a source of livelihood is no longer a viable option due to population pressure, over exploitation, conflict of interests in land use and impact of climate change. Nevertheless global demand for herbal supplements and remedies is very high, and given the unique biodiversity and thousands of plants species growing in PNG this sector deserves special attention and effort to promote medicinal and aromatic plants through cultivation supported by adequate research, development and extension activities in order to ensure their better conservation and utilization in trade and industry. Due to geographical location and remoteness, access to health services is limited in several parts of PNG. The use of medicinal plants and traditional medicine is widespread among these communities, thus presenting both challenges and opportunities. PNG has one of the lowest health indicators in the Pacific. The question is how best MAPs can be utilized for improving the health conditions? This paper attempts to assess the current status of production, utilization and commercial prospects of medicinal and aromatic plants in PNG and identify future needs of conservation and opportunities for their development.

KEYWORDS: Medicinal, aromatic, plants, traditional medicine, herbal industry, biodiversity

Submitted March 2014; Accepted March 2014

INTRODUCTION:

(Geography, ecology, biodiversity, and economy of PNG)

Lying just south of the equator, 160km north of Australia, Papua New Guinea (PNG) is part of an arc of mountains stretching from Asia, through Indonesia into the South Pacific [1]. PNG is the world's largest and highest tropical island [1]. The country is richly diversified in languages, culture, customs, traditions, native flora and fauna [2]. It is one of the 'last frontiers' which truly defines magnificence and richness of diversity. Geologically, PNG has the highly mountainous, arid to rain forest, coastal swamps to alpine forest areas [2].

The country consists of more than 600 islands, the lowlands (0-1200m) and highlands (1200-2800m). As many as 800 ethnic groups exist, and 800-850 distinct languages are spoken in the country [2, 3, 4]. It is known that close geographical relationship between areas of high biological and language diversity exists and that particular cultural practices and linguistic tradition are compatible with high biodiversity [5, 6, 7].

PNG is the 4th mega-bio diverse country in the world [8]. It has 70% of its land covered by tropical rainforest. It covers less than 1% world land mass but contains more than 5% of biodiversity [3,9]. The total number of plants and animal species exceeds 200, 000 species [10]. Scientists estimate that more than half of the plants and animals found in PNG have yet to be scientifically named. Fortunately, much of this biodiversity has remained intact for thousands of years because of the ruggedness of the terrain that made the interior lands inaccessible. Coupled with low population density it also ensured that these biodiversity was never overexploited.

With more than 30 million hectares of closed tropical forests, PNG ranks 9th among the most forested tropical countries of the world [10,11]. Though estimates of total world species vary dramatically, it is estimated that 15,000 to 20,000 individual species of vascular plants may be found in PNG. Of these, perhaps 60% are endemic to PNG, one of the highest rates of endemism in the world [12]. PNG is home to some of the world's most unique species including the world's largest butterfly, moth, lizard, and crocodile [13]. It is the home of the bird of paradise, the national symbol, crowned pigeons, cassowaries, and many other exotic species. More species of orchids, sugar cane, parrot, pigeon and kingfisher exist in PNG than anywhere else on Earth. It is said PNG is a "cornucopia of ecology" [13].

PNG is a culturally diverse nation consisting of hundreds of distinct tribes. The lives of most of these tribal groups are intimately linked to their environment. They still survive through subsistence farming, food gathering and

hunting. Their food, housing, clothing and ritual materials are still largely obtained from their immediate surroundings. This extended habitation of diverse environs by various tribal groups has led to an especially rich tradition of medicinal plant use in PNG with well over 50% of the population relying exclusively on medicinal plants for health care [14]. The decline of the environment threatens the way of life for many of PNG's tribes.

PNG's economy struggled after gaining independence in 1975 and many of its institutions including national health systems went under-funded [15]. The national economic outlook is now improving, reflecting a flood of First World industries investing in the exploitation of the extensive gold, gas, timber and fishery resources of PNG [15,16]. In 2003 the PNG Gross Development Product (GDP) was under 4 billion USD, in 2013 it is just under 16 billion [16]. The first Liquid Natural Gas (LNG) project is on track to begin exportation in 2014, projected to contribute an additional 8 billion USD to PNG's GDP [16]. However, benefits to PNG's rapidly expanding population of 7 million are slow in materializing. PNG lags behind in almost all indicators of human development including health [17].

Medicinal and aromatic plants (MAPs) are produced and offered in a wide variety of products, from crude materials to processed and packaged products like pharmaceuticals, herbal remedies, teas, spirits, cosmetics, sweets, dietary supplements, varnishes and insecticides. Global market for MAPs has been growing at seven per cent annually, capitalizing on the growing awareness of herbal and aromatic plants worldwide [18]. A World Bank commentary has observed that "while commercial cultivation of medicinal plants is taking place on a miniscule scale, this activity is poised for 'dramatic growth' in the coming decade and favours organic and mixed cropping to ensure 'good agricultural practices' [19,20]. The use of botanical raw material is in many cases much cheaper than using alternative chemical substances [18]. Importance of MAPs becomes even more critical when we look at the intricate relationship that exists between biodiversity, ecosystem and human health. Nature remains the mainstay of medicines today as half or more of the prescribed medicines come from the natural sources, directly or indirectly [18]. This further emphasizes the need to preserve, protect and develop PNG's abundant natural resources for economic and health benefit of the population. The question that needs to be addressed is whether MAPs could be developed on a sustainable commercial scale for the benefit of all those involved.

MEDICINAL AND AROMATIC PLANTS:

Aromatic plants

There are an estimated 400 types of essential oils traded in the world with USD 5b in value. Global trade for flowers and fragrances were worth USD 22b in 2010 [21]. However, there

are no commercial or large scale plantings of medical and aromatic plants in PNG, and there is little marketing and trade of these materials. Small scale production of essential oils is limited to domestic markets only. There are hundreds of plant species that are used by indigenous people for sickness and physical ailments, but these plants are not commercialized. They are used mostly by rural people.

A number of plants have commercial potential but government policy and finance is needed to develop an essential oil industry. Although essential oils are not currently produced at commercial scale, an estimated 30 aromatic plant species have been identified for potential development and industrial application in PNG. Some of these are mentioned here.

Cryptocarya masoy (Massoia bark): The bark and hardwood contains C-10 lactone, golden colored oil. An experimental pilot plant has been set up in Central Province of PNG to investigate and explore commercial potential of the oil. Massoia lactone has an odour that is described as sweet, coconut meat, creamy, milky and waxy [22].

Asteromyrtus symphyocarpa (Waria-waria tree): Wari wari oil is obtained fromn Waria-waria trees that grow prolifically in PNG's Western province [22,23]. It's a type of melaleuca, and its leaves contain oil with basically the same medicinal qualities as eucalypt oil. The Australia's science agency CSIRO (Commonwealth Scientific Industrial Research Organization) and Australia Tree Seed Centre (ATSC) in partnership with the PNG National Forest Authority and the PNG Biological Foundation has established a sustainable essential oil industry in Western Province.

The oil is used to treat cough, pains and infections but other claims have also been made about the uses of benefits of the oil locally including solving baldness and curing malaria and so forth but basically the oil is cineole rich oil like medicinal eucalpytus oil and has very similar uses [24]. There are five distillation stills in three villages, but a viable industry is yet to be developed. The Waria-waria oil is already popular in the Port Moresby markets, but it's hoped that eventually the oil can find a wider market and create a viable local industry.

Santalum macgregorii (Papua sandalwood): Due to its wide use this tree is overexploited and considered a threatened species [24]. However, measures have been recently introduced for sustainable harvesting and conservation of this tree. Sandalwood oil is a pale yellow liquid and used as general fixative in almost any perfume type.

Pogostemon cablin (Patchouli; also called patchouly or pachouli) is a species of plant from the genus *Pogostemon* [24]. It is a bushy herb

of the mint family, with erect stems, reaching two or three feet in height and bearing small, pale pink-white flowers. The heavy and strong scent of patchouli has been used for centuries in perfumes, and more recently in incense, insect repellents, and alternative medicines.

Aquilaria agallocha (Agarwood): The tree grows on high altitude. Agarwood is a resinous wood that occurs in tree *Aquilaria agallocha;* the resin produced is rich dark in colour [24].

Elettaria cardamomum (Cardamom oil): The plant was introduced in highlands of PNG in 1960s. It is cultivated in high altitude mountainous region of highlands. Small communities are engaged in organic farming of cardamom as part of small enterprise that generates some income for the people [24].

Tagetes genus (52 spp.): Essential oil known as tagette for the perfume industry is produced from some of the species. A number of these species abound in PNG [24].

Morinda citrifolia (Noni juice and oil): Noni juice and oil is produced from the fruits and root subtracts and sold in local markets. There are some large Noni farms in the country that supply raw materials to these producers [25].

Zingiber officinale and *Curcuma longa* (Ginger and Turmeric): Ginger and turmeric belong to the family Zingiberaceae and are grown widely throughout PNG. In both plants the underground stem (rhizome) is the commercial product. There are several pharmacological applications for these species. Both these plants are used in traditional medicines for their medicinal properties [25].

Other plant spp.: *Polygala paniculata* (Indian snake-root), *Euodia* spp., *Cymbopogon citratus* (Lemon grass), *Piper aduncum*, *Piper gibblimbum*, *Alipina* spp., *Melaleuca leucadendron*, *Eucalyptus* spp., *Plantifolia vanilla* (Vanilla) [24].

MAPs GENETIC RESOURCES – CONSERVATION, DEVELOPMENT AND GERMPLASM COLLECTION:

Interest and support for the conservation and development of medicinal plants is increasing in all parts of the world including PNG. This is due, in part, to a growing recognition given to the role of medicinal plants in the provision of culturally relevant and affordable health care, in creating sustainable livelihoods and in the vital conservation of biodiversity. This has also drawn the attention of the world community towards the need for creating mechanisms to ensure sustained development of the sector and to allow sharing of information between countries, organizations and agencies. Although plants are widely used in the traditional systems of medicine in PNG it is only recently that some interest is being shown by the private sector and NGOs in this area [26].

Plant genetic resources (PGR) not only form the foundation of agriculture and agricultural research but also of medicinal plants. Requirement for diverse genotypes of medicinal and aromatic plants that give higher yield of specific compounds used in medicine will never cease in developing new varieties [27]. Unfortunately no work is going on in PNG in the area of germplasm collection, characterization, evaluation, etc. However the need and importance of variety development for work in future cannot be underestimated. Also very little effort has been made in variety development, and cultivation of medicinal and aromatic plants. None of the important plants are cultivated for processing or product development or export and commercial development. There is also no Government policy in place for promotion and commercial development of medicinal and aromatic plants at present.

Since the incorporation of National Policy on Traditional Medicine in 2007 [28] however, systematic survey and documentation of medicinal plants has been undertaken and still continuing. Further details are given in the succeeding paragraphs.

CONSTRAINTS AND OPPORTUNITIES:

PNG health system is built on modern western medicine, though traditional medicine is widely used both in urban and rural areas. There is need to develop herbal industry in PNG to meet the potential demands in global and domestic market. The necessary growth drivers for development of herbal industry are health concerns, side effects of conventional drugs, higher confidence, cultural acceptability, and competitive pricing. In the past two decades there has been increasing appreciation toward maintaining health with natural products vs. curing disease with chemical drugs [29,30]. There is also growing awareness of side effects of synthetic drugs. In recent years consumers are showing greater confidence in scientifically validated and quality products of traditional medicines. Besides, herbal extracts and powders are comparatively cheaper than synthetic drugs and formulations [29,30].

MEDICINAL PLANTS AND TRADITIONAL MEDICINE PRACTICES IN PNG:

Traditional medicine is an important part of the health system in PNG. The Government of PNG adopted the National Policy on Traditional Medicine in 2007 [28]. The policy aims to improve the quality and delivery of traditional medicine and its practices and identifies ways of integrating traditional medicine into the country's primary health care system. Traditional healers are permitted to practice at village and district level. Indeed traditional healers and medicinal plants have become important health resources in rural areas, particularly where aid posts and health centres have closed. The use of traditional medicine is very much part of the lives of local communities

throughout the country. Each ethnic group has a long tradition of using plants and other natural materials for treating illnesses. Although no official data exists it is estimated that traditional medicine accounts for almost half of all health care delivered in the country. It is the only form of health care available in some remote parts of the country [31]. The knowledge of traditional medicine is passed on from many generations verbally, and mostly to family members. Some traditional medicine practices are unique and of cultural significance. There is now concerted effort to document and safeguard the traditional knowledge of medicinal plant usage as it is a national heritage. In some areas traditional medical knowledge is still kept secret and cannot be released or shared easily. In general, there is good awareness but there is also a strong perception that traditional medicine is not being utilized to its full potential [31].

Currently medicinal plant preparations are used to treat various ailments such as sexually transmitted diseases, asthma, diarrhoea/ dysentery, body/abdominal pain, headaches, boils/sores, tuberculosis, cold/cough, fever/ malaria and insect bites [31]. According to a 1999 national report, 80% of the population in PNG are using herbal medicines and traditional medicine therapies [32]. For people living in the most remote parts of PNG, distance from public health services often means that a traditional healer is their only option [32].

There is acceptance of traditional medicine by doctors trained in conventional medicine, and traditional healers do not object to their patients also seeking conventional medical treatment [33]. This mutual tolerance and acceptance contributes to the majority of the population utilising both forms of treatment. Despite the diversity of ethnic groups in PNG, there are several common concepts and beliefs around health and illness, including a universal belief in the power of sorcery, belief in the importance of adherence to customary law, and belief in the healing power of herbs and incantation. It should be noted that the national policy on traditional medicine explicitly excludes the use of sorcery [28].

TRADITIONAL MEDICINE HEALTH CARE INITIATIVES:

Although the Department of Health in PNG has recognised traditional medicine as a valuable health resource, little has been done with the practitioners at the community level whose services are patronized by seemingly large number of people. As part of the traditional medicine programme of the National Department of Health (NDoH) a series of activities directed at incorporating traditional medicine in the national health system have been initiated in the past ten to twelve years. These include systematic survey of medicinal plants and traditional medicine practices, establishment of medicinal plants database, training of traditional healers in primary care,

traditional healers register and network, research into medicinal plants and development of national herbal formulary [34, 35, 36].

A national office for traditional medicine within the NDoH and the Traditional Medicines Task Force were established in 1999. NDoH officially endorsed medicinal plant use in 2005 through the announcement of a Traditional Medicines Health Care Initiative [17,35]. The Taskforce is now charged with promoting the National Policy on Traditional Medicine (2007) nationwide, selecting 'safe and effective' traditional medicines, developing a training manual for traditional practitioners in primary care, and formalizing Traditional Healer Guilds in the each province [36,37]. Traditional medicine program has featured in National Health Plans since 2001 [17,38]. Cost of traditional medicine is not covered by the government. This policy provides a sound basis for defining the role of traditional medicine in national health care delivery, ensuring that the necessary regulatory and legal mechanisms are created for promoting, maintaining and development of traditional medicine, and that the authenticity, safety, efficacy, quality and rational use of therapies are assured. Besides enabling wider health coverage, introduction of traditional medicine into primary health care will reduce government medical expenditures. This is important at a time of severe financial constraints that the country is facing, particularly in terms of the funds available for purchase of modern drugs and medical supplies, and for providing human resources to health centres and aid posts. There is limited information on the number of traditional medicine practitioners in PNG. The national traditional medicines database [31] lists over 850 practitioners but the total number may be much higher. There are currently no exclusive traditional medicine training or education programmes at college or university level and no traditional medicine research institute in PNG.

QUALITY AND SAFETY OF TRADITIONAL MEDICINE:

Currently there is no regulation for herbal medicines or the practice of traditional medicine, though laws relating to the National Policy on Traditional Medicines (2007) are currently in development. The WHO has strongly advocated the use of quality traditional medicines and developed guidelines for member countries to adapt [29]. The Traditional Medicines Database, first started in 1999, holds details on medicinal plants and traditional medicines, and is viewed as a national resource. The Traditional Medicines Task Force, established by the NDoH in PNG in 2004, has been tasked to identify candidate herbal medicines from the database for inclusion in the primary health care formulary.

Herbal medicines are sold in local markets with medical, health and nutrient content claims.

PNG has no pharmacopoeia. However, *Medicinal Plants in Papua New Guinea* written and compiled by Rai and colleagues was published in 2009 with support from the WHO regional office. This publication describes traditional uses of 126 medicinal plants [25]. The tradition and culture of using herbs for variety of sicknesses is well entrenched in communities across Bougainville, an autonomous region of PNG. Recent publications by Rai et al [39, 40, 41] have given account of some of the commonly used plants in traditional medicine in Bougainville and highland regions of the country.

The lives of most of the tribal groups in PNG are intimately linked to their environment. This extended habitation by various tribal groups of diverse environs has led to an especially rich tradition of medicinal plant use in PNG. At least 80% of Papua New Guineans use traditional medicines, especially in rural settings where HIV infection is most prevalent [42]. Although many of the early botanical expeditions collected notes on traditional use, they have not been systematically assessed with respect to medicinal records, nor disseminated.

The National Health Plan of PNG, 2001-2010, adopted by the NDoH, created a Traditional Medicines Working Group to assist in the development of traditional medicines in the country. As an outcome of this early work the University of Papua New Group initiated a country-wide survey on traditional medicine practice, the collection of medicinal plant voucher samples, and developed the above mentioned database. The database provides the Taskforce with validated information concerning particular medicinal plant uses. The database contains comprehensive information on up to 4000 traditional medicines derived from 1000 plant species. The database is dedicated to documentations and preservation of traditional knowledge concerning medicinal plant use in PNG and to serve as repository of indigenous knowledge in traditional medicine [31]. It is also used to identify safe and effective traditional medicine practices and promote their usage in the community.

There are four major categories in the database: prescription database, chemistry and pharmacology database, photo-Image database, and practitioner database. Medicinal plants of commercial potential are listed in Table 1, and the most commonly used plants utilized in PNG traditional medicine are listed in Table 2 [31].

Table 1: Distribution of medicinal plants of commercial potential in PNG

Plant species	Region in PNG	Name of oil
Cryptocaria massoy	All coastal regions	Massoia oil
Canaga odorata	All coastal regions	Canaga
Jasminum sp.	All coastal regions	Jasmine
Drimis peperita	Tabubil and some highlands region	Drimis
Pongostemon cablin	Most regions of PNG	Patchouli
Piper nigrum	Most regions of PNG	Pepper
Kaempfera sp.	Central province	Kaempfera
Santalum macgrogreii	Central province	Sandalwood
Aquilaria sp.	Most coastal provinces	Eaglewood
Euodia hortensis	Many provinces of PNG	Euodia
Rosa damacena	Cooler parts of highlands	Rose
Polyanthus tuberosa	All coastal regions	Tuberose
Mentha sp.	Highlands	Mint
Curcuma longa	All coastal regions	Turmeric
Elettaria cardamomum	Most parts of PNG	Cardamom
Acorus calamus	Highlands	Acorus
Piper gibbilimbum	Highlands	Gibbilimbol
Melaleuca platyphylla	Western province	Melaleuca

Table 2: The most common 100 medicinal plants used in PNG traditional medicine*

Abelmoschus manihot	*Derris elliptica*	*Morinda citrifolia*
Abrus precatorius	*Desmodium umbellatum*	*Ocimum basilicum*
Acalypha wilkesiana	*Dioscera alata*	*Pandanus tectorius*
Ageratum conyzoides	*Dioscorea bulbifera*	*Pangium edule*
Alstonia scholaris	*Eleusine indica*	*Paspalum conjugatum*
Alstonia spectabilis	*Endospermum formicarium*	*Passiflora foetida*
Angiopteris evecta	*Endospermum medullosum*	*Pedilanthus tithymaloides*
Areca catechu	*Epipremnum pinnatum*	*Persea americana*
Artocarpus altilis	*Erythrina variegate*	*Pipturus argenteus*
Asplenium nidus	*Euodia hortensis*	*Plectranthus scuttelariodes*
Averrhoa carambola	*Euphorbia hirta*	*Polygala paniculata*
Bidens pilosa	*Fagellaria indica*	*Pometia pinnata*
Bixa orellana	*Ficus adenosperma*	*Premna obtusifolia*
Breynia cernua	*Ficus copiosa*	*Premna serratifolia*
Calophyllum inophyllum	*Ficus septic*	*Psidium guajava*
Capsicum annum	*Ficus wassa*	*Pterocarpus indicus*
Carica papaya	*Flagellaria indica*	*Ricinus communis*
Cassia alata	*Gnetum gnemon*	*Rungia klossii*
Casuarina equisetifolia	*Hibiscus rosa-sinensis*	*Scaevola tacadda*
Casuarina papuana	*Hibiscus tiliaceus*	*Sida rhombifolia*
Catharanthus roseus	*Homalium foetidum*	*Solanum torvum*
Centella asiatica	*Hornstedtia scottiana*	*Solanum tuberosum*
Citrus limon	*Imperata cylindrical*	*Sphaerostephanos alatellus*
Cocos nucifera	*Inocarpus fagifer*	*Sphaerostephanos unitus*
Codiaeum variegatum	*Ipomea pes-caprae*	*Sterculia ampla*
Coleus blumei	*Jatropha curcas*	*Syndrella nodiflora*
Commelina paleata	*Kalanchoe pinnata*	*Syzygium malaccense*
Cordyline terminalis	*Laportea decumana*	*Terminalia cattapa*
Costus speciosus	*Macaranga aleuritoides*	*Timonius timon*
Crinum asiaticum	*Mangifera indica*	*Vitex trifolia*
Curcuma longa	*Manihot esculenta*	*Wedelia biflora*
Cyclandophora laurina	*Merremia peltata*	*Zingiber officinale*
Cymbopogon citratus	*Metroxylum sagu*	
Cyperus rotundus	*Mikania micrantha*	

* Based on data obtained from the National Database on PNG Traditional Medicine [Ref: 31]

FUTURE THRUST:

Rising consumer interest in use of natural and organic products, sustained demand for medicinal and aromatic plants worldwide [43] and growing acceptance of alternative and complimentary medicines by health policy makers make it prudent for country like PNG to formulate policies and plans to use these resources for economic and health benefit of its people. Scientifically validated and standardized herbal products are in great demand and data are available to develop some suitable products from PNG herbs [37,44].

Using proprietary information accumulated in the National Database on Traditional Medicine in PNG it is possible to identify effective herbal preparations for range of conditions such as energy/wakeup, relaxation/sleep aid, HIV, pneumonia, cough suppressant, topical antibiotic/antifungal, pain or headache, fever, etc. PNG offers huge scope for development of small, medium and large scale industrial production of medicinal and aromatic plants. Timber logging which is one of the main income generation activities at present is detrimental to the sustainability of flora and is impacting adversely on ecology and biodiversity of the land [12]. It is incumbent upon the relevant Government agencies and departments** to formulate policies and action plans to address this important but hitherto neglected area. There is also scope for private sector to explore and engage in economic ventures utilizing medicinal and aromatic plants. Commercial production of some of these plant species will generate much needed income for village communities, and also help conserve valuable plant resources.

CONCLUSION:

Global herbal supplements and remedies market is forecast to reach US $107 billion by the year 2017 [30]. PNG is known as a treasure house of valuable medicinal and aromatic plants [45]. It is a small nation with rich biodiversity. PNG has an estimated 20,000 vascular plant spp. that offers tremendous opportunity for exploration, cultivation, production, and commercialization of plant products [23]. So far little has been done in this regard. Government support in establishing herbal industry, cooperation and collaboration from private sectors, government agencies, research institutions, and universities are needed to develop this sector.

It is important that valuable plant species are not only conserved but also promoted for commercial cultivation in order to meet the increasing demand especially in the export markets. Efforts should be made to develop suitable herbal products from indigenous plant species with in-built quality control and standardization measures. It is believed that production and accessibility to quality herbal

products within PNG will have positive impact in improving health standards of its people. There is demand locally and overseas for quality herbal products. PNG, with its rich traditional knowledge and plant resources, need to harness this potential.

***Department of Environment and Conservation; Dept. of Agriculture; PNG Forest Resource Authority; National Forestry Research Institute of PNG; National Agriculture Research Institute; Dept. of Trade and Industry; National Dept. of Health; and Dept. of Planning.*

ACKNOWLEDGEMENTS:

This paper was presented by the author at the Expert Consultation on Promotion of Medicinal and Aromatic Plants (MAPs) in Asia and the Pacific Region, Bangkok 2-3 December 2013 under the title "Country report on the status of medicinal and aromatic plants in Papua New Guinea". The event was organised by the Asia-Pacific Association of Agricultural Research Institutions (APAARI) and sponsored by Food and Agriculture Organization (FAO). The author is grateful to PNG National Agriculture Research Institute (NARI) and Professor Topul Rali for sharing some information presented in this report.

REFERENCES:

1. Mittermeier RA, Gil PR, Mittermeier CG, and Megadiversity. Earth's Biologically Wealthiest Nations. Cemex, Monterrey Mexico, 1997.
2. Beehler BM (ed.) Papua New Guinea Conservation Needs Assessment, 1993. Vol. 2: Papua New Guinea Department of Environment and Conservation. Boroko, Papua New Guinea.
3. Reid WV. How many species will there be? In: Tropical Deforestation and Species Extinction, Whitmore, T.C. and Sayer, J.A. (eds). 1992. Chapman and Hall, London. pp. 55 -73.
4. Loh J and Harmon D. A global index of biocultural diversity, Ecological Indicators, vol. 5, pp. 231-241, 2005.
5. Maffi L. Linguistic, cultural, and biological diversity, Annual Review of Anthropology, vol. 29, pp. 599-617, 2005.
6. Harmon D. Losing species, losing languages: Connections between biological and linguistic diversity, Southwest Journal of Linguistics, 15, 89-108, 1996.
7. Papua New Guinea's 5th National Report to the Convention on Biological Diversity, 2010.
8. Swartzendruber JF, 1993. Papua New Guinea Conservation Needs Assessment - Synopsis Report. Biodiversity Support Program. The Nature Conservancy, World Resources Institute, Washington, D.C. and the Government of Papua New Guinea, Department of Environment and Conservation, Boroko, PNG, Publication No. 29.
9. World Wildlife Fund. The Coral Triangle - At the centre of marine biodiversity, 2007. http://assets.panda.org/downloads/coral_triangle.pdf
10. John RJ. Biodiversity and Conservation of the Native Flora of Papua New Guinea in Papua New Guinea Conservation Needs Assessment; Beehler BM. (ed) Government of Papua New Guinea, Department of Environment and Conservation, Boroko, PNG. 1993, 15-31.
11. Shearman PL, Bryan JE, Ash, J, Hunnam, P, Mackey, B, and Lokes, B, 2008. The

State of the Forests of Papua New Guinea; Mapping the extent and condition of forest cover and measuring the drivers of forest change in the period 1972-2002. Available from: www.scienceinpublic.com/State%20of Forests%20of%20PNG_Concise.pdf.

12. Shearman PL, Ash J, Mackey B, Bryan JE, Lokes, B. Forest conversion and degradation in Papua New Guinea 1972 – 2002. Biotropica. 2009, 41, 379-390.
13. PNG Embassy [cited 2007]. Environment & Conservation, Papua New Guinea Wildlife and Enviornment: PNG Government/NGO Approach to Conservation. www.pngembassy.org/environment.html
14. Rai PP, 2004. Traditional Medicine in Papua New Guinea: Policy and Practices, Proceedings of the National Workshop on Traditional Medicine Policy and Practices held in Port Moresby, 28-30 July, 2004; (Edited by Prem P. Rai), Printed by University of Papua New Guinea.
15. Government of PNG, 2012. Health Service Delivery Profile, PNG. Developed in collaboration with WHO and National Department of Health.
16. PNG LNG Economic Impact Study An assessment of the direct and indirect impacts of the proposed PNG LNG Project on the economy of Papua New Guinea Prepared for ExxonMobil, 6 February 2008 (revised April 2009).
17. Government of Papua New Guinea, National Health Plan 2001-2010. Volume II, Chapter 12.
18. Newman DJ and Cragg GM. Natural products as sources of new drugs over last 25 years; Journal of Natural Products 70, 461 – 477, 2007.
19. wbln1018.worldbank.org/sar/sa.nsf/a22044d0c4877a3e8567de00520fa/fae63d87e2bd14038525687f0057e0d1
20. Zhang, F. WHO guidelines on good agricultural and collecting practices for medicinal plants. World Health Organization, Geneva, 2003.
21. Medicinal Plants and Extracts, MNS-ITC Report December 2011, Geneva, Switzerland.
22. Rali T. University of Papua New Guinea, Papua New Guinea, 2013 (Personal Communication)
23. John R. The endemic flora of New Guinea, patterns and conservation of the flora, Science in New Guinea Journal, vol. 32, 63-69, 2012.
24. Womersley, JS. (Ed.). Handbooks of the Flora of Papua New Guinea, 1995, Papua New Guinea National Herbarium, Forestry Research Institute, Lae, Morobe Province, PNG.
25. WHO 2009. Medicinal Plants in Papua New Guinea, Written and compiled by Prem P. Rai together with Simon Saulei, Uma Ambi, and Teatulohi Matainaho; published by the WHO Regional Office for the Western Pacific Region, Manila (2009); copyright WHO, Manila, Philippines,
26. Rai, PP, Ströble S and Klink C. A collaborative approach to preservation and valorisation of traditional health care knowledge in the Autonomous Region of Bougainville, Science in New Guinea Journal, 32, 15-20, 2012.
27. NBPGR Vision 2050. 2013. National Bureau of Plant Genetic Resources IARI Campus New Delhi, India.
28. National Policy on Traditional Medicine. National Department of Health, Waigani, NCD, Papua New Guinea. 2007.
29. World Health Organization. Regional strategy for Traditional Medicine in the Western Pacific (2011-2020). Manila, WHO WPRO, 2012. Available on: www.wpro.who.int/publications/2012/regionalstrategyfortraditionalmedicine.pdf
30. www.prweb.com/releases/herbal_supplements/herbal_remedies/prweb9260421.htm; accessed 28 November 2012.
31. Rai, PP and Saulei, S. Establishment of a Database on Indigenous Traditional Medicine in Papua New Guinea, In: Reframing Indigenous Knowledge Systems, Research Methodologies &

Indigenous Pedagogies. (Ed. Steven Winduo); 2010, UPNG; pp: 79-88.
32. U. Ambihaipahar and Rai PP, 2004, 'Traditional Medicine in Papua New Guinea 2001-2010: Achievements, Plans and Projections', In: Traditional Medicine in Papua New Guinea: Policy and Practices, (Ed: Prem P Rai), Uni of Papua New Guinea, pp. 8-13.
33. Rai PP and Maibani G. The Role of Traditional Medicine Practitioners in Managing HIV and AIDS-related Sicknesses in two Provinces of Papua New Guinea, Pacific Journal of Medical Sciences, Vol. 10, No.2, pp.1-14, 2012.
34. Rai, PP. Traditional medicine surveys, database and taskforce activities. Paper presented at the International Cooperative and Biodiversity Groups annual meeting at University of Utah, Utah, USA, June 2011. Unpublished report
35. Rai, PP. Traditional medicine initiatives in Papua New Guinea. Paper presented at the Experts Consultation Meeting on Regional Strategy for Traditional Medicine in the Western Pacific, 2011-2020. Hong Kong, November 2010. Unpublished report
36. Rai PP, Traditional Medicine Database and its Potential Role in Primary Health Care in Papua New Guinea, Medical Sciences Bulletin, 2004, 2, 20-22.
37. Rai PP. Development of Traditional Medicine Based on Scientific Research, Pacific Journal of Medical Sciences, 2008, vol. 5, 61-64.
38. Government of Papua New Guinea, National Health Plan 2011-2020 – Volume 1 Policy and Strategies
39. Rai PP. 2012. Traditional Uses of Plants for Health and Healing in Bougainville Part I, published by HORIZONT3000, Madang, Papua New Guinea
40. Waruruai J, Sipana B, Koch M, Barrows LR, Matainaho TK, Rai PP. An Ethnobotanical Survey of Medicinal Plants Used in the Siwai and Buin Districts of the Autonomous Region of Bougainville, J of Ethnopharmacology, 2011, 138, 564-577.
41. Jorim RY, Korape S, Legu W, Koch M, Barrows LR, Matainaho TK and Rai PP. An Ethnobotanical Survey of Medicinal Plants Used in Eastern Highlands of Papua New Guinea, Journal of Ethnobiology and Ethnomedicine, 2012, 8, 47-64.
42. Rai PP and Maibani G. The Role of Traditional Medicine Practitioners in Managing HIV and AIDS-related Sicknesses in two Provinces of Papua New Guinea; Pacific J. Medical Sciences, 2012, Vol. 10, No.2, pp 1-14.
43. Kroner S. 2006. Finding key sales opportunities in the natural products marketplace. www.spins.com/news.
44. Salopuka G. Standardization of herbal medicines of Papua New Guinea with a particular focus on Alstonia scholaris Linn. R. Br, M. Med. Sc. Thesis, Uni of Papua New Guinea, 2012.
45. Barrows LR, Matainaho TK, Ireland CM, Miller S, Carter GT, Rai PP, Gideon O, Manoka B, Piskaut P, K Kiapranis, Bugni T, Noro JN, Pond CD, Andjeclic CD, Koch M, Powan E, Pole AR, Jensen JB , Making the Most of PNG's Biodiversity: Establishment of an Integrated Set of Programs that Link Botanical Survey with Pharmacological Assessment in "The Land of the Unexpected, 2009, Pharmaceutical Biology,47 (8), 795-808.

INSTRUCTIONS FOR AUTHORS:

AIMS AND SCOPE:

Pacific Journal of Medical Sciences is a peer-reviewed, multidisciplinary journal published by the School of Medicine and Health Sciences (SMHS), University of Papua New Guinea (UPNG).

The aim of The Pacific Journal of Medical Sciences is to provide the forum for researchers, medical practitioners and other professionals to record, publish, and share ideas and research findings that serve to enhance the understanding of the aetiology, symptoms, diagnosis, prevention, control and management of human disease conditions world wide.

The Pacific Journal of Medical Sciences publishes original scientific research reports, case reports, short communications, letters to the editor and reviews, representing new and significant findings in all areas of medical, biomedical and health sciences (including epidemiology, public and environmental health). Book reviews, scientific news and conference proceedings are published on special request.

EDITORIAL POLICIES:

The Pacific Journal of Medical Sciences (Pac. J. Med. Sci.) editorial policies require that: All manuscripts accepted for publication must reflect novelty and originality of quality research and be of interest to a multidisciplinary audience. All papers submitted for publication are peer-reviewed by two anonymous reviewers and the editor-in-chief or a designated member of the editorial board.

The editorial board may request for review articles, commentaries or short reviews on contemporary medical or biomedical issues that the board considered important to the advancement of the aims of the journal.

Original research papers should be both complete and concise; they should essentially offer conclusive results, but they should not exceed 7,500 words, including abstract, tables, figures and references.

Short communications and reports should not exceed 3500 words, including abstract, tables, figures and references. Review articles should not exceed 6,000 words, including tables, figures and references. Letter to the Editor should be brief and to the point.

On preliminary editing, all manuscripts that fail to meet the basic requirements indicated above and those that contain significant and obvious typographical errors are returned without further processing.

Manuscripts submitted will be reviewed and considered for publication only if they have not been published, simultaneously submitted or

already accepted for publication in another journal. The author responsible for correspondence must show evidence of approval of all co-authors when submitting a paper for publication.

All relevant ethical approval for research involving human and animal subjects must conform to the provisions of the Declaration of Helsinki in 1995 (as revised in Edinburgh 2000).

Only research methods that comply with internationally accepted principles of humane animal experimentation are accepted for publication in The Pacific Journal of Medical Sciences. The decision to accept, revise or reject any manuscript for publication in The Pacific Journal of Medical Sciences is the responsibility of the editor-in-chief; this is done after reviewing the reports and comments from the reviewers, in consultation with members of the editorial board.

Disclaimer:

All statements and opinions expressed in any of the manuscripts published in The Pacific Journal of Medical Sciences are of the authors and co-authors, and not necessarily of the editors or members of the editorial board. The editor-in-chief and members of the editorial board of The Pacific Journal of Medical Sciences disclaim any responsibility or liability for such material and do not guarantee or endorse any products or services mentioned in the articles, nor guarantee any claims made by the authors of the articles.

SUBMISSION OF MANUSCRIPT:

Manuscript should be written in clear and concise English and be intelligible to those that are not specialists in the particular scientific area. Manuscript that does not satisfy these requirements but is acceptable for publication in the Pacific Journal of Medical Sciences because of its essential scientific content will be returned to the authors for extensive and appropriate revision, as recommended by the reviewers and editors.

A covering letter to clarify the following should accompany any manuscript submitted for publication in the Pacific Journal of Medical Sciences: (a) That the scientific data contained in the manuscript has not been published or submitted for publication in any other journal; (b) That ethical clearance and permission for the research had been obtained from the appropriate committee(s) in the institution(s) where the work was carried out; (c) That all the authors have read and approved the content of the manuscript; (d) The name, address and email contact of the author responsible for correspondence and for communicating with others about revisions and final approval of proof of manuscript; (e) Statements quantifying the contribution of each author to the manuscript (*this is a requirement in the latest*

guidelines of the International Committee of Medical Journal Editors);

Only electronic copy of the manuscript sent as e-mail attachment should be submitted using the approved format indicated in the appropriate sections of this document.

Manuscript should be sent by email to any of the following: pacjmedsci@gmail.com.; templevictor@gmail.com;

PREPARATION OF MANUSCRIPT:

Manuscripts should be prepared on one side of A4 paper, using double-spacing. Pages are to be numbered consecutively in the bottom right-hand corner. Manuscript should include the following sections: Title page, abstract and keywords, text, acknowledgements, references, tables and figures.

Style: The Pacific Journal of Medical Sciences uses both UK and US spelling. Only one or the other should be used throughout a manuscript. SI units should be used for all measurements. Use abbreviations to avoid repetition of long technical terms: Indicate the abbreviation in parentheses when the word is used in full for the first time. Use the approved generic names of chemical substances and drugs. Do not use trade names or brand names of chemicals and drugs.

Title page: The following should be on the title page: (a) Title of the manuscript – it should be concise and informative; (b) Short running title of not more than 40 characters (optional); (c) Name of each author (first name, middle initial and last name), including highest academic degree; (d) Name and address of institution(s) in which the work was carried out; (e) Name, postal address and email contact of the author responsible for correspondence; Source(s) of research or other types of support for the research project, if any,

Abstract and key words:

The abstract should not be more than 300 words. The following should be clearly stated in the abstract: the purpose of the study, basic procedures, main findings (specific results and statistical significance, if any), and principal conclusions. Abbreviations and references should not be included in the abstract. Not more than 8 key words should be put below the abstract. Key words are used to assist indexers in cross-indexing published articles and may be published with the abstract. Medical Subject Headings (MeSH) list of the Index Medicus should be used for selecting key words (www.nlm.nih.gov/mesh/meshhome.html)

Text:

Text of an original manuscript should be separated into the standard IMRAD format as follows: Introduction, Materials and Methods, Results, Discussion. Sections on Acknowledgements and References should be included.

Introduction: This section should: (a) summarize relevant previous work, using appropriate references, without any extensive review of the subject; (b) clearly state the purpose of the study and summarize the rational for the study or observation; (c) avoid given any data on the work being reported.

Materials and Methods: This section should: (a) clearly indicate either the sampling procedure or observational subjects; (b) give appropriate references for established techniques and procedures; (c) new techniques and procedures and extensive modifications of existing ones should be presented in sufficient details so that other researchers can easily reproduce and evaluate them; (d) indicate appropriate quality control procedures used for laboratory methods and techniques; (e) indicate ethical procedures if either human subjects were involved [if informed consent was obtained from each subject] or if appropriate guide line for using laboratory animals were followed [see editorial policies above]; (f) indicate statistical methods used, if any.

Results: Data obtained should be presented in logical sequence in the text, tables and figures should be adequately explained to facilitate their interpretation. Avoid duplicating the results by repeating in the text all the data presented in the tables and figures. The text in the results section should only emphasize or summarize the important data.

Discussion: Major findings should be highlighted before the minor findings. All findings should be related to other relevant studies, if any, using appropriate references. Indicate the implications of the findings and their significance or limitations, including implications for future research. When warranted, propose new hypotheses with appropriate data, but be sure to clearly label them as such. The conclusions should be linked with the goals of the study and be clearly supported by evidence / data. Include recommendations, if applicable.

Acknowledgements:

The following should be acknowledged: Research or other financial grants; Material support, Contributions of Institutions, Colleagues, and other relevant participants.

References:

The Pacific Journal of Medical Sciences uses the Vancouver system of referencing. The references should be numbered, using Arabic numerals in square brackets, in the order in which they are first used in the text, tables, figures, and legends. In the reference section, list the references in the order of appearance in the text, tables, figures and legends. ***Abstracts, unpublished data, oral communications, and personal communications should not be included in the reference section***. All references should

be verified against the original documents. In the reference section, the names of all authors should be included. Avoid using "et al." in the reference section. Names of journals should be abbreviated, using the approved style indicated in Index Medicus/PubMed. References should be listed according to the examples given below:

Journal articles:

Brander LC, Buess H, Haldimann F, Harder M, Hanggi W, Herrmann U, Lauber K, Niederer U, Zurcher T, Burgi U, Gerber H. Urinary iodine concentration during pregnancy in an area of unstable dietary iodine intake in Switzerland. J Endocrinology Invest. 2003, 26 5: 389 – 396.

Book:

Gillett JE. The health of women in Papua New Guinea. PNGIMR: Kristen Press, 1991

Chapter in a Book:

Chaney SG. Principles of nutrition II: Micronutrients. In: Delvin TM, editor. Textbook of Biochemistry with Clinical Correlations, 4th ed. Brisbane: Wiley-Less, 1997: 1107– 36.

Published proceedings paper:

Kruse-Jarres JD. Basic principles of zinc metabolism. *In*: Kruse-Jarres JD, Scholmerich J, editors. Zinc and diseases of the digestive tract. Proceedings of the International Falk workshop, Germany, 1996: 3 – 15.

Tables:

Tables should be numbered sequentially in Arabic numerals, typed double-space on separate A4 paper for each table; vertical lines should not be used to separate columns. Each table should be self-contained with a comprehensive but concise legend/heading; column headings should be brief, with units in parenthesis. All non-standard abbreviations used in tables should be explained in footnotes, using the following symbols in this sequence: *, §, ¶, #, $.

Illustrations:

Graphs, line drawings, bar charts, maps, etc., should be labelled as 'figures' and numbered consecutively, using Arabic numerals. All figures should be drawn using computer graphics. Legends should be brief but understandable without referring to the text. Photographs should be unmounted sharp, glossy black and white prints. Photographs should contain scale bars, not magnifications. Colour photographs are not acceptable. Figures, reproduced from another source, should be clearly indicated and appropriate references and written permission from the copyright holder must be submitted with the manuscript.

Electronic copy of manuscripts (e-mail):

When a manuscript is accepted for publication, the corresponding author will be required to send an electronic copy of the corrected or

modified manuscript by email. All email attachments should be scanned with anti-virus software before sending. Automatic software for referencing, footnotes, headers, footers, etc., should not be used during formatting.

All manuscripts should be formatted using MS WORD.

GALLEY PROOFS:

Galley proof will be sent by email to the correspondent author. Only minor corrections should be made, no major alterations to the manuscript will be accepted. Galley proof should be returned within maximum 5 working days from the date of receipt. If any major modification is made, the manuscript will be rejected at this stage. Correspondent author should correct all printing errors and ensure that the typesetting is accurate in the galley proof. Note that the correspondent author, not the publisher will be responsible for any such errors, should they occur in the published paper.

REPRINTS:

Reprints will not be sent to authors, because the PJMS is an on-line journal. The Web-link will be sent by email to the corresponding author. One copy of a paper may be sent to the correspondent author on request only, subject to availability of funds to cover postage. Authors may download and print the journal at their own expense.

COPYRIGHT:

Manuscripts accepted for publication in The Pacific Journal of Medical Sciences become the property of the journal. Therefore, all authors **may** be requested to sign a transfer of copyright form and send scan copy by email attachment to the editorial office. The copyright gives the journal the exclusive rights to reproduce, translate and distribute the article for academic purposes.

CHECKLIST FOR AUTHORS:

- Running title included (Can be omitted if not applicable);
- Abstract prepared according to instructions and include key words;
- Manuscript typed double-space on one side of A4 paper;
- References cited in square brackets in text and listed according to approved style for this journal; Uniform spelling throughout the text;
- Tables and Figs on separate A4 pages;
- Covering letter written as required by this journal;
- E-mail address of corresponding author indicated.