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Comparative studies of proximate, mineral and phytochemical compositions of pomegranate (*Punica granatum*) in peel, seed and whole fruit powder

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Abstract
Pomegranate (*Punica granatum*) has been used for thousands of years to cure a wide range of diseases like diabetes, cancer, osteoarthritis, cardiovascular diseases and other myriad ailments across different cultures and civilizations. This is because pomegranates have the potential to thin the blood, increases blood flow to the heart, reduce blood pressure, plaque in the arteries, bad cholesterol parallel increasing good cholesterol. For experiments, proximate composition (moisture, ash, protein, fat, crude fiber, carbohydrate and vitamin C) were done by using Association of Analytical Communities (AOAC) standard methods. Mineral compositions (Na, Ca, Mg, K, P, Fe and Zn) were determined by atomic absorption spectrophotometry. UV spectrophotometer was used to determine phosphorous. Solvent free extract filtrate was used with various alkaloidal reagents to determine phytoconstituents. The result of the analysis shows that the peel, seed and whole fruit powder contains moisture content, ash, protein, crude fiber and fat. It also contains sodium, calcium, magnesium, potassium, phosphorus and iron. The three different extracts from peel, seed and whole fruit powder were found to contain alkaloids, flavonoids, phenolic compounds, glycosides, saponins, carbohydrates and protein. All of these findings implied that minerals and phytochemicals and vitamin C from pomegranate peels, seeds and whole fruit might be potential resources for the development of antioxidant function dietary foods and are useful in preventive healthcare.

Keywords: pomegranate, phytochemicals, minerals, dietary foods

1. Introduction
Plants are always a rich source of valuable compounds that do not hint for primary metabolic path but incorporates thousands of secondary metabolic activities [1]. The therapeutic efficacy of many indigenous plants for various diseases has been described by traditional herbal medicinal practitioners. Natural products are the source of synthetic and traditional herbal medicine [2]. Pomegranate (*Punica granatum L.*) is one of such plants with high potential to contain valuable chemical compounds. India is major pomegranate exporter in the world [3]. Pomegranate is a popular fruit in Sri Lankan home gardens, especially in dry and intermediate zones [4]. The ripe pomegranate fruit can be up to five inches wide with deep red, leathery skin. The arils are contained in the fruit and they are separated by white/yellow color membranous pericarp [5]. The arils contain white, pink to purplish or crimson pulp which is juicy, sweet and variable in acidity, but some are quit tart. The juice is edible but seed (without juice) and peel are considered as inedible portion. This inedible portion is found to be with higher medicinal value [6].
Pomegranates are used in ancient historical uses in several systems of medicine for a variety of ailments [7]. In Ayurvedic Medicine, Pomegranate is considered as "Blood Tonic" and to heal aphthae, diarrhea, ulcers, diabetes, dental condition, erectile dysfunction, and protection from Ultra-Violet (UV) radiation [5]. According to the historical background and the modern investigation, pomegranate has a lots of health benefits [8]. Pomegranate contain higher amount of nutrients compared to many familiar fruits. They contain no fats, low sugar and only 80 calories per 100g serving, 5g of fiber and 15% of the recommended daily allowances of vitamin C [9].
Pomegranate supplies Vitamin C as other fruits and Vitamin B_5, B_9, and K. It contains vital minerals potassium and Zinc. Seed oil contains phytoestrogens, similar to the estrogen naturally produce in human body [10]. Pomegranate contains anti-inflammatory activity to repair the body tissue damage and reduces the inflammatory state of gum. Generally the seed and peel particles are considered as waste portion of pomegranate. Inedible portion of pomegranate also contain antioxidant, phenolic compounds and flavonoids in high quantities. Further it can be a good nutrient source. However, this waste portion has not been utilized in food industry [11]. Recently, chemical constituents and their bioactivities in all parts of pomegranate (*Punica granatum L.*), including leaf, seed, juice, husk and peel, have been investigated [12]. Pomegranate seed, the by-product of pomegranate juice processing, contains a range of nutraceutical components such as sterols, γ-tocopherol, punicic acid and hydroxyl benzoic acids [13]. Pomegranate peel is a rich source of tannins, flavonoids and other phenolic compounds [14]. Pomegranate peel extract has both antioxidant and antimutagenic properties and may be exploited as biopreservative in food applications and neutraceuticals. However, so far, there has been no attempt to investigate the antioxidant properties of pomegranate in meat products [15].
The main objective of this study was to investigated the ability of using the pomegranate by-products in food processing and preservation technology which having high nutritional value, a

better acceptability for consumer and good safe hygienic quality. Therefore, the present study was performed to throw the light on the nutritional value indices; proximate, mineral phytochemical composition and vitamins content and of pomegranate fruit peel, seeds and whole fruit powders as by-products of their processing.

2. Materials and Methods

2.1 Plant materials

Sound fruits of pomegranate collected from local market of Banasthali, India were used in this study. Peels, seeds and whole fruit were separated. Those were dried in a tray drier for 24 hours at 55ºC - 60ºC. Dried peels, seeds and whole fruit were grinded and graded with 60 grade mesh and kept in air tight container until use.

2.2 Extraction

10g of dried peel and seed powder was taken separately ethanol; methanol and acetone were used as solvents. The samples were kept in vertical shaker at 40ºC for 4 hours. Extracts were filtered and concentrated using force convectional environment. Concentrated samples were kept in a freezer (at -4ºC to -7ºC) for further use.

2.3 Proximate Analysis

The crucible was sterilized, dried up and weighed. The crucible was measured again after taking sample into it. It was further weighed once again after oven drying at 110ºC. It was weighed repeatedly until determining the constant weight. Each time before the weight was taken; the crucible was cooled down in desiccators to get accurate results. Nutrient analysis included moisture, fat, ash, fiber, protein, carbohydrate and vitamin C estimations were done. Fat was estimated by the use of petroleum ether as extract solvent in Soxhlet apparatus. Total ash was calculated after weighing the furnace incinerated residue at 550ºC for 12hrs. The protein was estimated through micro Kjeldahl's distillation method [16]. Percentage carbohydrate was calculated by difference method.

2.4 Mineral Analysis

Chemical estimations were estimated for determining sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), potassium (K), phosphorus (P), iron (Fe) and zinc (Zn). The aqueous digest was examined using Atomic Absorption Spectroscopic standard method for Na, Ca, Mg, P, Fe and Zn. K was calculated using flame photometer.

2.5 Phytochemical screening

Preparation of Extract

2.5.1 Decoction

1g of dried fruit pulp powder was boiled in 16ml of double distilled water until the volume was decreased to 4 ml. The decoction was strained and utilized for further estimations.

2.5.2 Soxhlet extract

Crude plant extract was produced using soxhlet extraction method. About 50g of peel, seed and whole fruit powder was homogenously packaged into a thimble and extracted with 250ml of different solvents distinctly. Ethanol, methanol and acetone were used as solvents. The extraction process continued until the solvent in siphon tube of an extractor turned neutral. Followed by it, the extract was poured in a beaker and kept above the hot plate and heated at 30-40ºC until whole solvent was evaporated. Dried extract was preserved in a refrigerator at 4ºC for the further usage in phytochemical estimations.

The extract and decoction were examined to test whether the bioactive compounds (phenolic compounds, glycosides, flavonoids, saponins, alkaloids, proteins, amino acids and carbohydrates) were present through the usage of standard methods [17, 18].

Tannins: Crude extract was mixed with 2ml of neutral $FeCl_3$. A dark green coloration indicated the presence of tannins.

Phenols: Crude extract was mixed with few drops of 10% solution of lead acetate. White precipitate indicated the presence of phenols.

Flavonoids: Crude extract was mixed with 5ml of dilute ammonia followed by the addition of concentrated H_2SO_4. A yellow coloration observed in the extract indicated the presence of flavonoids. The yellow coloration disappears on standing.

Saponins: Crude extract was mixed with 5ml of distilled water in a test tube and shaken vigorously. The formation of stable foam was taken as an indication for the presence of saponins.

Alkaloids: Crude extract was mixed with 2ml of 1% HCl and heated gently. Mayer's and Wagner's reagent were added to the mixture. Appearance of cream color precipitates with Mayer's reagent and appearance of reddish brown precipitates with Wagner's reagent indicates the presence of alkaloids.

Steroids: Crude extract was mixed with 1ml of chloroform, few drops of acetic anhydride and two drops of concentrated H_2SO_4. The development of a greenish coloration indicated the presence of steroids.

3. Results and Discussion

3.1 Proximate composition

The proximate composition was determined on the pomegranate peels, seeds and whole fruit powder results were shown in Table 1 The result of the analysis shows that the peel was having high content of moisture, ash and vitamin C as compared to seed and whole fruit powder. The seeds contain high content of fat and protein as compared to peel and whole fruit powder. The whole fruit powder contains maximum content of crude fiber and carbohydrate as compared to peel and seed powder.

Table 1: Proximate composition of pomegranate peel, seed and whole fruit powder

S. No	Component Analyzed	Peel	Seed	Whole Fruit
1.	Moisture	69.7±0.35	20.8±0.45	17.6±0.62
2.	Ash	5.6±0.05	5.3±0.19	3.3± 0.69
3.	Fat	1.2±0.29	4.8±0.29	1.4±0.22
4.	Protein	7.8±0.16	9.2±0.35	6.4±0.37
5.	Crude Fiber	19.0±0.97	12.6±0.26	24.4±0.41
6.	Vitamin C	21.5±0.06	20.6±0.01	19.0±0.06
7.	Carbohydrate	64.84±0.53	64.85±0.07	78.58±0.23

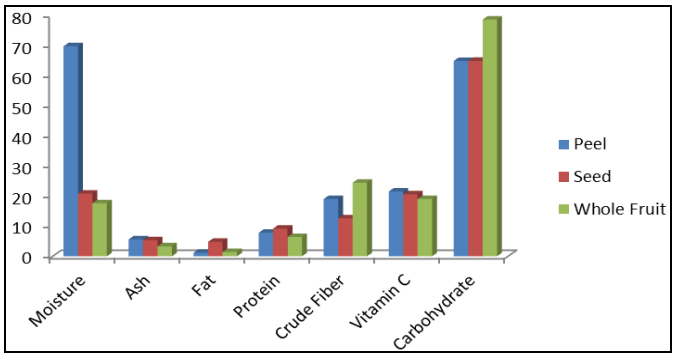

Fig 1: Proximate Composition of Pomegranate peel, seed and whole fruit powder

3.2 Mineral Composition

The minerals content of pomegranate peels, seeds and whole fruit powder were shown in Table 2. The concentration of phosphorus and zinc was found the highest in peel as compared to seed and whole fruit powder. The seeds found to have maximum content of magnesium and iron as compared to other. The content of sodium, calcium and potassium found to be higher in whole fruit powder as compared to peel and seed.

Table 2: Mineral Content of Pomegranate Fruit peel, seed and whole fruit powder

S. No	Component Analyzed	Peel	Seed	Whole Fruit
1.	Sodium	362.7±0.9	763.7±0.7	824.1±0.7
2.	Calcium	6679.5±0.9	7536±0.9	16237.41 ± 0.9
3.	Magnesium	728.23 ± 0.8	819±0.8	645.70 ± 0.8
4.	Potassium	524.80 ± 0.4	614±0.6	1644.47 ± 0.8
5.	Phosphorus	57.01 ± 0.7	48.05±0.3	33.96 ± 0.7
6.	Iron	18.33 ± 0.4	24.78±0.4	22.6 ± 0.7
7.	Zinc	16.2±0.06	5.70±0.1	11.0±0.10

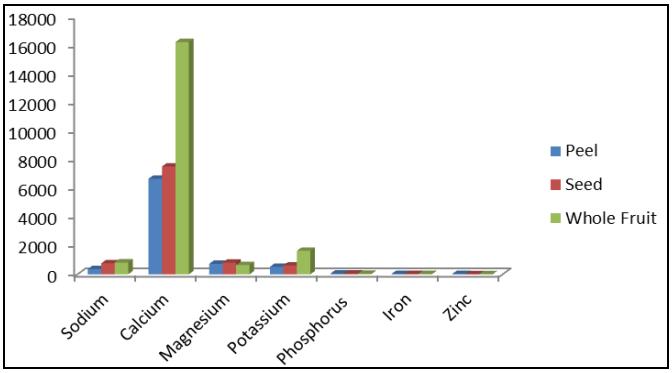

Fig 2: Mineral composition of pomegranate peel, seed and whole fruit powder

3.3 Phytochemical analysis

The curative properties of medicinal plants are perhaps due to the presence of various secondary metabolites such as alkaloids, flavonoids, glycosides, phenols, saponins and sterols. The various extracts of peel, seeds and whole fruit powder of *Punica granatum* have revealed the presence of alkaloids, glycosides, saponins, carbohydrates, proteins, phenolic compounds and flavonoids. Free amino acids were found to be absent in all the parts of pomegranate. From this analysis, ethanolic extract of seeds was found to have more constituents compared to peel and whole fruit extracts.

Table 3: Phytochemical analysis of pomegranate fruit peel, seed and whole fruit powder

S. No	Test used to detect phytochemicals	Peel	Seed	Whole fruit
1.	Alkaloids			
	Wagner's reagent	+	+	+
	Hager's reagent	+	+	+
	Mayer's reagent	+	+	+
2.	Saponins			
	Foam test	+	+	+
3.	Carbohydrates			
	Molish's test	+	+	+
	Barfoed's test	+	+	+
	Benedict's test	+	+	+
4.	Glycosides			
	Borntrager's test	+	+	+
	Legal's test	+	+	+
5.	Proteins			
	Biuret reagent	+	+	+
	Mllon's reagent	-	+	-
6.	Amino Acids			
	Ninhydrin test	-	-	-
7.	Phenolic Compounds			
	Ferric chloride test	+	+	+
	Gelatin test	+	+	+
	Lead acetate test	+	+	+
8.	Flavonoids			
	Alkaline reagent test	+	+	+

(+) Positive; (-) Negative

Sodium (Na) is very important physiological component and a primary extracellular cation in human beings. Sodium (Na) along with Chlorine (Cl) and Potassium (K) are electrolytes that regulate normal fluid balance in and out the cells and along with maintaining optimum acid-base balance in humans. Deficient potassium may result in muscle cramp and hypertension [19]. Potassium (K) is the most abundant mineral 1596 ppm content present in bael pulp powder. Human body requires a higher amount of potassium. Potassium is the prime electrolyte positioned in the body's cell and stored in muscle fibers with glycogen. It is an important part in helping in carrying out the transportation of glucose into the muscle cells. Deficient potassium levels results in nerve irritability, cardiac and mental disorder, muscular weakness and paralysis [19]. The Calcium (Ca) content was 94.9 ppm in bael fruit powder. It has numerous essential functions such as bone and dental tissues development, hormonal release, muscles contraction and glycogen metabolism [20]. The amount of Magnesium (Mg) was 243 ppm in bael fruit. Magnesium (Mg) helps in bones growth, maintaining a constant metabolic

process, keeping the flexibility of blood vessels, preventing cardiovascular diseases, and repairing injured cerebral cells [21]. Iron (Fe) is a vital trace constituent required in formation of haemoglobin, for regular working of the central nervous system and in carbohydrates, proteins and fats oxidation [22]. Either in a direct or in an indirect way, the iron status can shows an effect on the capacity of a cell to produce adenosine triphosphate (ATP) and on the glucose and other carbohydrates oxidation process [23]. [24] have determined that deficient iron levels result in deterioration of cognitive functioning, mainly among school going children [25]. Verbal performance may directly get affected by deficient iron status.
The preliminary phytochemical screening tests may be useful in the detection of the bioactive principles and subsequently may lead to the drug discovery and development. Further, these tests facilitate their quantitative estimation and qualitative separation of pharmacologically active chemical compounds [26]. The phytochemical screening in the present study has revealed the presence of alkaloids, glycosides, saponins, carbohydrates, proteins, phenolic compounds and flavonoids in the peel, seed and whole fruit extract. Further the presence of different phytoconstituents in the three different extracts may be responsible for the therapeutic properties of pomegranate. Flavonoids and phenolic compounds are a major group of compounds that act as primary antioxidants or free radical scavengers. Since these compounds were found to be present in the extracts, it might be responsible for the potent antioxidant capacity of pomegranate. The secondary metabolites (phytochemicals) and other chemical constituents of medicinal plants account for their medicinal value. For example, saponins have hypotensive and cardio depressant properties [27]. Glycosides are naturally cardioactive drugs used in the treatment of congestive heart failure and cardiac arrhythmia [28]. The presence of saponins in whole fruit and seeds extract and glycosides in all the extracts might play a role in the cardioprotective potential of pomegranate. Since the seed extract contains the components of both peel and whole fruit, it was found to contain more constituents. It would thus mean that in this study, the seeds extract had the highest number of bioactive compounds. Since the yield of bioactive metabolites in a plant extract also varies considerably with the [29, 30] method/solvent of extraction it is plausible that the ethanolic extracts were generally more potent than the aqueous extracts probably because the active principles in the plant dissolved more readily in and were better extracted by a less polar solvent (ethanol) than water. This is in agreement with many literatures reporting of differences in the activities of extracts obtained from the same morphological part of a plant using different solvents. For instance, the methanolic extract of the fruits of *Tetrapleura tetraptera* is more potent than the aqueous [31] extract. The preliminary phytochemical tests are helpful in finding chemical constituents in the plant material that may lead to their quantitative estimation and also in locating the source of pharmacologically active chemical compound.

4. Conclusion

A present result indicates that the fruit pulp powder of Pomegranate offers a good pool of nutrients. Fruit pulp, seeds and whole fruit were wealthy source of carbohydrates and vitamin C. In the light of these explored nutritional facts, it can be concluded that the studied part of pomegranate would exercise as a new source of superior quality food. The presence of phytoconstituents make the plant useful for treating different ailments and have a potential of providing useful drugs of human use. In the present study, we have found that most of the biologically active phytochemicals were present in the ethanolic, methanolic and acetone extracts of *Punica granatum* peel, whole fruit and seeds. Since the ethanolic extract of seeds contains more constituents it can be considered beneficial for further investigation.

5. Conflict of Interests

The authors declare that there is no conflict of interest regarding the publication of this paper.

6. Acknowledgement

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7. References

1. Elfalleh W, Hedia H, Nizar T, Yassine Y, Nizar N, Ali F, *et al*. Total Phenolic Contents and Antioxidant Activities of Pomegranate Peel, Seed, Leaf and Flower. Journal of Medicinal Plants Research. 2012; 6:4724-4730.
2. Gami B, Parabia MH. Pharmacognostic Evaluation of Bark and Seeds of Mimusops elengi. International Journal of Pharmacy and Pharmaceutical Sciences. 2010; 2(4):110-113.
3. Mertens T, Susanne U, Petra JS, Jolian R, Lal H, Hartmut D, *et al*. Absorption, Metabolism, and Antioxidant Effects of Pomegranate (*Punica granatum* L.) Polyphenols after Ingestion of a Standardized Extract in Healthy Human Volunteers. Journal of agricultural and food chemistry. 2006; 54(23):8956-8961.
4. Amararatne DIM, Weerakkody WAP, Jayakody JALP. Bioactive Properties of Fruit Juice of Pomegranate (*Punica granatum*) Grown in Dry Regions of Sri Lanka. Tropical Agricultural Research. 2012; 23(4):370-375.
5. Jurenka J. Therapeutic Applications of Pomegranate (*Punica granatum* L.): A Review. Alternative medicine review. 2008; 13(2):128-144.
6. Ahmed MM, Safaa EA. Protective Effect of Pomegranate Peel Ethanol Extract against Ferric Nitrilotriacetate Induced Renal Oxidative Damage in Rats. Journal of Cell and Molecular Biology. 2010; 7(2):35-43.
7. Christaki EV, Eleftherios M, Panagiota CFP. Dietary Benefits of Pomegranates in Humans and Animals. Journal of Food, Agriculture and Environment. 2011; 9(1):142-144.
8. Jagtap UB, Shrimant NP, Bapat VA. Evaluation of Antioxidant Capacity and Phenol Content in Jackfruit (*Artocarpus Heterophyllus* Lam.) Fruit Pulp. Plant foods for human nutrition. 2010; 65(2):99-104.
9. Hassan NA, Abeer AEH, Sayed HA. Phytochemicals, Antioxidant and Chemical Properties of 32 Pomegranate Accessions Growing in Egypt. World Applied Sciences Journal. 2012; 16(8):1065-1073.

10. Abbasi H, Karamatollah R, Ladan R. Extraction of Essential Oils from the Seeds of Pomegranate Using Organic Solvents and Supercritical Co2. Journal of the American Oil Chemists' Society. 2008; 85(1):83-89.
11. Parashar A, Gupta SK, Ashok K. Studies on Separation Techniques of Pomegranate Seeds and Their Effect on Quality of Anardana. African Journal of Biochemical Research. 2009; 3(10):340-343.
12. Lansky EP, Newman RA. *Punica granatum* (pomegranate) and its potential for prevention and treatment of inflammation and cancer. Journal of Ethnopharmacology. 2007; 109:177-206.
13. Liu G, Xu X, Hao Q, Gao Y. Supercritical CO2 extraction optimization of pomegranate (*Punica granatum* L.) seed oil using response surface methodology. LWT-Food Science and Technology. 2009; 42:1491-1495.
14. Li Y, Guo J, Yang J, Wei J, Cheng S. Evaluation of antioxidant properties of pomegranate peel extract in comparison with pomegranate pulp extract. Food Chemistry. 2006; 96:254-260.
15. Naveena BM, Sen AR, Vaithiyanathan S, Muthukumar M, Babji Y. Microwave cooking properties of chicken patties containing honey and vitamin-C. Journal of Food Science and Technology. 2007; 44:505-508.
16. Imamul Huq SM, Alam MD. A Hand Book on Analyses of Soil, Plant and Water, BACER-DU. University of Dhaka, Bangladesh, 2005, 55-59.
17. Trease GE, Evans MD. A text book of Pharmacognosy. 13th ed. Baillier, Tindal and Caussel, London, 1989, 144-148.
18. Harborne JB. Phytochemicals Methods. Chapman and Hall Ltd. London, 1973, 49-188.
19. Adotey DK, Serfor-Armah Y, Fianko JR, Yeboah PO. Essential elements content in core vegetables grown and consumed in Ghana by instrumental neuron activation analysis. African Journal of Food Science. 2009; 3(9):243-249.
20. Kaneko JJ, Harvey JW, Bruss ML. Clinical Biochemistry of Domestic Animals. 5th ed. Academy Press London, 1977, 619-702.
21. Sasaki S, Oshima T, Teragawa H, Matsuura H, Kajiyama G, Kambe M, *et al*. Magnesium status in patients with cardiovascular diseases. Rinsho Buori. 1999; 47:396-401.
22. Adeyeye EI, Otokiti MKO. Proximate composition and some nutritionally valuable minerals of two varieties of Capsicum annum. (Bell and Cherry peppers). Discovery and Innovation. 1999; 11:75-81.
23. Griffith HW. Complete guide to vitamins, minerals and supplements. 1st ed. Fisher Books Company press: New York. 1995; 92(11):4733-4737.
24. Bryan J, Osendarp S, Hughes D, Calvaresi E, Baghurst K, Klinken JW, *et al*. Nutrients for cognitive development in school-aged children. Nutrition Revision. 2004; 62(8):295-306.
25. Sungthong R, Mo-Suwan L, Chongsuvivatwong V. Effects of haemoglobin and serum ferritin on cognitive function in school children. Asia Pacific Journal of Clinical Nutrition. 2002; 11:117-122.
26. Varadarajan P, Rathinaswamy G, Asirvatahm D. Antimicrobial properties and phytochemical constituents of Rheo discolor. Ethnobotanical Leaflet. 2008; 12:841-845.
27. Olaleye MT. Cytotoxicity and antibacterial activity of methanolic extract of *Hibiscus sabdariffa*. Journal of Medicinal Plants Research. 2007; 1:9-13.
28. Brian FH, Thomas-Bigger J, Goodman G. The Pharmacological Basis of Therapeutics. Macmillan, New York: NY, USA, 1985, 7.
29. Clark TE, Appleton CC, Drewes SE. A semi-quantitative approach to the selection of appropriate candidate plant molluscicides-a South African application. Journal of Ethnopharmacology. 1997; 56:1-13.
30. Marston A, Maillard M, Hostettmann K. Search for antifungal, molluscicidal and larvicidal compounds from African medicinal plants. Journal of Ethnopharmacology. 1993; 38:215-223.
31. Adewunmi CO, Adesina SK, Marquis VO. On the laboratory and field trials of *Tetrapleura tetraptera*. Bulletin of Animal Health and Production in Africa. 1982; 30:89-94.