Sumerianz Journal of Biotechnology, 2018, Vol. 1, No. 1, pp. 25-29
ISSN(e): 2617-3050, ISSN(p): 2617-3123
Website: https://www.sumerianz.com

Original Article Open Access

Soft Drinks Associate with Low Levels of Some Bone and Infertility Markers in Women

Amadu A. A.[*]
Deparment of Chemical Pathology and Immunology, Faculty of Clinical Science College of Health Sciences, Bayero University Kano Nigeria

Kabir N.
Department of Biochemistry Faculty of Science Federal University Dutse, Jigawa State Nigeria

Bolori M. T.
Department of Community Medicine, University of Maiduguri, Maiduguri, Nigeria

Ahmad I. M.
Department of Biochemistry, Kano University of Science and Technology, P.M.B 3244 Kano Nigeria

Yunusa I.
Department of Biochemistry, Kano University of Science and Technology, P.M.B 3244 Kano Nigeria

Abstract

The consumption of soft drinks has increased drastically over the decades; important concerns raised are the medical and health implications of the increased consumption. We aimed to assess the effect of soft drink consumption on the serum levels of some bone and infertility markers in women. The biochemical investigations for serum calcium and phosphorus concentrations was carried out with Random Access Biochemistry auto-analyzer (KENZA 240 TX) using Bio-Labo diagnostics kits, while serum hormonal levels; prolactin, Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH), progesterone, estrogen and vitamin D were assayed using Abbot Architect i1000 SR (USA) reagents. A total of 128 newly diagnosed women with primary infertility were screened; out of which 112 (87.5%) consumed soft drinks while 16 (12.5%) do not. Sixty-eight (53.1%) participants consumed more than 4 bottles per day and orange soft drinks happens to be the drink of choice for 40 (31.2%) participants. A significantly high level of serum inorganic phosphate ($P < 0.05$) in subject that consumed soft drinks was observed. Serum levels of calcium and vitamin D showed no significant difference ($P > 0.05$) with consumption. Serum level of progesterone was observed to be significantly ($P < 0.05$) low in subjects consuming soft drinks. Consumption of soft drinks might be associated with high inorganic phosphates and reduced serum level of serum progesterone.

Keywords: Bone markers; Infertility; Soft drinks.

1. Introduction

Infertility is defined as failure to achieve clinical pregnancy after 12 months or more of regular unprotected sexual intercourse in a woman of reproductive age [1]. It affects more than 50 million couple worldwide [2]. Infertility rate is low in Africa compared to the developed countries [3]. Ikechebulu, *et al.* [4] evaluated 314 couples from south eastern Nigeria out of which 214 (65%) had primary infertility while 110 (35%) had secondary infertility. Hormonal imbalance has been considered of great importance in the causes and diagnoses of female infertility. Luteinizing hormone levels in the blood rises sharply by about 6 to 8 folds within 24 to 48 hours before ovulation. Follicle stimulating hormone (FSH) is responsible for the growth and development of the Graafian follicle that when matured releases the ovum or egg. The Grafian follicle also releases estrogen. When ovulation is due, the pituitary gland releases LH which causes the release of the egg from the ovary. The rise in estrogen has a negative feedback mechanism on the pituitary gland to control FSH secretion and to start making more LH. This LH causes eggs to be released from the ovary, process called ovulation [2].

Important concerns have been raised over the years regarding the medical and health implications of increased soft drink consumption [5]. Detrimental effects of soft drink consumption include enamel softening [6], hypokaleamic myopathy [7], diabetes mellitus [8], chronic kidney disease [9], low bone mineral density (BMD) [10], osteoporosis [11], spontaneous abortion [12]. Caffeinated soft drinks have been linked with delayed conception [13] and infertility (ovulatory) [14].

Soft drinks contain phosphoric acid which interferes with calcium absorption and contributes to imbalance that leads to additional loss of calcium [15] it is also involved in steroid biosynthesis in the adrenal glands and ovaries conversion of pregnenolone to progesterone [16]. This affects the activity of cyclic adenosine mono-phosphate (cAMP), the release of LH [17] and follicle selection [18]. Calcium is also required to initiate acrosomal reaction with its attendant release of enzymes, and membrane alteration needed for successful egg-sperm interaction [19].

*Corresponding Author

Vitamin D deficiency was reported to be associated with soft drink consumption in premenopausal women [20]. There exists a link between insufficient circulation of vitamin D concentration, health outcomes and low bone mineral density (BMD) [21]. Cola soft drinks have been identified to be linked with decrease concentration of plasma vitamin D in rats [22]. There is however some evidence which shows that apart from being the classical steroid hormones, vitamin D also modulates reproductive processes in both men and women [23-25]. Biological actions of vitamin D are mediated through the vitamin D receptors (VDR) that are distributed across various tissues including skeleton, parathyroid glands as well as reproductive tissues [26-28]. Lower levels of serum vitamin D are associated with lower testosterone, estradiol and LH [29]. Hence this study aims to assess the effect of soft drinks consumption on the serum level of the bone and infertility markers among women in Kano, one of the most populous states in Nigeria.

2. Materials and Methods

2.1. Location

This cross-sectional study was carried out at Aminu Kano Teaching Hospital (AKTH), Kano Nigeria

2.2. Study Design

This was a cross sectional descriptive study.

2.3. Inclusion Criteria

The Study included all the women who were on visit to Aminu Kano Teaching Hospital on account of primary and secondary infertility.

2.4. Exclusion Criteria

Women who were either below or above age range of 17 to 50 years, women diagnosed as diabetic patients, or with cardiovascular conditions, and those with acute/chronic kidney diseases were excluded. Others included those with previous or current pituitary, thyroid or parathyroid disorders or surgery and those with already identified cause of inability to conceive.

2.5. Ethical Consideration

Ethical clearance was obtained from the Aminu Kano Teaching Hospital. Informed consent was also obtained from each participant for the purpose of the study.

2.6. Sample Collection

Venous fasting blood sample ($5cm^3$) was collected in a plain tube. The blood was allowed to clot and centrifuged at 5000 rpm for four minutes and serum harvested. The samples were refrigerated at -20^0C before analysis.

3. Methods

The biochemical investigations for serum calcium and Phosphorus was done by Random Access Biochemistry auto-analyzer (KENZA 240 TX) using Bio-Labo diagnostics kits, while serum hormonal levels of prolactin, follicle stimulating hormone (FSH), Luteinizing Hormone (LH), progesterone, estrogen and vitamin D were assayed by Abbot Architect i1000 SR (USA) using abbot Architect reagents**.**

4. Statistical Analysis

Results are presented as mean and standard error of mean (Mean ± SEM). Differences in the parameters between study group and controls were tested with Students' T - test. Data were analyzed with SPSS (Statistical Packages for Social Sciences) version 16.0 Inc., Chicago, IL, USA). Values were considered significant when $P < 0.05$.

5. Results

Table-1. Demographic distribution of women with infertility

	Population (%)	**Frequency (%) Yes**	**Frequency (%) No**
Tribes Hausa	76 (59.4)	72 (94.7)	4 (5.3)
Yoruba	24 (18.8)	12 (50.0)	12 (50.0)
Ibo	20 (15.6)	20 (100.0)	0 (0.0)
Others	8 (6.2)	8 (100.0)	0 (0.0)
		X^2 =38.496; p=0.000***	
Occupation			
House Wife	60 (46.9)	52 (86.6)	8 (13.4)
Business	40 (31.2)	32 (80.0)	8 (20.0)
Civil Servant	28 (21.9)	28 (100.0)	0 (0.0)
		X^2 =6.095; p =0.047*	

Age (yrs)			
15-20	12 (9.4)	12 (100.0)	0 (0.0)
20-25	4 (3.1)	4 (100.0)	0 (0.0)
25-30	32 (25.0)	28 (87.5)	4 (12.5)
30-35	20 (15.6)	20 (100.0)	0 (0.0)
35-40	24 (18.8)	16 (66.7)	8 (33.3)
40-45	16 (12.5)	12 (75.0)	4 (25.0)
45-50	20 (15.6)	20 (100.0)	0 (0.0)
		X^2=19.810; p=0.003**	

NS=non-significant; S=significant; *=significant; **=very significant; ***=highly significant

Table-2. Frequency and type of soft drinks consumption in study subject

Qty. Per Day	Type(35 cl)	Frequency (%)
1 per day	Orange soft drinks	32 (25.0)
2 per day	Cola soft drinks	68 (53.1)
3 per day	Orange soft drinks	8 (6.2)
4 per day	Others	4 (3.1)
None	None	16 (12.5)

Table-3. Effect of soft drinks on markers of infertility in female

Bio-markers	Soft Drinks Consumption				
	Yes	No	Normal Range	Units	P-value
Calcium	1.98±0.15	2.07±0.15	2.10-2.80	mmol/L	0.057^{NS}
Inorganic Phosphate	1.85±0.54	1.49±0.44	0.94-1.70	mmol/L	0.004**
Vitamin D	20.09±4.60	20.23±2.60	20.00-50.00	ng/mL	0.909^{NS}
Progesterone	0.32±0.32	1.61±3.37	1.00-20.00	ng/mL	0.000***
Prolactin	273.21±206.90	325.50±200.34	72.00-511.00	mu/L	0.344^{NS}
FSH	6.78±4.10	12.38±14.08	4.70-21.50	miu/L	0.001**
LH	8.64±3.75	10.00±6.68	18.00-12.00	miu/L	0.438^{NS}
Estrogen	412.75±209.01	472.00±142.70	96.00-436.00	pg/mL	0.275^{NS}

NS=non-significant; S=significant; *=significant; **=very significant; ***=highly significant

Table 1 shows the socio-demographic characteristics of the studied subjects according to tribe, occupation, age group and lifestyle. Out of One hundred and twenty-eight participants recruited for the study, 76 (59.4%) were Hausa, 24 (18.8%) Yoruba, 20 (15.6%) Ibo and 8 (6.2%) other tribes. From the study sample, 112 (87.5%) consumed soft drinks while only 16 (12.5%) do not. Sixty (46.9%), 40 (31.2%) and 28 (21.9%) study participants were housewives, business owners and civil servants respectively. Fifty two housewives (86.6%) were found to be consuming soft drinks. Twenty-eight (87.5%) participants consuming soft drinks were observed to be within age group 25-30 years. Table 2 shows the frequency, quantity and the type of soft drinks consumed by the study subjects. Sixty-eight (53.1%), 32 (25.0%), 8 (6.2%) and 4 (3.1%) participants drank 4 bottles (35 CL) of Cola soft drinks, 1 (35CL) bottle of orange soft drink, 3 bottles(35 CL) of orange soft drink and 4 bottles of other type of soft drink (not 35 CL) per day.

Table 3 shows the effect of soft drinks consumption of some biomarkers of bone and infertility. From the results, serum calcium levels of study participants consuming soft drinks compared with controls (1.98±0.15mmol/L vs. 2.07±0.15mmol/L) were found to be not significantly different ($P > 0.05$). A significantly high ($P > 0.05$) levels of serum inorganic phosphate (1.85±0.54mmol/L) were observed in participants who consume the drinks compared to the control group (1.49±0.44mmol/L). Serum progesterone was found to be significantly reduced ($P < 0.05$) in study participants consuming the drinks (0.32±0.32ng/mL) compared to controls (1.61±3.37ng/mL). Serum vitamin D, prolactin, FSH, LH and estrogen levels in study participants consuming soft drinks were observed to be not significantly different ($P > 0.05$) to the control group. However, weak positive correlation ($r = 0.315$, $P = 0.05$) between soft drink consumption and the serum concentration of calcium as well as weak negative correlation ($r = -0.025$, $P = 0.05$) between soft drink consumption and serum concentration of vitamin D were observed. A strong negative correlation ($r = -0.547$, $P = 0.05$) was found between soft drink consumption and the serum concentration of inorganic phosphate. This study indicated that most infertile women did consume more than one soft drink per day. Kassem, *et al.* [30] reported 96.3% consumption of soft drinks in female teenagers aged 13-18. Lew and Barlow [31] reported 49.4% female teenagers consume soft drinks 2-5 times a week and 32.5% consumed soft drink daily. According to Tucker, *et al.* [32], Coca-Cola was the soft drink of choice taken at least 5 times per week per person among women in the age bracket 29 to 83 yrs. The study showed that serum levels of inorganic of those consuming soft drink were high than those in the control group. This may lead to the formation of calcium-phosphorus complex making calcium unavailable for absorption and hence triggering the stimulation of parathyroid hormone release. Fernando, *et al.* [33]; Mazarigos-Ramos, *et al.* [34]; Heany and Rafferty [35] reported higher parathyroid hormone, hyper-phosphotaemia and low serum calcium in post-menopausal women who consumed more than one bottle of soft drink per day. Since calcium dependent mechanism are involved in steroid biosynthesis in the adrenal glands and ovaries. Calcium plays a critical role in the metabolism of the cells of the reproductive

system and it is required for the activity of cyclic adenosine mono-phosphate (cAMP) and the release of LH [17]. In the light of this effect on calcium brought about by soft drink consumption, the low Calcium level may be one of the risk factors for infertility.

Our study found lower level of progesterone in participants consuming soft drinks. Moltz, *et al.* [36] reported lower hormonal levels of progesterone, estradiol, FSH and LH in infertility. Furthermore, lower levels of FSH and LH may result inhibition of Graffian follicle formation. This further reduces estrogen level which needed to stimulate the endometrium Olootu, *et al.* [37]. The condition where the bones become brittle and fragile as a result of loss of tissue brought about by hormonal changes, and deficiency of calcium is referred to as Osteoporosis. The body maintains a steady state of Phosphorus and calcium ratio in the blood to enable formation of new bones and remodeling of old ones. Therefore, if there is increased intake of soft drinks containing phosphoric acid, phosphates in the blood increases, leading to urinary loss of calcium. The loss of calcium in the blood leads to the activation of parathyroid hormone (PTH), causing the release of calcium from the bones for maintaining the balance. When this process continues over a long period of time, it results into weakness of the bone [32]. There has been an association between soft drinks and bone mineral density (BMD) in women [32], this is adduced to the fact that women have smaller bones overall and are at a higher risk of osteoporosis and nutritional anomaly due to lesser physical activities [32]. This study did not observe significant change in serum vitamin D levels with soft drinks consumption. Though an experiment conducted in rats found out that sugar sweetened beverages decrease the concentration of plasma vitamin D [22].

6. Conclusion and Recommendation

Consumption of soft drinks was associated with significantly high inorganic phosphates and reduced serum level of progesterone which may predispose to infertility. However, the replication of our findings in another experimental setting is recommended.

References

[1] WHO, 2013. "Infertility definitions and terminology, human reproductive programme."

[2] Maya, N., Seth, R. F., Ties, B. S., and Vanderpoel, G. A. S., 2012. "National, regional and global trend in infertility: a Systematic analysis of 277 healthy surveys." *Plos. Med.*, vol. 9, p. e1001356.

[3] Gerais, A. S. and Rushwan, H., 1992. "Infertility in Africa." *Popul. Sci.*, vol. 12, pp. 25-46.

[4] Ikechebulu, J. I., Adinma, J. I., Orie, E. F., and Ikegwuonu, S. O., 2003. "High prevalence of male infertility in the southeastern." *Nigeria J. Obstet. Gynecol*, vol. 23, pp. 657-659.

[5] Atsushi, I., Satoshi, I., Hiroshi, T., Kazutoshi, M., Suzuki, N., and Yamamoto, A., 2010. "Effects of cola intake on fertility: a review." *Health*, vol. 2, pp. 997-1001.

[6] Jensdottir, T., Holbrook, P., Nauntofte, B., Buchwald, C., and Bardow, A., 2006. "Immediate erosive potential of cola drinks and orange juices." *Journal of Dental Research*, vol. 85, pp. 226-230.

[7] Packer, C., 2009. "Cola-induced hypokalaemia: A supersized problem." *International Journal of Clinical Practice*, vol. 63, pp. 833-835.

[8] Acheson, K., 2005. "Caffeine and insulin sensitivity." *Metabolic Syndrome & Related Disorders*, vol. 3, pp. 19-25.

[9] Saldana, T., Basso, O., Darden, R., and Sandler, D., 2007. "Carbonated beverages and chronic kidney disease." *Epidemiology*, vol. 18, pp. 501-506.

[10] McGartland, C., Robson, P. J., and Murray, L., 2003. "Carbonated soft drinks consumption and bone mineral density in adolescence: the Northern Ireland Young Hearts project." *J. Bone Miner Res.*, vol. 18, pp. 1563-1569.

[11] Rapuri, P. B., Gallagher, J. C., Kinyamau, H. K., and Ryschori, K. I., 2001. "Caffein intake increases the rate of bone loss in elderly women and interacts with vitamin D Receptor genotypes." *Am. J.Clin.Nutri*, vol. 74, pp. 694-700.

[12] Signerrello, L. and McLaughlin, J., 2004. "Maternal caffeine consumption and Spontaneous abortion: A review of the epidimologic evidence." *Epidimology*, vol. 15, pp. 229-239.

[13] Bolúmar, F., Olsen, J., Rebagliato, M., and Bisanti, L., 1997. "Caffeine intake and delayed conception: A European multicenter study on infertility and sub-fecundity." *American Journal of Epidemiology*, vol. 145, pp. 324-334.

[14] Chavarro, J., Rich-Edwards, J., Rosner, B., and Willett, W., 2009. "Caffeinated and alcoholic beverage intake in relation to ovulatory disorder infertility." *Epidemiology*, vol. 20, pp. 374-381.

[15] Amato, D., Maravilla, A., and Montoya, C., 1998. "Acute effects of soft drink intake on calcium and phosphate metabolism in immature and adult rats." *Rev. Invest. Clin.*, vol. 50, pp. 185-189.

[16] Seifi, H., Farzaneh, N., and Mohri, M., 2005. "Relationship between fertility, serum calcium and inorganic phosphate in dairy cows." *Iranian Journal of Veterinary Research*, vol. 6, p. 12.

[17] Hurley, W. L. and Doane, R. M., 1989. "Recent development cause of infertility of lactating cows." *Brit. Vet J.*, vol. 124, pp. 126-130.

[18] Thys-Jacob, S., Donovan, D., Papadopoulus, A., Sarrel, P., and Bilezikian, J. P., 1999. "Vitamin D and Calcium Dysregulation in Polycystic ovarian syndrome." *Steriod*, vol. 64, pp. 430-435.

[19] Yanagimachi, R., 1981. *Mechanism of fertilization in mammals in fertilization and embryonic development invitro. Mastrioanni Jr,L. and Biggers,J.D (eds)*. New York: Plenum press. pp. 88-182.

[20] Duchaine, S. and Diorio, C., 2014. "Association between Intake of Sugar-Sweetened Beverages and Circulating 25-Hydroxyvitamin D concentration among premenopausal women." *Nutrients*,

[21] Ross, A. C., Manson, J. E., Abrams, S. A., Aloia, J. F., Brannon, P. M., Clinton, S. K., Durazo-Arvizu, R. A., Gallagher, J. C., Gallo, R. L., *et al.*, 2011. "The 2011 report on dietary reference intakes for calcium and vitamin d from the institute of medicine: What clinicians need to know." *J. Clin. Endocrinol Metab.*, vol. 96, pp. 53-58.

[22] Garcia-Contreras, F., Paniagua, R., and Avila-Diaz, M., 2000. "Cola beverage consumption induces bone mineralization reduction in ovariectomized rats." *Arch. Med. Res.*, vol. 31, pp. 360–3365.

[23] Eyles, D. W., Smith, S., Kinobe, R., Hewison, M., and Mc Crath, J. J., 2005. "Distribution of vitamin D receptors and 1-alpha-hydroxylase in human brain." *Journal of Chemical Neuropathy*, vol. 29, pp. 21-30.

[24] Blomberg, J. M., Nelsen, J. E., Jorgensen, A., Raypert-De, M. E., Kristensen, D. M., and Jorgensen, N., 2010. "Vitamin D receptors and Vitamin D metabolizing enzymes are expressed in the human male reproductive tract." *Human Reproduction*, vol. 25, pp. 1303-1311.

[25] Elizabeth, L. and Barbara, O.-P., 2012. "Vitamin D and fertility: A systematic review." *European Journal of Endocrinology*, vol. 106, pp. 765-778.

[26] Kinuta, K., Tanaka, H., Moriwake, T., Aya, K., Kato, S., and Seino, Y., 2000. "Vitamin D is an important factor in Estrogen biosynthesis of both female and male gonads." *Endocrinology*, vol. 141, pp. 1317-1324.

[27] Agic, A., Xu, H., Altgassen, C., Noack, F., Wolfler, M. M., Diedrich, K., Friedrich, M., Taylor, R. N., and Hornung, D., 2007. "Relative expression of 1,25-dihydroxy vitamin D3 Receptor, Vitamin D,1-A hydroxylase, Vitamin D 24-hydroxy and Vitamin D 25-hydroxylase in endometriosis and gynecological cancers." *Reproductive Sciences*, vol. 14, pp. 486-497.

[28] Parikh, G., Varadinova, M., Suwandhi, P., Araki, T., Rosenwaks, Z., Poretsky, L., and Seto-young, 2010. "Vitamin D-regulates steriodogenesis and insulin-like growth factor binding proein-1(IGFBP-1) production in human ovarian cells." *Hormone and Metabolic Research*, vol. 42, pp. 754-757.

[29] David, M. L., Abdelouhid, T., and Stephen, R. P., 2012. "Association of hypogonadism with vitamin D status .The European male ageing study." *European Journal of Endocrinology*, vol. 166, pp. 77-85.

[30] Kassem, N. O., Wee, J. W., Modeste, N. N., and Johnston, P. K., 2003. "Understanding Soft drink consumption among female adolescents using theory of planned behavior." *Health Education Research*, vol. 18, pp. 278-291.

[31] Lew, K. and Barlow, P. J., 2005. "Dietary practices in Singapore and Malaysia." *Singapore Medical Journal*, vol. 46, p. 282.

[32] Tucker, K. L., Morita, K., Qiao, N., Hannan, M. T., Cupples, L. A., and Kiel, D. P., 2006. "Colas, but not other carbonated beverages, are associated with low bone mineral density in older women: The Framingham Osteoporosis Study." *Am. J. Clin. Nutr.*, vol. 84, pp. 936-942.

[33] Fernando, G. R., Martha, R. M., and Evangelina, R., 1999. "Consumption of soft drinks with phosphoric acid as a risk factor for the development of hypercalcaemia in postmenoupausal women." *J.Clin. Epidimol.*, vol. 52,

[34] Mazarigos-Ramos, E., Guerro-Romero, F., Rodriquez-Moran, M., Lazcano-Burciaga, G., Paniagia, R., and Amato, D., 1995. "Consumption of soft drinks with phosphoric acid as a risk factor for the development of hypercalcaemia in children: A case control study." *J.Peadiatr*, vol. 126, pp. 940-942.

[35] Heany, R. P. and Rafferty, K., 2001. "Carbonated beverages and Urinary Calcium excreation." *Am.J.Clin Nutri.*, vol. 74, pp. 343-347.

[36] Moltz, L., Leidenberger, F., and Weise, C., 1991. "Rational hormone diagnosis in Normocycitic functional sterility." *Journal of Fertility*, vol. 51, pp. 56-68.

[37] Olootu, W. E., Amballi, A. A., and A., B. T., 2012. "A review of female infertility ,Important etiological factors and management." *A Journal of Microbiology and Biotechnology Resources*, vol. 2, pp. 379-385.