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Macro and Micro Nutrient Composition of Groundnut (*Arachis hypogaea*) Leaf and Calcium Oxalate Shape and Arrangement by using SEM-EDX

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ABSTRACT

Groundnut (Arachis hypogaea) is an important oil crops. The fodder of groundnut use as animal foods. Groundnut leaf contain large amount of calcium (Ca) and the particles of calcium have unique shape and arrangement. Leaf is the rich source of minerals like magnesium, potassium, calcium and chlorine. The different varieties of leaf were analysed by using SEM-EDX, MP-AES and biochemical analysis showed the different mineral concentration. One thing new in the analysis found that the calcium particles have unique shape and size in all the varieties and this is the first time report the calcium particles shape and arrangement on groundnut leaf.

Key words: *Minerals, SEM-EDX, Calcium*

INTRODUCTION

India is the second largest producer of groundnuts after China. Groundnut is the largest oilseed in India in terms of production. It accounted for 35.99 per cent of the oilseeds production of the country during 2007-08. Gujarat is the largest producer contributing 25 per cent of the total production followed by Tamil Nadu (22.48 per cent), Andhra Pradesh (18.81 per cent), Karnataka (12.64 per cent) and Maharashtra (10.09 per cent) during 2006-07. Groundnut contains on an average 40.10 per cent of fat and 25.30 per cent of protein and is a rich source of calcium, iron and vitamin 'B' complex like thiamine, riboflavin, niacin and vitamin 'A'. It has multifarious usages. It is used not only as a major cooking medium for various food items but also for manufacture of soaps, cosmetics, shaving creams and lubricants. In fact, it plays a pivotal role in the oilseed economy of India. India exports groundnut kernels, shell, hand picked selected (HPS) groundnut and oil cake forms. Groundnut haulms and leaves serve as a rich source of cattle feed and raw material for preparation of silage. Being a leguminous crop, groundnut is also grown in crop rotation as it synthesizes atmospheric nitrogen and adds 100-120 kg of nitrogen in the field per hectare per season. It maintains the fertility of soil and helps in reducing soil erosion.

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The cake has several uses in feed and infant food formulations. Groundnut provides considerable amounts of mineral elements to supplement the dietary requirements of humans and farm animals. Groundnut seeds are reported to contain 9.5 to 19.0% total carbohydrates as both soluble and insoluble carbohydrate[1,3,5,7,9]. The stovers from cereals have low nitrogen and high fibre contents and poor digestibility, and therefore have low nutritive value, while the stover from legumes are of high nutritional quality[8]. Especially, leaves of legume crops can provide good quality forage material in farms. Crop residues of groundnut (*Arachis hypogaea* L.) provide important feed resources for livestock production[2,6]. The by-products of peanut contain many functional compounds, such as protein, fiber and polyphenolics, which can be incorporated into processed foods to serve as functional ingredients.

MATERIAL AND METHODS

The samples were used in this study leaves of GJG-20, GJG-22, GJG-17, GG-21 and GG-5 Groundnut varieties. Samples were collected from the biotech plot in the summer season in sterile bags. From each plant 10 leaves were collected.

Scanning Electron Microscopy (SEM) Analysis

Sample preparation for SEM and analysis

The SEM analysis two leaves samples were used. Small part of leaf cut with the help of cutter and fix on the sample holder (stub) of SEM. Samples were dried in incubator at 40^0C for 1 hrs. Then, gold coating using sputter coater. After the coating images were taken at different magnification using SEM (EVO18). Parallel the elemental detection was carried out on leaf at different spot by using Elemental detector (EDX) which is inbuilt in SEM. Elemental analysis was carried out using TEAM software.

Chemical Analysis

Sample preparation and analysis

Remaining eight leaf samples were oven dried and grind in mixture to prepare fine powder and for analysis purpose. sample digestion[4] 0.5gm fine powder of sample in digestion tube then add 7ml HNO_3 Let it pre-digest for 4-6 hour, Digest the sample by Mars-6 Microwave Digestion system. Transfer the solution to 50ml volumetric flask and make up the solution 50ml by mille water and Store this solution to reagent bottle for further analysis.

Trace Elements Analysis by MP-AES

MP AES instrument was standardized by NIST certified standard .Sample were run on triplicate and its average value is utilize in table. Following table represents the methods and instruments were used for measure the different parameters

Table 1: Parameters and methods used for chemical analysis

Parameters	Method
Digestion	Nitric acid digestion
Trace elements	MP AES (Microwave Plasma Atomic Emission Spectroscopy)
Total N	Micro Kjeldahl method
Total P	Venado molybdo phosphoric yellow color method
Total K	Flame photometer
Total S	Terbidity method
Total Na	Flame photometer
Total Protein	Micro Kjeldahl method

RESULT AND DISCUSSION

Result of SEM analysis

Groundnut leaves SEM images were taken at 15kv EHT in high vacuum mode 500 to 2000 magnification observed the stomata and leaf architecture. During the images acquisition unique particle and its arrangement was found on the leaf. After EDX analysis of that

particles was identified as calcium oxalate. For confirmation of the particle that it was calcium or not. Analysis of leaf samples were carried out in Microwave Plasma Atomic Emission Spectroscopy (MP- AES). compare the result of the EDX and MP-AES, it was found that the Calcium concentration higher than the other trace elements present in the leaf. So, it was confirmed that this was calcium particles. Fig.1 Show the SEM images of different groundnut varities with particle shape and arrangement on the leaf

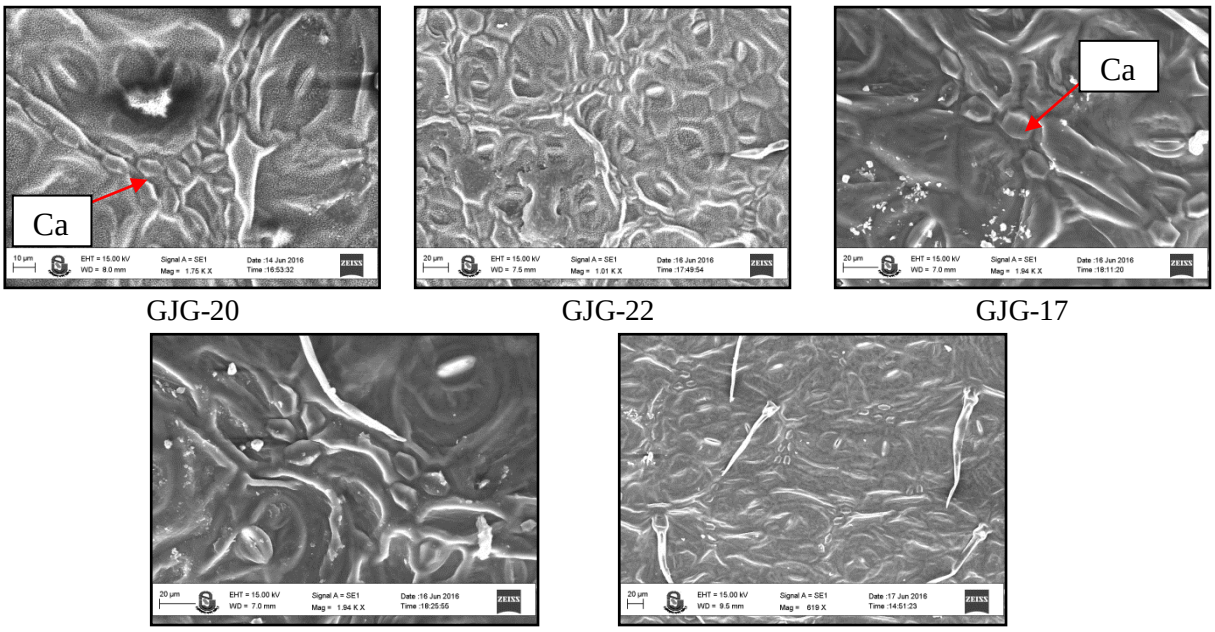
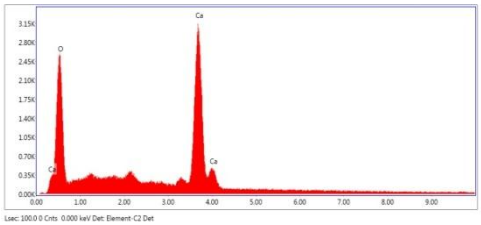


Fig. 1: SEM images of Groundnut varieties

SEM-EDX analysis was showed the different element present on the leaf. The fig.2 A, C, E, G and I shown the elemental composition of the specific particle as discussed above present on the leaf. based on that it was identified that the particles were calcium oxalate. Fig.2 B, D, F, H and J showed the elemental composition of other spot on the leaf means other than the calcium particles and it was shown that the other element were present on the leaf.

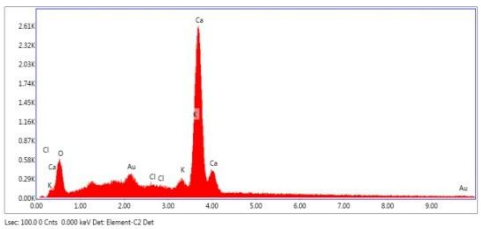
A



GJG-20

Element	Weight %	Atomic %	Net Int.	K ratio	Z	R	A	F
O K	57.24	77.03	393.52	0.1130	1.0661	0.9609	0.1852	1.0000
CaK	42.76	22.97	568.45	0.3927	0.9032	1.0406	1.0057	1.0108

B



Element	Weight %	Atomic %	Net Int.	Kratio	Z	R	A	F
O K	29.83	52.14	73.31	0.039	1.121	0.930	0.117	1.000
AuM	2.16	0.31	19.88	0.031	0.639	1.341	1.324	1.709
ClK	0.50	0.39	5.81	0.004	0.944	1.003	0.944	1.031
K K	2.82	2.02	26.93	0.027	0.938	1.014	0.982	1.073
CaK	64.69	45.14	474.5	0.613	0.955	1.019	0.985	1.007

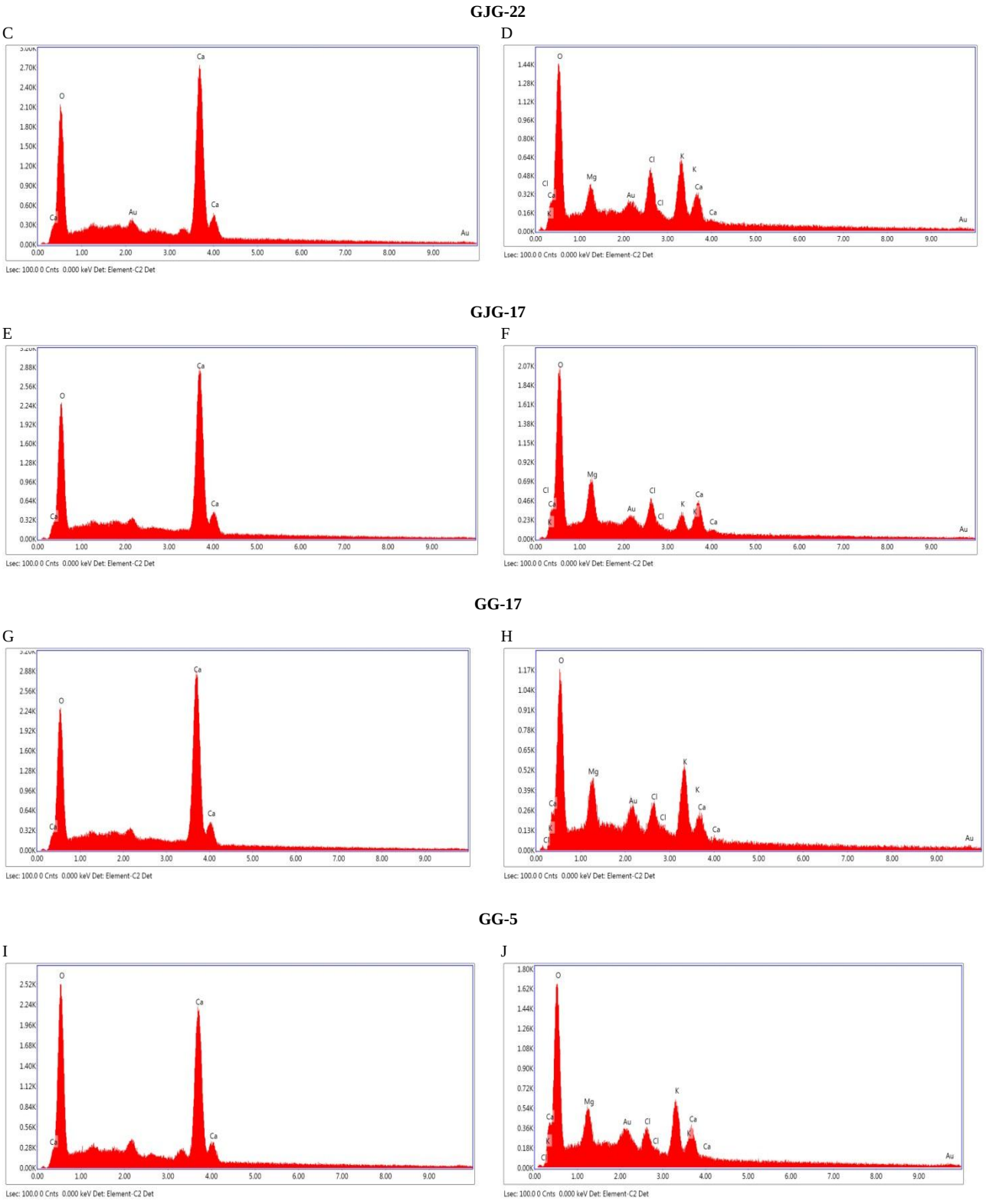


Fig. 2: SEM-EDX analysis of different groundnut varieties

RESULT OF CHEMICAL ANALYSIS

Chemical analysis of leave samples were performed by different methods. it was found that high amount of nitrogen follow by potassium and calcium. other micro element like Fe, Cu, Zn etc as shown in table 2. found in samples.

Table 2: MP-AES and Chemical analysis of groundnut leaf samples

Sample Name	Unit	GJG-20	GJG-22	GJG-17	GG-21	GG-5	Average
N	%	3.29	3.81	3.79	3.59	3.16	3.53
P	%	0.07	0.06	0.05	0.05	0.06	0.06
K	%	2.19	2.27	2.06	2.04	1.68	2.05
S	%	0.04	0.05	0.05	0.05	0.06	0.05
Na	%	0.03	0.04	0.02	0.03	0.04	0.03
Ca	%	1.57	1.52	1.89	2.02	1.51	1.70
Mg	%	0.49	0.58	0.64	0.57	0.54	0.57
Cu	ppm	11.29	11.32	10.46	9.80	11.15	10.80
Fe	ppm	142.93	535.16	126.15	138.88	378.39	264.30
Mn	ppm	71.73	65.22	104.27	81.08	97.54	83.96
Zn	ppm	17.83	17.60	19.38	18.09	15.93	17.76
Ti	ppm	9.48	11.79	10.73	10.23	11.05	10.65
V	ppm	44.48	65.39	29.45	35.80	59.59	46.94
Cr	ppm	1.10	2.12	1.31	2.95	3.23	2.14
Mn	ppm	73.51	65.61	78.78	73.48	102.40	78.75
Sr	ppm	296.80	229.10	285.40	294.52	277.04	276.57
Co	ppm	10.85	28.83	18.51	20.37	28.62	21.43

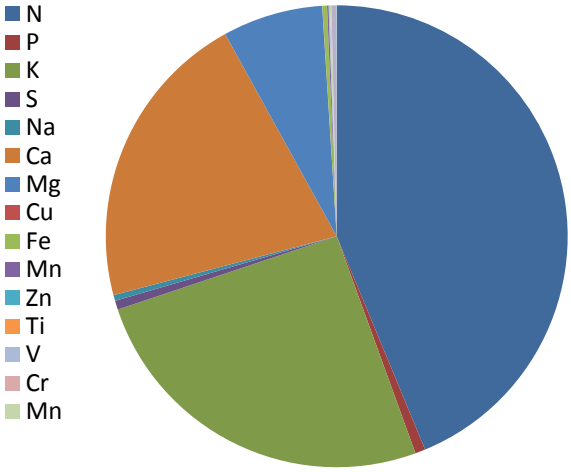


Fig. 3: Pie-chart of Elemental composition

CONCLUSION

The Scanning Electron Microscopy-EDX, MP-AES and chemical analysis of groundnut's leaves, it was found that the leaves of groundnut contain the high amount of nitrogen than potassium and calcium. other than these trace elements copper, iron, zinc etc. were present in leaf. Specific particles present on the groundnut leaf was identified as calcium by the comparison of EDX and MP-AES analysis. Groundnut leaves were good source of trace elements which was used as animal fodder for the full fill the requirement on micro-nutrients.

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