

GU J. Phytosci.



## GU JOURNAL OF PHYTOSCIENCES

### A Quarterly Peer-Reviewed Journal

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### **Department of Botany**

Ghazi University, City Campus Dera Ghazi Khan- 32200 Punjab, Pakistan https://gudgk.edu.pk/phy-to-science-journal

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### **Table of Content**

S. No.	Title of Manuscript and Authors	Page No.
1	Diversity of Wild Vegetation Along the Aridity Gradient Habitat of Dera	
	Ghazi Khan	74-88
2	Qurat-ul-Ain, Allah Bakhsh Gulshan and Faisal Hussain Evaluation of Selected Landraces for Yield and Yield Attributes from	
2	District Bannu Against Drought Stress	
	Tahir Muhammad, Muhammad Abid Khan, Saadullah Khan Laghari, Allah Bakhsh	89-96
	Gulshan, Faisal Hussain, Anila Nijabat, Saher Nawaz, Abdul Basit and Naseem	
	Ullah	
3	First Record of <i>Pisolithus tinctorius</i> from Sangla Hill, Nankana Sahib,	
	<b>Punjab, Pakistan</b> Ayesha Noor Awan, Nousheen Yousaf, Saher Nawaz, Atia Nazir, Aneela Nijabat,	97-102
	Zainab Naseem, Saadullah Khan Leghari, Allah Bakhsh Gulshan, Faisal Hussain	97-102
	Muhammad Abid Khan, and Muhammad Awais Arif	
4	<b>Concentration of Proximate and Heavy Metals in Four Selected Vegetables</b>	
	Irrigated by Municipal Wastewater	103-109
	Saadullah Khan Leghari, Mudassir Asrar Zaidi, Sana-Ur- Rehman Sheikh, Azhar	100 107
5	Ali Merri and Muhammad Sadir Effect of Drought and Waterlogging on Cotton crop in Dera Ghazi Khan	
Э	Muhib Hussain, Allah Bakhsh Gulshan and Faisal Hussain	110-118
6	Effect of Coal Fly Ash on Seedling Growth, Productivity, Biochemical and	
	Heavy Metals Uptake of Zea mays L. and Soil Characteristics in Semi-arid	
	and Climatic Conditions of Quetta Valley	119-128
	Rafiullah, Saadullah Khan Leghari, Ateeq Ullah Kakar, Allah Bakhsh Gulshan,	
7	Faisal Hussain, Sayed Qaim Shah, Abdullah Baloch and Muhammad Iqbal Jakrro Analytical Study of the Historical Contribution of Muslims in Horticulture	
/	During 800-1250 AD	129-133
	Sohail Akhtar	129 155
8	A Systematic Review on Synthesis of Nanoparticles and Their Applications	
	Abdul Basit, Allah Bakhsh Gulshan, Faisal Hussain, Afshan Fareed, Asia Kainat	134-144
-	and Muhammad Waqas	
9	Nanobiotechnology: As Emerging Field in Agriculture	145-150
	Usama Ahmad Khan	

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Hussain, F., S. S. Shaukat, M. Abid, F. Usman and M. Akbar. 2014. The effect of fungicides alone and in conjugation with chitin on the control of some fungal pathogens associated with chilli seeds. *World Appl. Sci.* J., **32(2)**: 977-985.

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### Diversity of Wild Vegetation Along the Aridity Gradient Habitat of Dera Ghazi Khan

#### Qurat-ul-Ain, Allah Bakhsh Gulshan\* and Faisal Hussain

Department of Botany, Ghazi University, Dera Ghazi Khan, Pakistan

#### Abstract

The investigation was carried out of wild vegetation along aridity gradient from Sakhi Sarwar to Ghazi Ghat in Dera Ghazi Khan Landscape. Presence/absence data were collected from 13 sampling sites. We used classification as well as ordination methods to analyze the data structure. The main emphasis was on classification, ordination being used in part to check whether the classification results reflect in an adequate way the main floristically gradients in the data set, and also to detect relations between some environmental factors and the composition and structure of natural vegetation. The major axes brought by the ordination were related to soil characteristics. The application of multivariate statistic {detrended correspondence analysis (DCA) and cluster analysis} allowed an interpretation of weed species spatial distribution and assemblage. From the results of classification (cluster analysis) samples from fields could be divided into distinct plant associations based on floristic composition. We have shown that there is a clear relation between physical and chemical features of the substrate and the association defined by numerical analysis. In this respect, soil texture and organic matter seems particularly important in shaping the natural communities.

Keywords: Okra; Sowing time; Growth; Yield; Climatic connnnditions

#### 1. Introduction:

The ecological variations within inclined landscapes may show different spatial configurations. Landscapes having a spatially continuous variations (or spatial dependence) can be considered homogeneous. In contrasts, landscapes showing a discontinuous variation in space (or spatial independence) can be considered heterogeneous (Kolasa & Rollo, 1991). The potential role of landscape heterogeneity over plant species diversity and distribution has been suggested by different authors using conceptual (Shmida & Wilson, 1985; Auerbach & Shmida, 1987) and mathematical (Palmer, 1992) models.

The relationship between species and functional diversity is thus central in identifying mechanisms of biodiversity effects (Diaz & Cabido 2001; Hooper *et al.* 2005; Mason *et al.* 2005). The concept of functional diversity (FD) remains however rather complex because there are questions about how to define, measure and assess FD

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variability (Petchey & Gaston, 2006). One approach that has gained currency considers the FD as the extent of functional trait variation among the species in a community (Tilman, 2001, Petchey & Gaston, 2002).

Species richness is expected to be higher in the heterogeneous landscapes; because of the closer distance between dissimilar habitats, species may be present unfavorable habitats supported by nearby populations in favorable habitats. It has been proposed that this phenomenon, named mass effect (Shmida & Wilson, 1985), may be determination of the species per site. In arid ecosystems structural and functional variation is primarily controlled by water availability (Meir, 1973). In flats landscapes spatial changes in water availability depend mainly on precipitation. Precipitation varies continuously in space generating homogenous gradient of water availability in flat areas. In hilly or mountain landscapes water availability is also influenced by slope, aspect and elevation. At the landscape scale these topographic variable shows a discontinuous variation. Consequently, water availability and plant species distribution in arid and semi-arid ecosystems (Whittaker & Niering, 1975; Yeaton & Cody, 1979; Burke *et al.*, 1989; Dargie & El-Demerdash, 1991). The increase in plant species richness with the precipitation within the range of arid and semi-arid conditions has been documented (Aronson & Shmida, 1992).

The effects of changes in the spatial configuration of water availability gradient on species composition or diversity remain poorly explored. In this study we describe the floristic and diversity patterns of inclined moisture, analyze their relationship with water availability gradients, and explore effects of the spatial configuration of these gradients in landscape of contrasting topography. Biodiversity has recently emerged as an issue of both scientific and political concern primarily because of an increase in extinction rates caused by human activities (Ehrlich & Wilson, 1991). Several very large experiments (Tilman & Downing, 1994; Naeem et al., 1994; 1995; Kareiva, 1994; 1996; Tilman, 1996) have addressed the relationship between biodiversity, measured as species richness, and ecosystem function. However, they have failed to reveal a clear causal effect (Huston, 1994). After a revision of some of the problems and hidden treatments in these experiments, Huston, (1997) concluded that they do not provide evidence that increasing biodiversity improve ecosystem function and that "both local species diversity and the rate ecosystem processes such as productivity are determined by the amount and variability of the fundamental environmental resources that regulate the plant growth and ecosystem processes". Species richness patterns in relation to environment need to understand before drawing conclusions on the effect of biodiversity in ecosystem processes. Numerous problems regarding the study of species richness need to clarify, including the role of disturbance (Grime, 1979; Huston, 1994), and the relative importance of biotic versus abiotic factors (Grime, 1979; Cornell & Lawton, 1992; Austin & Gaywood, 1994).

In the present investigation we want to explore the distribution and community structure of wild vegetation along the aridity gradient of Dera Ghazi Khan Range. From this study, the followings objectives are considered to seek during the survey.

#### 1.1. Objectives:

- To assess the wild vegetation along the aridity inclined from upland area to lowland land of Dera Ghazi Khan from west-east landscape.
- To examine the relationship of vegetation and the substrate
- To measure the composition and community structure of vegetation from the study area.

#### 2. Materials and Methods:

#### 2.1. Study Area:

Quetta The study was conducted from west to east up to the area of 50 km along the aridity gradient from Suleiman Mountain range Sakhi Sarwar upland area (29° 58′ 45″ N, 70° 18′ 21″ E) to Ghazi Ghat lowland area of Indus River (30° 6′ 2″ N, 70° 52′ 22″ E) in Dera Ghazi Khan district, Punjab province, Pakistan. The zone has extensive alluvial plains and protracted over the larger portion of the Punjab province. These were bordered by foothills of Suleiman Range on the west. The soil of the area is characterized by sandy to loamy. The height diverges from 284 m from to 114 m from sea level and overall climate of the area is dry with little rainfall. The winter season is comparatively cold while the summer is very hot, and the temperature is usually rising about 115 °F (46 °C). Due to the desolate Suleiman Mountains and the sandy soil of the sites, winds are very common during the summer.

#### 2.2. Collection of Vegetation Data:

Survey of wild vegetation from piedmont to alluvial landscape was conducted during 2014. A total of 13 stands were selected along the west- east transect approximately 50km long. The sampling of wild vegetation was selected in three different regions (Figure 1): upper piedmont, lower piedmont, and alluvial plains. These sites were selected to encompass a wide range of topo-edaphic conditions corresponding to different landscapes.

At each 5 km, 2 sampling units were selected. The sampling fields> 50×50 m was surveyed. The vegetation was sampled determined the individual species frequency and over all plant richness. In each field, the composition of vegetation was recorded ten 1.0m×1.0m quadrates. All the plant species within each quadrate were recorded and the voucher specimens were taken to the laboratory. Nomenclature of vegetation follows by the flora of Pakistan. To determine the relationship between spatial distribution and Soil properties were measured soil physical and chemical analysis. Soil was sampled from various depths; 0-10 cm, the surface layer of soil, from centre to each sampling site. Aggregated samples were sieved through a 4-mm screen to remove stones and large pieces of organic material. Following sieving, a portion of each aggregated sample was processed for soil physical and chemical properties.

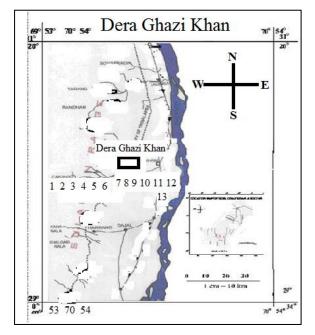


Figure 1. Shows the locations of study stands along the aridity gradient habitat of Dera Ghazi Khan

#### 2.3. Soil Reactions (pH):

The pH values show that soils of Dera Ghazi Khan District are slightly alkaline, with no significant difference between one site and another as the zone and site main factors were not significant in overall analysis of variance. The pH is a negative logarithm of Hydrogen ion concentration. The soil of Dera Ghazi Khan was neutral to slightly alkaline from upland area to low land area. The differential differences among the sites were responsible for the occurrence of significant interaction between sites and zones in the overall analysis.

#### 2.4. Electrical Conductivity (μs/cm):

The EC values show that soils of Dera Ghazi Khan district are non-saline, with no significant difference between values of different sites are considered, it can be seen that EC ranges is high in association B, while those of association A is very low ranges. The amount of solute concentration relatively declines from piedmont to alluvial soil, thus the Electrical Conductivity decline from association A to Association D. In fact, the soil EC appears to be site specific in different zones as the interaction between zones and site was significant in the overall analysis of variance.

#### 2.5. Soil Organic Matter Content:

There are significant differences in the organic matter between different zones of Dera Ghazi Khan District. The results show that the soils of Dera Ghazi Khan are poorer in organic matter than those of either Dera Ghazi Khan, which do not differ between themselves. Organic matter differs from one site to another, as the sites the main factor was significant in ANOVA. Natural vegetation soils tend to increase the organic matter from association A to association D and maximum in association B along the Quetta Multan Road from Sakhi Sarwer to Ghazi Ghat.

#### 2.6. Soil Texture (%):

Data for soil texture analysis reveals that mostly all soil of the Dera Ghazi Khan district are sandy clay loam, with no appreciable difference between different sites as neither the main factor nor their interaction were significant in the overall analysis of variance, but some stands also contain clay (sample 10 and 11) This shows the properties of silt, sand and clay in different zones and sites.

#### 2.7. Data Analysis:

In this study, we used classification as well as ordination methods to analysis the data structure. The main emphasis was on classification ordination being used in part to check whether the classification results in an adequate way the main floristically gradient in the data set, and also to detect relation between some environmental factors and the composition and structure of weed vegetation. Species and sites data were arranged into a matrix. The matrix data were used to analyze the species diversity, cluster analysis and ordination by using the statistical computing program MVSP (Version 3.2).

#### 2.8. Classification:

We used information statistical methods of data classification. This technique is based on method of the diverse species and was used as a quick and efficient way to obtain an initial clustering of samples. Information Statistical method is a hierarchical rather than chronological clustering method. Information Statistical methods chronological clustering method has a special constraint, which means that only adjacent sampling subunits can be grouped. The clustering analysis was performed with using the default option "farthest neighbor" with incorporating Pearson coefficient. Presence- absence data was used for this analysis.

#### 2.9. Ordination:

Indirect gradient analysis was performed separately for sample fields using the programmed detrended correspondence Analysis (DCA) (ter Break, 1987). Rare species (those with relative frequency lesser than 1%were omitted from the analysis and default option were used. Eigenvalues for the first two axes of the ordination (measure of their importance) Eigenvalues> 0.5 were obtained for axes 1; axes2, with Eigenvalues < 0.4, was less important. As DCA axis 1 and 2 explained most of the variation in the date sets, only these axes were considered for further analysis. In both date sets samples and species were regularly spread along the axes of the ordination to assess the compatibility of two method of data simplification (Dargie & El-Demerdash, 1991). The relationship between sample/species scores and environmental variables were determined using Pearson correlation. DCA was calculated with computer software program MVSP version 3.2.

#### 2.10. Analysis of Variance and Correlation:

Differences are soil parameters between the plants associations were estimated by using the analysis of variance (ANOVA). Appropriate graphs were drawn to illustrate the difference between the plant associations delineated by the cluster analysis. The relationship between soil character and DCA axis 1 and 2 were determined using Pearson correlation. The calculations were made with the help of MINITAB present version 15.0-statistical computer software.

#### 3. Results:

#### 3.1. Classification of Wild Vegetation by Normal Cluster Analysis:

Four vegetation groups (associations) were recognized by the normal cluster analysis (Table 1). These groups were delineated at two levels of division of hierarchical diagram (Figures 2). The most obvious attribute signified by the cluster analysis was the separation of the entire wild vegetation field from more sandy texture soil (Association A) than the field of more loamy texture soil (Association A and Association B) at first level of division. Then the field of more loamy texture soil (Association C) from more clayey texture soil (Association D) at second level of division. So, at second level of division, four associations of distinctively different vegetation vicinity emerged due to different soil characters (Figures 2).

Table 1: Names of associations/ number of stands by the Normal Cluster Analysis

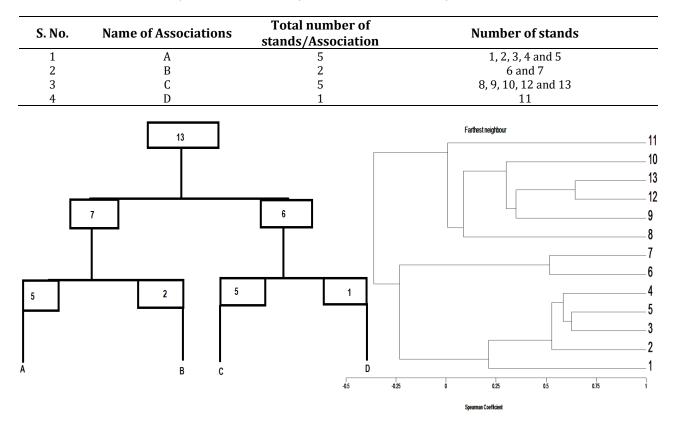


Figure 2: (a) Dendrogram of stands by Normal Cluster Analysis. Number of stands in respective boxes, (b) Dendrogram of Normal Cluster Analysis of stand of 13 sampling sites. (A= Association-A, B= Association-B, C= Association-C, D= Association-D)

#### 3.2. Classification of Wild Vegetation by Inverse Cluster A

Five associations were recognized by the inverse cluster analysis from the wild vegetation of the study sites (Table 2). These associations were delineated at three levels of division of hierarchical diagram (Figures 3). The most obvious attribute signified by the Inverse cluster analysis was the separation of the entire wild vegetation fields from more sandy texture soil (Association-A) than the field of more sandy texture soil (Association-A) separated from sandy loam texture soil at second level of division. Then the field of more loamy texture soil (Association-C) from more clayey texture soil (Association D and E) at third level of division. So, at third level of division, five associations of distinctively different vegetation vicinity emerged due to different soil characters (Figures 3).

Table 2: Names and numbers of plant communities/ associations by the Inverse Cluster Analysis.

S. No.	Plants Communities	Total Number of Species	Name of Species/ communities
1	А	6	Aerua persica, Calotropis procera, Cenchrus celeries, Citrulus colocynthes, Acacia modesta, Cynodon dactylon
2	В	8	Rhazya stricta, Suaeda fructicosa, Leptadaenia pyrotechnica, Zizyphus numularia, Panicum antidotale, Tribulus terrestris, Acacia jacuomentii, Acacia nilotica
3	С	7	Prosopis cinneraria, Capparis aphylla, Saccharum munja, Salsola foetida, Fagonia indica, Withania somnifera, Solanum surrettense
4	D	9	Desmostachya bipinnata, Cyperus rotundus, Pulicaria crispa, Tamarix indica, Arundo donax, Bracharia ramosa,Cressa cretica, Paganum harmala, Parkinsonia aculeata
5	Е	9	Typha elephantina, Nelumbium speciosum, Dalbergia sisso, Phoenix dactylifera, Phyla nodiflora, Azadirecta indica, Solanum nigrum, Alhagi maurorum, Physalis pubescens

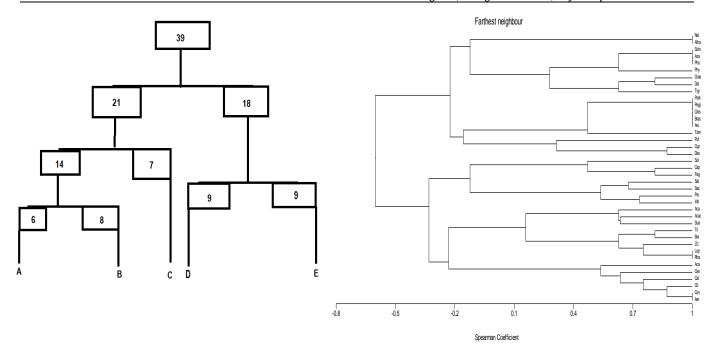


Figure 3: (a) Dendrogram of species by Inverse Cluster Analysis. Number of species in respective boxes, (b) Dendrogram of Inverse Cluster Analysis of 39 species from 13 sampling sites. (*A= Association-A, B= Association-B, C= Association-C, D= Association-D*)

#### 3.3. Ordination:

DCA the results obtained from classification. The results of the DCA ordination are used to plot the scatters of classificatory results. Figure 4 shows the four major groups/ associations of wild vegetation derived from Normal Cluster Analysis, although some species were common and found in all associations identified by classification. The vegetation associations identified by multivariate were structurally floristically distinct from each other. Table 3 shows the floristic composition of these associations. A description of the groups/ associations identified by classification will know be presented.

Allalysis.				
Species Name	Association-A	Association-B	Association-C	Association-D
Acacia jacumantii	2.5	1.0	1.0	
A. modesta	0.5	0.5		
A. nilotica	0.5	1.5	0.5	
Aerua persica	3.0	2.0		
Alhagi maurorum			1.5	1.0
Arundo donax		0.5	0.5	
Azadirecta indica			0.5	1.5
Calotropis procera	3.0	1.5	0.5	
Capparis aphylla	0.1	0.1	3.0	
Cenchrus celeries	0.5			
Citrullus colocynthes	1.0			
Cressa cretacia		1.0	1.0	
Cynodon dactylon	1.0	1.5	1.0	
Dalbergia sisso		0.5	0.5	1.0
Desmostachya bipinnata		1.5	2.5	1.0
Fagonia indica	0.5	1.5	1.5	
Leptadaenia pyrotychnica	0.1	1.5		
Nelumbium speciosum			1.5	2.0
Panicum antidotale	0.5	1.0		
Peganum hermala		0.5	0.5	
Phoenix dactylifera		0.5	1.5	1.0
Phyla nodiflora			0.5	0.5
Physalis pubescens			0.5	0.5
Prosopis cinneneria		0.5	3.4	
Pulicaria crispa			0.5	
Rhazya stricta	0.5	2.5		
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Table 3: Relative frequency	of species	association	(A-D)	identified in	n wild	vegetation	by Normal	Cluster
Analysis.								

3.3.1. Association-A: Aerua persica and Calotropis procera Coummunity:

Association A consist of five different sampling sites. Sites: 1, 2, 3, 4 and 5 (Table 1). All these stands were existed at right hand side (at the lower score) along the DCA axis 1. *Aerua persica* and *Calotropis procera* was the dominant species and have equal share with relative frequency 3.0 (Table 3). Soil of this group was sandy loam with soil characteristics i.e., EC (10<sup>6</sup>) ranges 156-166, pH ranges 6.4-7.4, Soil saturation percentage ranges 21-26 and organic matter ranges 1.6-2.3 % (Table 4).

Table 4: Physical and chemical	properties of soil samplings	collected from of 13 stands

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Soil Sample/ Stand	EC X 10 <sup>6</sup>	рН	0.M %	P (ppm)	K (ppm)	N (ppm)	Ca+ Mg meq /L	Soil Satu. %	Texture of Soil
1	156	6.4	2.3	14	320	3.5	2.94	26	Sandy loam
2	166	5.4	2.2	15	369	2.5	3.8	21	Sandy loam
3	160	7.46	1.6	16	358	2.9	4.2	23	Sandy loam
4	180	8.6	3.2	7.6	325	4.3	4.08	38	Sandy clay loam
5	172	7.6	2.5	12.5	219	3.8	3.69	48	Sandy clay loam
6	310	8.4	0.89	6.9	278	4.4	3.98	39	Sandy clay loam

Diversity of Wild Vegetation Along the Aridity Gradient Habitat of Dera Ghazi Khan

Saccharum munja

Salsola foetida

S. surrettenese

Solanum nigrum

Suaeda fruticose

Tamarix indica

Tribulus terristeris

Typha elephantina

Withania somnifera

Zizyphus nimularia

 7	390	7.8	0.47	6.4	302	2.90	2.0	54	Clay loam
8	320	8.1	0.52	5.9	298	2.76	1.70	56	Clay loam
9	310	7.9	0.47	6.4	312	2.56	1.94	52	Clay loam
10	453	8.4	0.69	5.9	136	4.3	3.80	46	Clay loam
11	504	8.5	0.66	5.9	182	2.4	3.40	45	Clay loam
12	306	8.1	0.56	4.8	205	3.6	3.30	55	Clay loam
 13	398	7.76	2.02	17	2.3	542	2.29	38	Silt Clay loam

From the Pearson's correlation of soil characteristics with the DCA axis-1 and 2, it was observed that the soil texture significantly differences from the upland area- low land area. The correlative values of the results (0.804 P-Value= 0.001\*\*\* Table 5) showed significant difference in the soil texture between the right- and left-hand side of the DCA axis-1 (Figure 4).

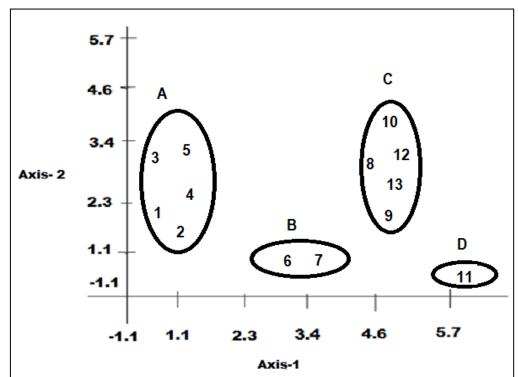


Figure 4: The DCA graphs (axis 1 and axis 2) plot of 13 sampling stands. Stands of Association (A-D) given in respective boxes. (*A= Association-A, B= Association-B, C= Association-C, D= Association-D*)

3.3.2. Association-B: Rhazya stricta and Suaeda fructicosa community:

Association B included two different sampling sites. Sites: 6 and 7 (Table 1). All these stands were scattered at the middle score of DCA axis-1 of the ordination diagram (Figure 5). *Rhazya stricta* and *Suaeda fructicosa* are dominant and association making species having relative frequencies i.e., 2.0 and 3.0 (Table 3) respectively. Other xerophytic species such as *Aerua persica, Fagonica indica* and *Leptadaenia pyrotechnica* were also the member of this association having different degree of relative frequencies (Table 4).

Table 5: Correlation: Axis 1, Axis 2, EC, pH, O.M, P (ppm), K (ppm), N (ppm), Ca+Mg, Soil Sat. and Texture

Axis 1	Axis 2	EC	рН	0.M	P (ppm)	K (ppm)	N (ppm)	Ca+ Mg	Soil Sat.	Texture
Axis 2	0.206 0.499									
EC	0.611 0.027	0.538 0.058								

	0.433	0.181	0.569							
рН	0.139	0.553	0.042							
0 M	-0.365	-0.042	-0.690	-0.379						
0.M	0.220	0.891	0.009	0.201						
D (nnm)	-0.436	0.028	-0.525	-0.669	0.649					
P (ppm)	0.136	0.929	0.065	0.012	0.016					
K (nnm)	-0.611	-0.474	-0.642	-0.385	0.114	-0.028				
K (ppm)	0.027	0.102	0.018	0.193	0.711	0.928				
N (ppm)	0.270	0.213	0.260	0.014	0.200	0.490	-0.735			
м (ррш)	0.371	0.485	0.391	0.965	0.512	0.089	0.004			
Cal Ma	-0.382	0.229	-0.353	-0.040	0.447	0.191	0.147	-0.292		
Ca+ Mg	0.197	0.451	0.236	0.897	0.125	0.533	0.632	0.333		
Soil Sat.	0.678	-0.133	0.562	0.655	-0.617	-0.773	-0.303	-0.089	-0.544	
5011 Sat.	0.011	0.665	0.046	0.015	0.025	0.002	0.314	0.771	0.055	
Texture	0.804	0.226	0.816	0.616	-0.515	-0.448	-0.739	0.505	-0.587	0.757
Texture	0.001	0.457	0.001	0.025	0.072	0.125	0.004	0.078	0.035	0.003

Cell Contents: Pearson correlation

Soil of this group was sandy loam and sandy clay loam in texture with soil characteristics i.e., EC (10<sup>6</sup>) ranges 172-310, pH neutral to slightly alkaline ranges 7.6-8.6, Soil saturation percentage ranges 38-48 and organic matter ranges 0.89-3.2 % (Table 4). From the Pearson's correlation of soil characteristics with the DCA axis-1 and 2, it was observed that the soil texture and Soil saturation percentage were significantly/ positively increased but the exchangeable potassium negatively correlated from the DCA-axis-1. The correlative values of the results (0.804 P-Value= 0.001\*\*\*, 0.678 P-value= 0.011\*\* and -0.611 P-value= 0.027\*\* respectively Table 5) It showed that significant difference in the soil texture and soil saturation percentage between the right- and left-hand side of the DCA axis-1 (Figure 5).

#### 3.3.3. Association-C: Prosopis cinneraria and Capparis aphylla community:

Association C included five different sampling sites. Sites: 8, 9, 10, 12 and 13 (Table 1). All these stands were scattered at the high score of DCA axis-1 and axis-2 of the ordination diagram (Figure 6). *Prosopis cineraria and Capparis aphylla* are the dominant species of this association and having the highest relative frequencies i.e., 2.5 (Table 3). Other frequently species were *Desmostachya bipinnata, Saccharum munja* and *Alhagi Maurorum* with relative frequencies ranges 1.5-2.5 (Table 3).

Soil of this group/ association was clay loam with soil characteristics i.e., EC (10<sup>6</sup>) ranges 310-453, pH ranges 7.8- 8.5 mostly alkaline in nature, Soil saturation percentage ranges 45-56 and organic matter ranges 0.47-0.69 % (Table 4). From the Pearson's correlation of soil characteristics with the DCA axis-1 and 2, it was observed that the soil texture significantly differences from the upland area- low land area. The correlative values of the results (0.804 P-Value= 0.001\*\*\* Table 5) showed significant difference in the soil texture between the right- and left-hand side of the DCA axis-1 (Figure 6).

#### 3.3.4. Association-D: Typha elephantina and Nelumbium speciosum community:

Association D included only one sampling site 11 (Table 1). This stand was scattered at the highest score of DCA axis-1 of the ordination diagram (Figure 6). *Typha elephantina* and *Nelumbium speciosum* are the dominant species of this association having relative frequencies i.e., 2.0 (Table 3). *Azadirecta indica, Dalbergia sisso, alhagi maurorum, Phyla nodiflora* and *Physalis pubescens* were found more or less in this Association D.

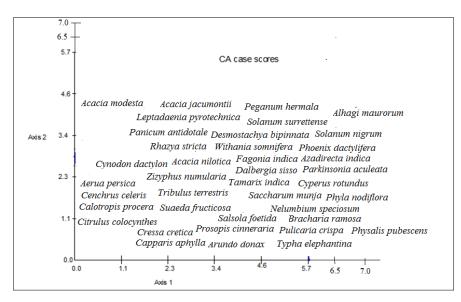


Figure 5: The DCA (Axis-1 and Axis-2) plot of the 39 plant species of study area

Soil of this group was clay loam and silty loam in texture with soil characteristics i.e., EC (10<sup>6</sup>) ranges 306-504, pH ranges 7.7-8.5, Soil saturation percentage ranges 38-55 and organic matter ranges 0.56-2.0 % (Table 4). From the Pearson's correlation of soil characteristics with the DCA axis-1 and 2, it was observed that the soil texture significantly differences from the upland area- low land area. The correlative values of the results (0.804 P-Value= 0.001\*\*\* Table 5) showed significant difference in the soil texture between the right- and left-hand side of the DCA axis-1 (Figure 6).

#### 3.4. Correlates of Ecological Attributes

The results (Table 4) clearly demonstrated the relationship between distribution of vegetation and edaphic factors. The eigenvalues of DCA axis 1 and 2 were very large in comparison with lower order axes, and later was ignored (Table 6).

Table 6: Eigenvalues of 38 species and cumulative percentage of DCA Axis 1-4 of natural vegetation along the aridity gradient.

Axis	Eigenvalues	Percentage	Cum. Percentage
Axis 1	1.00	24.519	24.519
Axis 2	0.603	14.781	39.289
Axis 3	0.571	12.682	51.981
Axis 4	0.264	6.469	58.45

When the four natural vegetation associations produced by the cluster analysis are plotted on the first two axes as a scatter diagram. The DCA showed considerable difference in habitat preference of the plant vegetation studied. Figure 2 and 3 shows the occupied of vegetation Associations of the sites on the DCA axes. It is clear that species such as *Acacia modesta, Aerua persica, Calotropis procera Citrullus colocynthes,* and *Cenchrus celeris* (Association A) are found on the right side on DCA axes. Species of association B such as *Rhazya stricta, Suaeda fructicosa, Acacia jacuonentii, Leptadaenia pyrotechnica, Acacia nilotica, Zizyphus numularia and panicum antidotale* are scattered at the high score of DCA axis-1 and axis-2 of the ordination diagram (Figure 4 and 5). The clay loam soil of association C with *Capparis aphylla, Arundo donax, Prosopis cineraria, Fagonia indica, Salsola foetida, Dalbergia sisso, Tamarix indica and Withania somnifera* occupied mostly middle and on the large, distributed axes of the ordination diagram (Figures 5 and 6). The association D containing species like *Typha elephantina, Nelumbium speciosum, Physalis pubescens, Alhagi maurorum, Cyperus rotundus, Phyla nodiflora* and *Parkinsonia aculeata* are present at the high score of the ordination diagram and some species of this association are hydrophytes and found in the flooded area of the Dera Ghazi Khan Landscape.

The second axes of the ordination diagram were clearly demonstrated that *Peganum hermala* is the species occur only at middle of the ordination graph having high EC and PH and high O.M % in comparison with all observed stands. *Acacia modesta, Acacia jacuomentii, Peganum harmala, Solanum surrettense* and *Leptadaenia pyrotechnica* are found at the high score of the ordination figure of the axix-2 (Figure 6) having very low EC, slightly basic pH and medium O.M % in comparison with all observed stands.

#### 3.5. Distribution of Species

The Table 6 gives the loading of two DCA axes, eigenvalue for 39 species (Table 6) of plants found in the area under observation. The highest loading of first DCA axis *were Peganum hermala, Acacia jacumentii, Solanum surrettense, Alhagi maurorum* and *Leptadaenia pyrotechnica* and showed this arrangement in the Table 6.

#### 4. Discussion:

The spatial distribution of wild plant community research usually focuses on the individual species to determine its environmental features and suitable management strategies in particular area. In this study we collected the information regarding the distribution of natural vegetation along the aridity gradient of Dera Ghazi Khan. This approach can provide an opportunity to identify factors that should be subjected to more detailed about the distribution of species at diversified environment of the study area. The data show that environmental factors, such as climate and disturbance, account for a large proportion of the variance of different components of biodiversity. The measurements of species diversity (richness, Simpson, evenness), however, did not vary similarly to other components of biodiversity (such as the functional diversity or the number of rare species in a community) along the considered environmental gradients. In particular, the increased species diversity with increased grazing intensity or moisture was not followed by similar trajectories for functional diversity. The Functional diversity, for example, did not differ between the most species' poor semi-arid locations and the most species-rich humid ones. Semi-arid rangelands, despite their relatively lower species diversity, showed a more even composition in different growth forms and life cycles than the species diverse humid grasslands, which were composed mainly by herbaceous perennials, as hypothesized by Vesk et al. (2004). This shows that the functional differentiation among species' and species richness can vary rather independently to each other. It has been noted that environmental filters might limit species composition to a given range of functional characteristics (Diaz & Cabido 2001) thus limiting the degree of variation of functional diversity with respect to species diversity (Grime, 2001; Hooper et al., 2005). The increased species richness within a more homogenous pool of traits thus might lead to a finer division of the available niche space among similar species rather than to a greater functional diversity (Diaz & Cabido, 2001). Results also suggests that drought, for some plant traits, might imply the coexistence of functionally dissimilar species (Mason et al., 2005), resulting probably in complementary uses of resources (Stubbs & Wilson, 2004). The indices of species and functional diversity varied also differently in response to sheep grazing regime. The stronger positive effect of grazing intensity on species diversity in humid conditions in comparison with arid ones is generally consistent with the prediction that competitive exclusion in the absence of disturbance would be faster in productive environments (Huston, 1994; Dupre & Diekmann, 2001; Leps, 2005; Cingolani et al., 2005; Bakker et al., 2006; Petru et al. 2006).

In the location where species diversity markedly increased with grazing, functional diversity presented the opposite pattern and decreased with grazing. The decrease of FD might confirm that livestock act by removing species that lack set of traits for persisting under grazed conditions, i.e., to avoid or tolerate defoliation (Diaz et al., 2001). This selection of traits should thus decrease the overall dissimilarity among species. However, the effect of grazing on FD tended also to be positive in more arid locations. This could depend on the fact that, in more patchy vegetation, grazing might also increase heterogeneity in resource distribution (Adler et al., 2001) and promote the coexistence of species with dissimilar resource acquisition strategies (Stubbs & Wilson, 2004) through an increase of aggregation patterns (Pugnaire et al., 2004). The potential link between species spatial distribution and dissimilarity of their traits (Stubbs & Wilson, 2004) opens up field for further research. The role of grazer's characteristics and size (Cingolani et al., 2005; Bakker et al., 2006) might also exert an influence on FD variation: animal selective effects on species' traits might be less marked in sheep grazing systems than in cattle grazing ones, as cattle are normally more generalists in their consumption. Our study showed an overall low co-variation between species and functional diversity (Fig 1 and 2), indicating that the mechanisms that support the coexistence of many species not necessarily support the functional differentiation among those species (Huston, 1994; Fukami et al., 2005). This implies that the diversity of species need not capture consistently the functional differences among species that regulate ecosystem processes. If we assume that ecosystem functioning is mainly dependent on functional diversity, because functional

diversity is a link between species traits and ecosystem processes (Loreau *et al.*, 2002; Petchey & Gaston, 2006), then the link between species diversity and ecosystem functioning might be context dependent.

As discussed above, the degree of dependence of functional diversity on species diversity might be mostly related to the amplitude of the species' traits pool and on how species divide into the niche space available. This is a potential ground for understating the mechanism that regulate the functioning of biodiversity under different environmental constraints (Schmid & Hector, 2004). The development of new indices of FD, such as the functional generalization of the Rao index, will also help testing whether the increase of FD reflects higher degrees of niche differentiation, lower resource competition and more efficient resource use (Mason et al., 2005). In this study, species that are catalogued by local floras as "very rare" were related principally to ungrazed habitats (at least in all but the driest locations), which were generally less species diverse. Thus, the species diversity needs not be a good indicator of the potential contribution of communities to the maintenance of diversity at the regional level (Pyka"la" et al., 2005). As disturbance may promote ruderal species, with generally a low conservation interest (Spitzer et al., 1993; Canals & Sebastia, 2000), the increase of the number of species with grazing may not reflect a high conservation value in species composition. This evidences that species diversity might not always be a synonym of ecological quality (Canals & Sebastia, 2000), for example in terms of conservation priorities, because species identity is not taken into account into biodiversity indices (Magurran, 2004). Finally, the use of different biodiversity indices could lead to the definition of different criteria by which areas/ management should be promoted. However, at the same time, the independence of different biodiversity components might offer the advantage of a better insight into mechanisms that regulate species coexistence and their links with ecosystem properties. Biodiversity studies can thus attain a more complete understanding of diversity, and its functioning, by the compared analyses of various components of diversity.

The analysis of complex ecological data required more advance tool than those currently available if valuable information is not to be last (Begon *et al.*, 1996). More complex modeling method, Detrended Correspondence Analysis (Hill & Gauch, 1980), can greatly assess interpretation by retaining more information about the data set. DCA is particularly better able to handle the complex data set and was used in the present investigation for data simplification and correlation of the edaphic variables included in the present study. When the plant associations produced by the cluster analysis are plotted on the first two axes as a scattered diagram (Figures 5 and 6) the two procedures of data simplification can be seen to have given very similar results. We take these results to confirm the clear-cut boundaries of the associations of natural vegetation in the space defined by ordination axes.

The ordination axes may represent in some way the major ecological variables which affect the stands in these data, and we use the plant and soil characteristics of the associations (Figure 5 and 6) to discuss these overriding features of the environment. The results shown in the correlation suggest that a large number of factors can influence the distribution of vegetation in the study area (Table 5). Although spatial distribution and plant species assemblage are poorly explained by morphological attributes, they are strongly associated with edaphic factors. The most important factor affecting vegetation communities was the soil texture, soil saturation percentage, Electrical conductivity and exchangeable potassium (R= 0.804, 0.678, 0.611 and -0. 611, p=0.001\*\*\*; P=0.011\*\* and P= 0.027\*\* respectively Table 5). These findings agree the findings of Qiang & Hu, (1999), Qiang & Liu, (1996), Qiang et al. (1994) and Qiang & Li, (1990). In the study area, the natural vegetation distribution is potentially affected by soil properties (Hüppe & Hofmeister, 1990; Pinke & Pal, 2008; Pinke et al., 2010; Zare et al., 2011). Analysis with DCA confirms that there is a clear relationship between soil physical and chemical feature of the substrate and the associations defined by the numerical analysis (Table 4) which surely shows that these associations of plant species are not randomly assemblage. The DCA results showed that soil texture, soil saturation percentage, electrical conductivity and exchangeable potassium criteria are the most important factors for the distribution and assemblage plant species. Petry et al. (1991) observed the influence of organic matter, while Andreasen & Streibig, (1990) and Sharma, (1986) observed the influence of soil texture on vegetation. In this respect, soil texture seems particularly important in plant community construction and distribution; soil texture governs the hydrological properties and soil moisture retention, dynamics of soil organic matter and influence the availability of nutrients to plants (Sperry et al., 2002) which are of overriding importance in agriculture fields (Udoh et al., 2007). The distribution of species like Aeua persica and Calotropis procera seems to be more influenced by soil texture. These species were confined to sandy loam textured soil (association A) and are altogether absent in samples from soils having high silt or clay content. Numerical analyses confirm these findings. Beside the soil texture, soil pH explains a significant role in community composition and plant assemblage. Species belonging to association A occur only on more basic soil. However, the more general reasons behind community differences on soils of different pH remain to be investigated thoroughly. The establishment of Typha elephantine and Nelumbium speciosum is dependent on the availability of soil moisture and is thought to occur through infrequent flood events and found at the left side of the diagram (Figure 6)

at the high score of DCA axis-1 and followed the findings of (Roberts, 1993; Eberhard, 1999; Stefano, 2002). In these areas flooding regimes together with soil properties appear to be important determinants of floristic variations and distribution. The results of the cluster analysis and again the scatter of ordination diagrams (Figures 5 and 6) demonstrate the boundaries between vegetation type and species distribution: some species were restricted. None of the divisive protocol increased the separation. The substrate of all these communities is found through the interaction of various agriculture practices and basic geo-morphological processes of surface movement through the action of wind and water. These factors may be contemporary or historical and range from sweeping changes that have taken place in the hydrological system of the study area, reviewed by Allchin *et al.* (1978), Meher-Homji, (1985), Gupta (1986) and Dasti & Agnew (1994) to current agricultural activities of the local farmers. These anthropogenic disturbances could well give rise to the observed, apparently arbitrary, changes in the community composition in the long term also it has been suggested that certain soil properties may be related. For the area under study, therefore, we suggest that the flora is a residual of generalists capable of persisting through disturbance regimes of various time scale and destructiveness, and that this can explain the difficulty of describing clearly the vegetation in terms of discrete communities.

#### 5. Conclusion:

The current investigation highlights a number of research areas, where knowledge about vegetation communities is very poor. The study of wild growing vegetation is a complex structure. Summarizing the main findings, it may be concluded that both classification and ordination are able to delimit the communities along the aridity gradient of the study area. Edaphic properties at the regional scale are an important factor that governs the community structure from the xerophytic sandy loam textured soil to wet clayey loam soil. The findings of this study provide clear evidence about the distribution pattern of communities is influenced by habitat and edaphic factors. Ordination as well as classification showed the clear cut boundaries of the wild vegetation along the aridity inclined of proposed study area and delimit into four association/ groups of vegetation from xerophytic community A and B (irrigated by hill torrents) – canal water community C (Mesophytes) to flooded area community D The final findings of this study demarcated the wild vegetation into three clear cut boundaries of the spatial diversity of the area of vegetation of study area of Dera Ghazi Khan.

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Diversity of Wild Vegetation Along the Aridity Gradient Habitat of Dera Ghazi Khan

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### Evaluation of Selected Landraces for Yield and Yield Attributes from District Bannu Against Drought Stress

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#### Abstract

*Brassica rapa* play an important role in oil production for daily consumption. In the present study 8 different landraces i.e., SSK, SAK, SU, BWA, KA-1, KA-2, IN, BA were collected from the different region of Bannu along with NG as a control from Sarai Naurang Agriculture research station. All the samples were grown in three replicate each in two different blocks. The one block was kept as a control while the other was treated with drought stress. Data for PH, LL, LW, NLP, NSP, NSS, SD, TSW, PGW in replicated from both the blocks were recorded. The data was subjected to correlation and ANOVA and LSD. In control high yield was recorded for KA-1 followed by IN, BA, NG, respectively while maximum seed diameter showed by BA followed by KA-2. BA seed showed resistance against drought stress while IN showed high yield against drought stress. The study further recommends the molecular analysis of different genes involved in pathways that influence plant yield.

Keywords: Yield; District Bannu; Drought Stress; Phenotypic; Landraces

#### 1. Introduction:

The *Brassicaceae* previously known as family Cruciferae, carries 338 genera and around 3700 species. Among the other genera of the family, the Genus Brassica is economically more vital and significant (Rakow & Raney, 2003) that comprised of 37 different species, including annual and biennials and even weedy, wild type and some domesticated crops species. It is believed that the vegetable species of the Genus Brassica domesticated in region between Mediterranean and Sahara of mild winter trailed by dry and hot summer while many species are found as invasive species in Australasia and in North and South America (Rakow, 2004). In addition, many species also grow in as invasive weeds in the Americas (North and South) and Australasia (Rakow, 2004). This genus contains six important annual species which are cultivated globally. The oil obtained from these species is used for human consumption in addition to other industrial uses and the meal after the extraction of oil is used as animal feed. as oilseed crops, condiments, fodder or vegetables Three of six species are diploids (2x) viz., *B. rapa* (2n=20); *B. nigra* 

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(2n=16) and *B. oleracea* (2n=18), while the remaining three (*B. juncea*, 2n=36; *B. napus*, 2n=38; *B. carinata*, 2n=34) are amphidiploids (4x) derived from chromosome doubling in the hybrids obtained from three possible crosses among the diploid species. Commonly four Brassica species have been cultivated as oilseed crops viz., *B. napus*, *B. rapa*, *B. carinata*, *B. juncea* globally (Song *et al.*, 1990) and they are also important in Pakistani context. *Brassica*, as mentioned earlier, is used as vegetable, grain, oilseed and green compost for soil restoration (Gomez-Campo, 1980; Williams & Hill, 1986). In Pakistan, oilseed *Brassica* is cultivated every year on 482,000 acres with the production of 186,000 t of seed, which produced 61000 t oil (GoP, 2013). In Pakistan five species viz., *B. rapa*, *B. napus*, *B. juncea*, *B. carinata* and *Eruca sativa* of rapeseed and mustard are cultivated in particular (Munir and Khan, 1984). Furthermore, Brassica species are vital sources of potassium; dietary fiber; vitamins A, C, and E; phenolics; and other health-enhancing factors (Fahey *et al.*, 1995; Zhao, 2007; Esawi, 2015). Brassicaceae contains glucosinolates which are broken down to isothiocyanates known to mitigate tumor development and resist a range of heart diseases and human cancers (Christopher *et al.*, 2005; King, 2015; Esawi, 2015). The plants comprising high amount of glucosinolate may be further utilized as a potential genetic source for breeding (Faltusová *et al.*, 2011). Brassica vegetables inhibit major diseases such as Alzheimer's, and some of the functional declines associated with aging (Esawi, 2015; King, 2015; King, 2015).

*Brassica rapa* is one of the important vegetable oil crop species, belong to important family Brassicaceae. It consists of wide ranges of leafy or root vegetable types such as Chinese cabbage, pak-choi etc. that used as human diet (Zhao *et al.*, 2005). Estimation of various qualitative and quantitative traits provides a clear picture of improve diverse genotypes. Among these characters the yield parameter plays a vital role for new variety development (Ali *et al.*, 2013; Azam *et al.*, 2013). Rapeseed-mustard is the major contributor among traditional oilseed crops (Ali & Mirza, 2005). Rapeseed-mustard is being grown on marginal lands scattered in whole Pakistan on considerable area of 228 thousand hectares (Anon., 2008).

Morphological evaluation is the primary step in description and categorization of the germplasm (Taylor et al., 1991). Morphological differences in plant species are the result of long-term selection/ adaptation in different parts of the world where the species were initially cultivated. Different techniques have been effectively used to find the pattern of phenotypic diversity in species indicating genetic differences of a variety of crops (Dias et al., 1993; Amurrio et al., 1995). To expand the existing genetic pool, breeders collect and evaluate the germplasm from all over the world for their genetic diversity (Alo et al., 2011). Evaluation of phenotypic characters enhances the efficiency of germplasm collection management (Nisar et al., 2008) as well as the genetic improvement of crops leading to the higher genetic gain (Basheer et al., 2015). Plants growing under field conditions are subjected to various environmental stresses, such as high or low temperature, drought and salinity. Plants naturally face many unfavorable environmental conditions, such as organic and inorganic stresses. In the case of all other stresses, the heavy metal pressures have a negative impact on the crops yield and growth. Heavy metal stress induces unlike responses in the plants, ranging from biochemical reactions to the yield of the crops (Basit *et al.*, 2022). At any given point of time, plants may have to face two or more stresses (e.g., drought and salinity). Among these stresses, drought is the most serious problem for global agriculture, approximately affecting 40% of the world's land area (UN Environment Management Group, 2011). Even worse, global climate change is predicted to lead to extreme temperatures and severer prolonged drought in some parts of the world, which will have a dramatic impact on crop growth and productivity (Trenberth et al., 2014).

The loss of farmable land due to either salinization or drought has posed a major challenge for maintaining world food supplies for the growing population. Thus, there is an urgent need to develop varieties that can maintain optimum yield levels under abiotic stresses. This objective has been highlighted by the United Nations Secretary General Kofi Annan: "We need a Blue Revolution in agriculture that focuses on increasing productivity per unit of water—more crop per drop" (Pennisi, 2008). However, due to the multigenic and quantitative nature of stress tolerance in plants, efforts to improve crop performance under drought and salinity have been elusive. This challenge has given plant scientists an impetus to improve drought and salt tolerance. Among the major food crops, Brassica crops are the most affected by drought and salinity, due to the fact that they are mainly grown in arid and semiarid areas.

#### 1.1. Objectives:

- To collect different landraces at District Bannu.
- To evaluate the landraces for yield attributes and drought tolerance

#### 2. Materials and Methods:

#### 2.1. Study Area:

The present investigation was carried out in 2018-2019 in the Department of Botany, University of Science and Technology, Bannu. Continuous visits were taken in 2018 to the different areas of Bannu, FR Bannu and adjacent area of the district Lakki Marwat. Eight different landraces i.e., SSK, SAK, SU, BWA, KA-1, KA-2, IN, BA with NG as control were collected from the different areas based on their visual observation and were grown in two different blocks in the green house of the University of Science and Technology Bannu.

The first block contains three replicates for each landrace with control. The Second block contained each landrace with control under drought stress. The 1<sup>st</sup> block was well watered while the second block was irrigated in order to provide maximum moisture before one week of sowing and after the field was maintained under rain-fed condition by withholding water. was kept under drought stress. Once germination occur the excess of weeds were removed, and pesticides were sprayed in whole field.

Phenotypic data of three selected plants was taken regularly and its various important quantitative agromorphological traits were studied such as days to flower initiation (DFI), days to 50% flower (50% DF), days to flower completion (DFC), leaf length (LL), leaf width (LW), pod shatter resistance-I-IV, plant height (PH), main raceme length (MRL), pods per main raceme (P/MR), stem thickness (ST), pod length (PL), pod width (PW), seeds per pod (S/P), seed yield per plant (SY/P) and 1000 seeds weight.



Figure 1: Field nursery of Brassica landraces for Control Vs Drought stress

#### 2.2. Statistical Analysis:

The data of both control and drought was analyzed for analysis of variance and subsequently in Statistic v.8.1. Further the data were also subjected to Pearson correlation among the traits of both matrices.

#### 3. Results:

#### 3.1. Phenotypic evaluation of control plots:

The control data subjected to the Pearson's correlation revealed the results presented in Table 1. Highly significant correlation was found between number of seliqua per plant and number of seeds per seliqua, between leaf length and leaf width, followed by seed diameter and thousand grain weight. Similarly, significant correlation

was found between leaf length and number of seeds per seliqua, followed between leaf length and number of seliqua per plant. Leaf width also showed significant correlation with number of seeds per seliqua and number of seliqua per plant. Leaf length was also correlated with number of leaves per plant. Seliqua width was also significantly correlated with thousand seeds weight.

PH LL LW NLP SL SW NSS SD **TSW** 0.5309 LL 0.8984\*\* LW 0.3743 0.7648\* NLP 0.6357 0.5527 0.6501 0.2075 SL 0.0850 0.7169 -0.0683 -0.2492 0.0336 SW 0.4678 -0.11730.2074 0.7873\* 0.7493\* 0.5810 0.4753 -0.1872 NSP 0.2069 0.7934\* 0.7543\* 0.4736 0.5969 -0.1854 NSS -0.0076 0.8527 SD 0.2253 -0.3071-0.5612 -0.2065 -0.34800.5924 0.1290 -0.2305 0.3360 0.1038 0.7369\* -0.1375 0.8262\*\* TSW 0.1276 0.5374 0.6700\* 0.4505 0.3529 0.1360 0.4373 -0.0823 -0.1143**PSW** 

Table 1: Pearson correlation between different traits of control

PH= (high in KA2) LL= (high in IN), LW= (high in IN), NLP= (High in BA), SL= (high in IN), NSP= (high in IN and BA, SSk), NSS= (high IN), SD= (BA, KA2,), TSW= (KA2), PSW= (KA1, IN, BA, NG)

In control plots maximum plant height (128.67 cm) was noted for KA-2 while minimum (102.67 cm) was recorded for SAK. Grand mean was 117.11 cm with SD of 8.9876 for plant height. The coefficient of variability for this trait was 7.6745%. The leaves length ranged from 10.667-18 cm. The grand mean for the LL was 14.259 cm with SD of 2.2117. CV% for LL was (15.511). For leaf width grand mean of 6.5704 cm was calculated. Leaf width ranged from 401667-9.1667 cm with 24.29% of coefficient of variations. The broad leaves were exhibited by IN while narrow leaves were found in the SAK. The grand mean for this trait was 6.5704 cm with 24.729% coefficient of variation and 1.6248 SD.

The highest number of leaves per plant was noted for the NG while lowest number of leaves per plant was recorded for SAK. The grand mean for the trait was 32.815 cm with CV of 33.832% and 11.102 SD. Similarly, grand mean of 3.7926 cm was calculated for seliqua length. The trait ranged from 3.1-6.4 cm. The 26.603% coefficient of variation and 1.009 SD was noted for the trait, respectively. The seliqua width ranged from 0.3533-0.45 mm. The grand mean for Seliqua length was 0.4074 mm with 8.5887% of coefficient of variation with SD of 0.035.

	РН	LL	LW	NLP	SL	SW
SSK	122.33BC	15.167ABC	7.7667AB	34.333BC	3.9B	0.42AB
SAK	102.67E	10.667C	4.1667C	15.333E	3.2667BC	0.4167AB
SU	103E	12.167BC	5.0667BC	25.333CD	3.1C	0.3533B
BKW	123.33AB	13.833ABC	6.4333ABC	23.333DE	3.5667BC	0.3967AB
KA-1	117CD	12.933ABC	6.8ABC	26.333CD	3.3BC	0.43AB
KA-2	128.67A	14.1ABC	5.1BC	41.667AB	3.6667BC	0.45A
NG	121BC	16.33AB	8.2A	50.667A	3.3BC	0.3633AB
BA	121.33BC	15.133ABC	6.4333ABC	41.667AB	3.6333BC	0.45A
IN	114.67D	18A	9.1667A	36.667B	6.4A	0.3867AB
Mean	117.11	14.259	6.5704	32.815	3.7926	0.4074
SD	8.9876	2.2117	1.6248	11.102	1.009	0.0350
CV	7.6745	15.511	24.729	33.832	26.603	8.5887
Min	102.67	10.667	4.1667	15.333	3.1	0.3533
MAX	128.67	18	9.1667	50.667	6.4	0.45

Table 2: Yield and yield attributes of control of different plant traits of Brassica landraces

Table 2: (Continued)

	NSP	NSS	SD	TSW	PSW
SSK	119.67AB	17.667BC	1.6333DE	2.5833EF	5.2157AB
SAK	54.33CD	16BC	1.9833B	2.77DEF	3.6870BC
SU	55CD	18.333B	1.62DE	2.3233F	3.1580BC
BKW	86BC	15.333BC	1.72CD	3.2917CD	2.4690C
KA-1	33.33D	14.667BC	1.8633BC	3.0367CDE	7.1223A
KA-2	34.33D	13.667C	2.1933A	6.1733A	3.2233BC
NG	112AB	15BC	1.5333E	2.2333F	6.144A
BA	125.67A	13.667C	2.2A	4.4333B	6.4163A
IN	139A	26.667A	1.6333DE	3.338C	6.5617A
Mean	84.407	29.536	1.8178	3.4289	4.8886
SD	41.164	13.917	0.2537	1.3141	1.7632
CV	48.768	47.119	13.955	38.323	36.068
Min	33.333	13.667	1.53	2.24	2.469
MAX	139	48.596	2.2	6.17	7.1223

PH= (high in KA2) LL= (high in IN), LW= (high in IN), NLP= (High in BA), SL= (high in IN), NSP= (high in IN and BA, SSk), NSS= (high IN), SD= (BA, KA2,), TSW= (KA2), PSW= (KA1, IN, BA, NG)

The number of seliqua per plant ranged from 33.333-139 cm with grand mean of 84.407 cm, CV of 48.768% with SD of 41.164. The highest number of seed per seliqua was recorded for IN while lowest for the trait was recorded for KA-2 and BA. The grand mean for NSS was 29.536 cm with 13.955% of coefficient of variations and 13.917 of SD. Seed diameter ranged from 1.53-2.2 mm. highest seed diameter was recorded for BA followed by KA-2. Grand mean for the trait was 1.8178 cm with CV of 13.955% with standard deviation of 0.2537. Highest 100 seed weight of (6.17 g) was recorded for KA-2 while lowest of (2.24 g) was recorded for NG which used as a control. Grand mean of (3.4289 cm) with 38.323% CV and 1.3141 SD was recorded. The most important trait per plant seed yield was weighted. Highest value of (7.1223) was recorded for KA-1, followed by IN, BA, and NG respectively. Grand mean of (4.8886 cm) with (36.068%) of CV and (1.7632) of SD was estimated.

#### 3.2. Phenotypic evaluation of drought stress:

The drought stressed data subjected to the Pearson's correlation revealed the results presented in Table 4.3. Highly significant correlation was found between leaf length and seliqua length followed by between leaf length and leaf width. Leaf length was also highly significantly correlated with plant height, number of leaves per plant, seliqua width, number of seliqua per plant, number of seed per seliqua, and per seed weight. Similarly, plant height was highly significantly correlated with seliqua length their number per plant and number of seeds within seliqua but significantly correlated with seliqua length, their number per plant and number of seeds within seliqua but significantly correlated with number of leaves per plant seed weight. Number of leaves per plant was highly significantly correlated with seliqua length, number of seliqua per plant seed weight. Number of seeds per seliqua. Seliqua width was highly significantly correlated with seliqua length and per plant seed yield. Number of seliqua per plant also showed significant correlation with per plant seed yield (Table 3).

	PH	LL	LW	NLP	SL	SW	NSP	NSS	SD	TSW
LL	0.7923**									
LW	0.7087*	0.9388**								
NLP	0.5567	0.9038**	0.9317**							
SL	0.7024*	0.9492**	0.8461**	0.8891**						
SW	0.889**	0.787**	0.6951*	0.5747	0.7523*					
NSP	0.7355	0.8704**	0.9325**	0.8142**	0.775*	0.5961				
NSS	0.5624	0.9369**	0.8784**	0.9278**	0.9147**	0.5798	0.7841**			
SD	0.7305*	0.6348	0.493	0.472	0.7569*	0.8412**	0.4791	0.4817		
TSW	0.4394	-0.159	-1696	-0.3915	-0.1961	0.351	-0.0357	-0.4451	0.3651	
PSW	0.6406	0.7962**	0.7309*	0.6493	0.6707*	0.7777**	0.5355	0.7559*	0.4439	-0.109

PH= (high in KA2) LL= (high in IN), LW= (high in IN), NLP= (High in BA), SL= (high in IN), NSP= (high in IN and BA, SSk), NSS= (high IN), SD= (BA, KA2,), TSW= (KA2), PSW= (KA1, IN, BA, NG)

Morphological evaluation of selected Germplasm when subjected to drought stress revealed highest plant height of (93 cm) for BA, while lowest (89.667 cm) were recorded for SU. Grand mean and standard deviation for plant height was (77.741 cm) and (10.862), respectively. Similarly highest leaf length (13.667 cm) was recorded for IN while (7.2 cm) was noted for SU. Grand mean and SD for leaf length was (9.7815 cm) and (1.8052), respectively. Narrow leaves of (2.9333cm) were recorded for SU while broad leaves of (5.9333 cm) were recorded for IN. grand mean (4.2852 cm) and SD (0.8273) was recorded for leaf width. Number of leaves among the Germplasm ranged from 8-25. Grand mean of (12.963 cm) and SD (5.2874) were noted for NLP. Highest Seliqua length of (4.84 cm) measured for IN while SU expressed lowest length of (2.72 cm). Grand mean of (3.2826 cm) and SD (0.6467) was recorded for SL. Highest silequa width of (0.4533 cm) was recorded for BA while lowest of (0.3067 cm) was noted for SU. Grand mean (0.3822 cm) and SD (0.0518) was noted for Siliqua length. Highest coefficient of variation was recorded for NLP, followed by number of seliqua per plant. Lowest coefficient of variation of reported for leaves length.

	PH	LL	LW	NLP	SL	SW
SSK	79.333B	10.333B	4.5B	14.667BC	3.2867BC	0.3833ABC
SAK	67D	9.367BC	4.0667BC	12BC	3.02BC	0.3633BC
SU	63D	7.2C	2.9333C	8C	2.72C	0.3067C
BKW	68D	8.5BC	3.833BC	10.333BC	3.0533BC	0.3733ABC
KA-1	73.333C	8.733BC	4.1BC	8.667C	2.7533C	0.34C
KA-2	88.667A	10.467B	5AB	16.333B	3.3933B	0.44AB
NG	77.667BC	9.333BC	4.2BC	12.333BC	2.9333BC	0.34C
BA	93A	10.433B	4BC	9.333BC	3.5433B	0.4533A
IN	89.667A	13.667A	5.9333A	25A	4.84A	0.44AB
Mean	77.741	9.7815	4.2852	12.963	3.2826	0.3822
SD	10.862	1.8052	0.8273	5.2874	0.6467	0.0518
CV	13.973	18.456	19.307	40.789	19.701	13.546
Min	63	7.2	2.9333	8	2.72	0.3067
MAX	93	13.667	5.9333	25	4.84	0.4533

Table 4: Yield and yield attributes of drought stressed of different plant traits of Brassica landraces

Table 4: (continued)

	NSP	NSS	SD	TSW	PSW
SSK	37B	15B	1.51B	2.38C	3.189AB
SAK	25.333BC	15.667B	1.5133B	2.15C	3.582A
SU	13.333C	11.667B	1.6033B	2.66ABC	2.1813C
BKW	28.333BC	12.667B	1.7967AB	2.6433ABC	2.6507ABC
KA-1	43.333B	12.667B	1.5133B	3.0333ABC	2.6007BC
KA-2	46.667AB	14.667B	1.89AB	3.4AB	3.358AB
NG	43.333B	14.667B	1.5333B	2.60333BC	2.7903ABC
BA	35.667BC	14.333B	2.0767A	3.56A	3.3487AB
IN	68A	21.333A	2.08A	2.1533C	3.5847A
Mean	37.889	14.741	1.7241	2.7315	3.0317
SD	15.414	2.7977	0.2422	0.5062	0.4940
CV	40.681	18.979	14.050	18.531	16.296
Min	13.333	11.667	1.51	2.15	2.1813
MAX	68	21.333	2.08	3.56	3.5847

PH= (high in KA2) LL= (high in IN), LW= (high in IN), NLP= (High in BA), SL= (high in IN), NSP= (high in IN and BA, SSk), NSS= (high IN), SD= (BA, KA2,), TSW= (KA2), PSW= (Ka1, IN, BA, NG)

Number of seliqua per plant ranged from 13.333-68. IN showed the high value while SU exhibited the lowest. Grand mean of (37.889 cm) with 15.414 was recorded for the trait. Highest number of seeds per seliqua of (21.333) was recorded for IN while lowest of (11.667) was noted for SU. Grand mean of (14.741 cm) with SD of (2.7977) was recorded for the trait. Seed diameter was ranged from 1.51-2.08 mm. IN exhibited highest value followed by BA. Lowest was recorded for SSK. Grand mean of (1.7241 cm) with SD of (0.5062) was recorded for the trait. Highest 100-seed weight of (3.56 g) was recorded was recorded for BA while lowest of (2.15 g) was recorded

Evaluation of selected landraces for yield and yield attributes from District Banuu against drought stress

for SAK. Grand mean of (2.7315 cm) with (0.5062) of SD was recorded for the trait. Plant seed yield for drought stress ranged from 2.1813-3.5847 cm. highest yields was recorded for IN followed by SAK. Lowest was recorded for SU. Grand mean of (3.0317 cm) with SD of (0.4940) was recorded for the trait (Table 4).

#### 4. Discussion:

Greater seed diameter was recorded for IN (2.08mm) followed by BA (2.07mm), while high number of seeds (21.33) in seliqua (NSS) was recorded for IN and highest 68 number of seliqua per plant was recorded for IN. Agromorphological based variation is important to screen best genotypes in field experiment (Martins *et al.*, 2006; Iqbal *et al.*, 2015) and to utilize such genotypes for further biochemical and molecular studies. The study of genetic diversity is a train that drives breeding for efficient utilization and development of improved varieties to meet future challenges (Sreenivasa *et al.*, 2020). Therefore, the proper strategies and planning is needed to evaluate local and exotic germplasm and to screen best genotypes among these for both qualitative and quantitative characters (Meena *et al.*, 2017). On the other hand, the conservation of germplasm is so important in so many reasons that one to retain the threatened or endangered species and another to preserve the vital germplasm domesticated and selected by our forefathers for many generations which could be used in novel breeding strategies for enhancing the yield quality and resistance against various biotic and abiotic stresses (Mickelbart *et al.*, 2015).

The present experiment was an attempt to document, preserve and screen out *Brassica rapa* landraces of District Bannu for various traits of essential contribution yield and other potency. Significant correlation was observed in many traits in the collected landraces which are in line with previous observation in *Brassica carinata* studied by Zada *et al.* (2013) and findings in *Brassica napus* by Gyawali *et al.* (2013). However, the yield and yield attributing traits response were varied with type of landrace collected.

Phenotypic traits associated with drought-tolerant crops serve as important breeding tools in identifying stress-tolerant genotypes and in introducing such tolerance traits into cultivated genotypes. However, most modern cultivars of *Brassica* crops are sensitive to drought and thus do not perform well under field stress conditions. In the present selection of landraces Germplasm, the landraces KA-1, showed high yield per plant followed by IN, BA, NG, respectively. While when stressed with drought the IN showed high resistant against the drought followed by SAK. SAK was not among the top high yielding landrace which shows IN and SAK high genetic resistance against drought tolerance. Drought tolerance in plants is recognised as a quantitative trait conditioned by many genes through various pathways. When drought stress is perceived by the plant, expression patterns are changed in genes, including those involved in water transport, osmotic balance, oxidative stress and damage repair (Zhang *et al.*, 2014).

Further the overall traits correlation harmony increases during drought stressed. One of the earliest attempts was made by Richards & Thurling, (1979b). *B. rapa* grown in a drought environment was assessed for grain yield improvement response to joint selection for yield, harvest index, 1000-seed weight and seeds per pod. It was shown that the joint selection was 20% more effective than direct selection for yield only under drought.

#### 5. Conclusion and Recommendation:

- > BA followed by KA-2 showed maximum seed diameter in control
- ▶ Highest yield per plant showed by KA-1 followed by IN, BA, NG, respectively.
- > During drought stress the seed diameter of BA remained high while KA-2 dropped severely.
- In normal condition the BA has high seed diameter than KA-2 but the thousand seed weight was higher in KA-2 which suggests that KA-2 is highly dependent on water.
- It is recommended that the seed diameter of BA should be mapped for genes involved and could be transferred to KA-1 or any high yield variety of Pakistan that is better adaptable to the local condition.
- ➢ KA-1 is recommended for irrigated land.
- > IN is recommended initially for rain-fed area for cultivation

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# First Record of *Pisolithus tinctorius* from Sangla Hill, Nankana Sahib, Punjab, Pakistan

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#### Abstract

In this study a species, *Pisolithus tinctorius* belonging to Gasteroid Mycota have been collected during several field tour of Shangla Hill, Pakistan from July-August 2019. This species has been described, illustrated and identified and are presented here, as a new record from Sangla Hill, Pakistan.

Keywords: Congo Red; Gasteroid; Gelba; Sangla Hill

#### Introduction:

The genus *Pisolithus* alb. & Schwein. described by Albertini and Schweinitz, 1805 are the member of order Boletales. This taxon is being described from dry and arid region of Pakistan. Shangla Hill is a tehsil in District Nankana Sahib of Punjab province. The soil of Shangla Hill is loamy with acidic pH of 6.7-6.9 (Naseer *et al.*, 2017). *Pisolithus* comprises complex of numerous species forming ectomycorrhizal association worldwide with the variety of woody plants. (Singla *et al.*, 2004; Yakhlef *et al.*, 2009).

The species of *Pisolithus* are thought to be host specific in nature and the species found mostly associated with *Eucalptus* spp. Are *P. albus, P. arrhizus, P. marmoratus* and *P. microcarpus* (Ducousso *et al.,* 2012). They may act as a bioremediating agent in cadmium effected sites and can also reduce the fungal diseases caused by *Fusarium moniliforme, Cylindrocarpon destructans and Rhizoctonia solani* on *Pinus sylvestris* (Cairney *et al.,* 1999; Kasuya *et al.,* 2010).

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#### 1.1. Objectives:

- To describe and illustrated the new species.
- To identify and record first time a new species from Sangla Hill, Pakistan

#### 2. Materials and Methods:

#### 2.1. Study Area and Collection of Specimens:

The specimens were collected from various sites of Shangla Hill from July-August 2019. The basidiomata morphologically analyzed and photographed on the spot. The anatomical characterization was done by preparing slides in 5 % potassium hydroxide and Congo Red, observed under light microscope (Labomed Lx200). The main characters were noted, measured, photographed and illustrated.

#### 3. Results:

Pisolithus tinctorius (Pers.) Coker & Couch, Gasteromycetes E. U.S. Canada (Chapel Hill): 170 (1928)

#### 3.1. Morphological Characterization:

Basidiomata irregular shape, club-shaped, sub globose, pyriform, whitish yellow to brown, 30–40 mm in diameter × 40–50 mm in height; pseudo stipe up to mm long, basally attached to substratum or buried below ground, yellow mycelial strands covered with soil particles; ostiole lacking. Exoperidium off white to brownish-black, smooth or rough, peeling off in patches to expose the endoperidium. Endoperidium dark brown, firm, rugulose, typically cracking apart apically to allow dehiscence. Gleba consists of three layers upper cinnamon-brown; middle white and at base yellowish, at maturation ferruginous to snuff-brown powdery mass formed by the breakdown of peridioles. Peridioles are hyphal aggregation containing spores arranged in a small black chamber, ovoid-ellipsoid white to yellowish brown and later dark brown to ferruginous as mature, distributed throughout the powdery gleba, more apparent at the base and becoming pulverulent toward the apex (Figure 1).

#### 3.2. Microscopic Characterization:

Basidiospores 8.2–11.4  $\mu$ m (without ornamentation 5.9–8.8  $\mu$ m), globose to sub globose, yellowish green in 5% KOH, ornamented, verrucose, apedicellate, thick walled with no oil droplet. Pseudoperidiolar hyphae hyaline, septate, branching rare, thin walled, 2.85–5.7  $\mu$ m in diameter (Figure 2). Peridium consist of 2–4.5  $\mu$ m, hyaline, septate, rarely branches, thin-walled hyphae with frequent clamp connections. Rhizomorphs consist of septate hyphal cells 4.5–12.5  $\mu$ m reddish-brown in 5% KOH (Figure 3).

#### 3.3. Examination of Collected Specimen:

**PAKISTAN:** Punjab, District Nankana Sahib, Sangla Hill, Morar Chak 42, on ground, under *Eucalyptus camaldulensis*, at 194 meters (636 ft.) a.s.l., 16<sup>th</sup> July 2019, A.N. Awan (SH-K). Morar Ruriwala, on soil, under *Eucalyptus camaldulensis*, 194 meters (636 ft.) a.s.l., 25<sup>th</sup> July 2019, A.N. Awan (SH-G); Morar Chak 42, on ground, under *Eucalyptus camaldulensis*, at 194 meters (636 ft.) a.s.l., 17<sup>th</sup> July 2019, A.N. Awan (SH-J).





Figure 1: (Plate 138) A–F. Morphology of basidiomata of *Pisolithus tinctorius* (SH-K). A. Basidioma. D. Exoperidium. E & F. Mature basidiomata with gleba. Scale bars: A= 6.6 cm, B= 5.2 cm, C= 8.7 cm, D= 5.8 cm, E= 5 cm, F= 6.3 cm

New Record of Pisolithus tinctorius from Sangla Hill, Nankana Sahib, Punjab, Pakistan

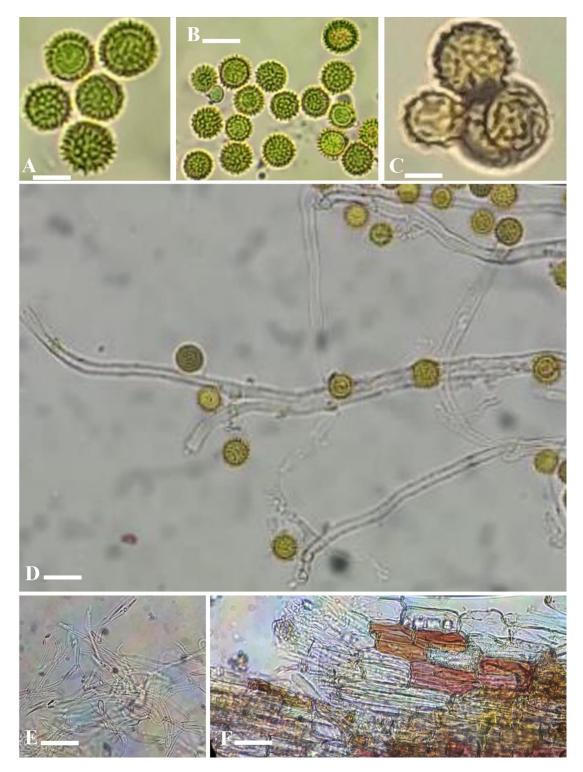


Figure. 2 (A–F). Light micrographs of microscopic features of *Pisolithus tinctorius* (SH-K). A–C. Basidiospores. D. Capillitium. E. Exoperidium. F. Rhizomorph hyphae. Scale bars: A= 6.8 μm, B= 13 μm, C= 4.8 μm, D= 15 μm, E= 21 μm, F= 14.2 μm.

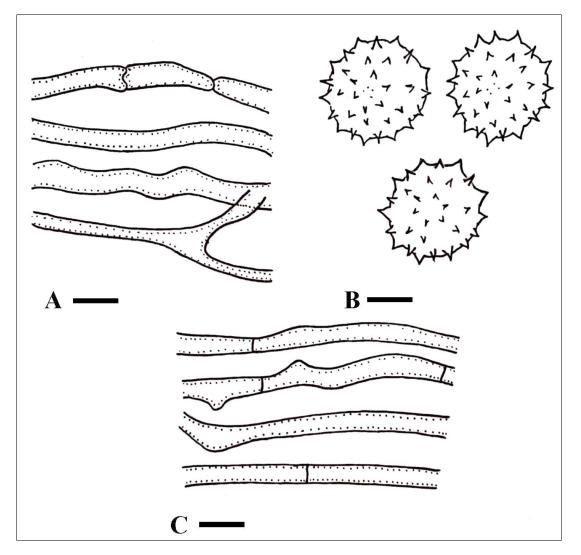


Figure 3: (A–C). Illustrations of microscopic features of *Pisolithus tinctorius* (SH-K). A. Capillitium. B. Basidiospores. C. Exoperidium. Scale bars: A= 9 μm, B= 4.8 μm, C= 8.4 μm

## 4. Discussion:

The genus *Pisolithus* was first described by Allbertini and Schweinitz in 1805. The group comprises complex of many species which cannot be differentiated by morphological characters (Singla *et al.*, 2004). This genus being worldwide in distribution and form ectomycorrhizal association with variety of woody plants (Marx *et al.*, 1975; Garbaye *et al.*, 1988; Duponnois & Ba, 1999; Singla *et al.*, 2004; Yakhlef *et al.*, 2009; Lebel *et al.*, 2018). The total number of species are up to 18 (Kirk *et al.*, 2008; Lebel *et al.*, 2018). *Pisolithus* was firstly considered as monotypic, but on the basis of morphological, basidiospores, culture characteristics, enzymes and mycorrhizal variations several species have been discovered so far (Bronchart *et al.*, 1975; Watling *et al.*, 1995; Anderson *et al.*, 1998; Singla *et al.*, 2004; Kasuya *et al.*, 2010).

The general characteristics of the genus include gasterocarp of variable shape and size with well-developed rooting system at its base (Razaq & Shahzad, 2004). *P. tinctorius* (SH-11, SH-J and SH-K) is characterized by medium to large, off-white to brown basidioma with well-developed yellow to brown rhizomorphs and often grow in clump of two with pseudo stipe having yellowish green, crowdedly ornamented basidiospores mostly growing under *Eucalyptus* tree. *P. tinctorius* found to be associated with 15 species of *Eucalyptus* (Pradhan *et al.*, 2011). The species have close resemblance morphologically with *P. albus* in term of color of basidiomata and *P. albus*, *P. arrhizus* and *P. calongei* as all of them have strong resemblance in having closely related shape of basidioma, basidiospore are 8.2-11.4 µm in diam. which is almost like *P. calongei*, *P. flavus* and *P. albus*.

From Pakistan it has been reported from Karachi (Razaq & Shahzad, 2004), Lahore and Mansehra (Yousaf *et al.*, 2013; 2014). In present study it has been reported first time from Sangla Hill, Punjab, Pakistan. Furthermore, the fungus is also known as dye ball and used to color the wool and are edible in small amount when young (Razaq & Shahzad, 2004).

### 5. Conclusion:

From the present investigation it is concluded that *Pisolithus tinctorius* is a new record for Sangla Hill, Punjab Pakistan and plains of Punjab are rich in diversity of gasteroid mushrooms. This will keep other researchers to utilize this data of taxonomy in their research work like biotechnology, ethnobotany and fungal ecology.

#### 6. References:

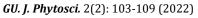
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New Record of Pisolithus tinctorius from Sangla Hill, Nankana Sahib, Punjab, Pakistan



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## Concentration of Proximate and Heavy Metals in Four Selected Vegetables Irrigated by Municipal Wastewater

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## Abstract

The study was conducted to determine the level of heavy metals such as: Lead (Pb), Cadmium (Cd), Nickel (Ni), Copper (Cu), Manganese (Mn), Zinc (Zn), Iron (Fe) and Proximate such as: Moisture, Ash and Protein in four abundantly used vegetables in Quetta, Balochistan, Pakistan. The foliage and fruits of Brinjal (*Solanum melongena* L.), Chili (*Capsicum annuum* L.), Karela (*Momordica charantia* L.) and Khira (*Cucumis sativus* L.) were collected from the city areas where these were cultivated with waste and fresh (Control) waters. The food samples were digested by dry ashing procedure and their minerals were determined by atomic absorption spectrophotometer. Results exhibited that there was slightly significant variation in elemental composition of waste and control watered vegetables. The results also specified that heavy metals in vegetables irrigated with wastewater were highly significantly build-up as compared to control water vegetables. The average range of various metals in foliage and fruits of wastewater vegetables were 1.9-2.4mg/kg and 1.8-2.2mg/kg for Pb, 0.14-0.25mg/kg and 0.13-0.20mg/kg meant for Cd, 0.73-1.01mg/kg and 0.70-0.91mg/kg intended for Ni, 0.86-1.20mg/kg and 0.80-1.13mg/kg for Cu, 0.16-0.24mg/kg and 0.11-0.20mg/kg stand for Mn, 1.26-1.60 mg/kg and 1.20 - 1.50mg/kg for Zn and 1.90-2.96mg/kg and 1.50-2.90 mg/kg for Fe, respectively. Result showed that the solitary Pb and Cd concentration in waste watered vegetables were found more than the allowable limit and the foliage exhibited more accumulation of heavy metals with respect to fruit.

Keywords: Toxic metals; Proximates; Vegetables; Wastewater; Irrigation

## 1. Introduction:

In semi-urban ecology, the municipal discarded water is frequently used for the crop cultivation, because of its informal expediency and shortage of underground water. Irrigation with wastewater is identified as a significant contributor of heavy metals to the soil. Extreme buildup of heavy metals in cultivated soils by untreated wastewater could not finish in soil impurity, on the other hand also impact on food purity and security as described by

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Muchuweti *et al.* (2006). Water and food are important for our lives; these act as a media by which we are exposed to numerous toxic metals. Mapanda *et al.* (2005) described those heavy metals are easily stored in the edible and leafy parts of vegetables, with respect to fruit or grain crops. Bahemuka & Mubofu, (1991) and Perveen *et al.* (2012) noticed that vegetables take-up heavy metals and accumulate them in their eatable and uneatable parts in great enough amounts to cause clinical difficulties among humans and animals consuming such metal-rich plants as quantified by Alam *et al.* (2003). Hussain *et al.* (2011) stated that ash, moisture and fibers concentration in vegetables and other different plant species has been deliberated energetic to the humanoid fitness and as well for the topsoil features. In Pakistan, total ecological area is about 79.6 million hectares, around 27% of the area is presently in agriculture, whereas the area of about 253000.6 hectares was for vegetables for the period of 2011-12 designated by Khokhar, (2014). The total means of water in Quetta Balochistan, (study area) are not adequate to meet up its necessities particularly of irrigation. Therefore, most of the vegetables grown in Quetta region depend upon wastewater.

The current research was focused to link the toxic metals buildup and configuration of proximate in edible parts and foliage of four frequently cultivated vegetable types using municipal wastewater and its comparison with fresh water irrigated vegetables. Cultivation of vegetables by means of contaminated water in Quetta is very commercial exercise. Intended for these fact-finding investigation seven different toxic metals such as Lead, Cadmium, Nickel, Copper, Zinc, Manganese & Iron and proximate composition in *C. annuum* L, *S. melongena* L., *M. charantia* L. and *C. sativus* L. were quantified.

## 1.1. Objectives:

- To link the toxic metals buildup and configuration of proximate in edible parts and foliage of four frequently cultivated vegetable types using municipal wastewater and its comparison with fresh water irrigated vegetables.
- To explore the fact-findings investigation of seven different toxic metals.
- To compare the toxic metals with fresh water irrigated vegetables.

## 2. Materials and Methods:

#### 2.1. Collection of Samples:

Foliage and fruit of entirely grown vegetables such as *C. annuum* L., *S. melongena* L., *M. charantia* L. and *C. sativus* L. cultivated with wastewater (along railway track near the campus University of Baluchistan Quetta) were collected. For the comparison same type of vegetables were also collected from fresh water cultivated vegetables from the Colony University of Baluchistan, Quetta heaving same environmental and soil conditions (Leghari *et al.,* 2015).

## 2.2. Pre-treatment of vegetable samples:

Collected leaves and edible part samples were carefully cleaned with purified water to eliminate any dust elements and dehydrated at room temperature for one hour and then dried at 70°C for 24 hours in oven. Dehydrated samples were crushed and sieved to obtain fine ash. All the vegetables samples were put in baggage of polythene for further analysis.

## 2.3. Analysis of Proximities:

For moisture content examination from the vegetable parts standard procedure as described by Anwar *et al.* (2011) was used. Contents of ash were assessed using method giving by Anonymous, (1977). For the investigation of protein contents in vegetable method was used as defined by Anonymous, (1985).

## 2.4. Heavy Metal Analysis:

For heavy metal analysis wet digestion technique and dry ash scheme given by Farooq *et al.* (2008) and Hseu, (2004) were used. Then concentrations of Lead, Cadmium, Nickel, Copper, Zinc, Manganese and Iron were calculated through atomic absorption spectrophotometer (Perkin Elmer Model 1100) in scientific laboratory at

Department of Botany university of Balochistan Quetta, Pakistan. For the comparisons of means of fresh and waste watered vegetables parts, paired-samples two tail t-test was used with the help of SPSS 16 computer software.

## 3. Results:

#### 3.1. Proximate Analysis of Moisture Contents:

The contents of moisture in leaves and eatable parts of waste watered vegetables extended between 73.5-90.3% and 75.4-95.6%, whereas in fresh water grown vegetables was 79.5-98.6% and 83.5-98.9% respectively. Among vegetables the *C. annuum* L. showed lowest moisture contents and *S. melongena* L. indicated. The consequences of paired-sample t-test at 95% confidence level specified that there was slightly to highly significant (0.138-0.006) variation in moisture contents between waste and freshwater irrigated vegetables as show in Tables 1.

Table 1: Summary of paired-sample t-test for the proximate composition (%) of four Vegetables

STE		C. ann	uum L.	S. melo	ngena L.	M. cha	rantia L.	C. sat	ivus L.
Parameters	Water source types	Foliage	Fruit	Foliage	Fruit	Foliage	Fruit	Foliage	Fruit
Moistur e (%)	Wastewater Control t-value Sig (2 tailed)	73.5±3.1 79.5±3.5 7.7 .017	75.4±3.0 83.5±3.0 12.8 .006	90.3±4.1 98.6±3.6 6.3 .044	95.6±3.5 98.9±4.1 2.3 .138	78.9±3.5 80.9±4.0 3.1 .115	83.5±3.0 85.2±4.0 2.3 .137	83.0±2.5 93.7±3.0 7.2 .019	85.5±3.1 95.5±3.5 8.3 .009
Ash (%)	Wastewater Control t-value Sig (2 tailed)	1.6±0.1 1.9±0.1 7.2 .019	1.4±0.0 1.8±0.1 2.0 .186	1.8±0.0 2.0±0.1 15.6 .004	1.6±0.1 1.9±0.0 11.7 .007	2.6±0.0 2.7±0.1 53.7 .000	2.3±0.1 2.4±0.0 22.5 .002	1.7±0.1 2.0±0.1 41.6 .001	1.5±0.0 1.9±0.1 39.8 .001
Protein (%)	Wastewater Control t-value Sig (2 tailed)	30.4±3.0 26.0±3.1 6.1 .026	35.7±4.5 30.5±2.5 12.6 .006	32.4±2.1 30.7±3.3 7.1 .020	38.4±4.5 35.6±3.6 4.3 .040	33.6±3.6 30.3±2.9 8.8 .024	40.2±3.0 34.4±2.0 6.2 .066	33.3±3.7 32.2±2.4 6.4 .024	37.3±3.0 35.1±3.5 4.0 .058

Sig. = Significant value, ± = Stander deviation

#### 3.2. Ash Level Content:

Ash contents in waste watered leaves was establish 1.6-2.6% and its eatable parts delimited 1.4-2.3%, while vegetables leave with fresh watered ranged 1.9-2.7% and in edible parts it was 1.8-2.4%. *C. annuum* L. showed lowest and *M. charantia* indicated largest contents of ash. Data of paired-sample t-test at 95% confidence level for Ash contents revealed highly to slightly significant (0.000-0.186) difference between waste and fresh watered vegetables in doth part (leaves and edibles).

#### 3.3. Protein Concentration:

The concentration of protein in leafy and eatable parts from wastewater irrigated vegetables was ranged 30.4-33.6% and 35.7-40.2%, whereas fresh water irrigated vegetables showed 26.0-32.2% and 30.5-35.6%, respectively (Tables 1). Data of paired-sample t-test at 95% confidence level for protein indicated that there was highly to slightly significant (0.006-0.066) variation in both part (*i.e.,* leaves and edible part) between fresh water treated vegetables.

## 3.4. Heavy Metals:

#### *3.4.1. Lead (Pb) Level:*

The concentration of pb in foliage and eatable parts of waste watered vegetables was fluctuated between 1.9-2.4mg/kg and1.8-2.2 mg/kg, while in unpolluted mature samples it was established 0.1-0.26 and0.05-0.24mg/kg, respectively. *M. charantia* L. showed highest contents of Pb in their leaves and eatable parts and *C. annuum* L. exhibited lowest. Statistical analysis using paired-sample t-test at 95% confidence level revealed that there was highly significant (0.000) variation in pb level between waste and fresh watered vegetables.

#### 3.4.2. Cadmium (Cd) Content:

Cadmium (Cd) in foliage and eatable part of waste watered vegetables ranged between 0.15-0.25mg/kg and0.13-0.20mg/kg, while in freshwater samples it was 0.06-0.09mg/kg and0.03-0.08mg/kg, respectively. Similar result was also reported in leafy and non-leafy vegetables irrigated with wastewater (Yusuf *et al.*, 2003; Al-Jassir, 2005). Highest concentration of Cd in this study was noted in *M. charantia* L. and lowest was found in *C. annuum* L. for both parts grown with *waste* and fresh water, respectively. Results of paired-sample t-test at 95% confidence level for Cd contents revealed that the difference was highly significant (0.006-0.001) between fresh and wastewater irrigated vegetables (Tables 2).

Table 2: Summary of paired-sample t-test for the variation in Heavy metals (mg/kg) composition of different vegetables irrigated with fresh and municipal wastewater

	C. annuum L.		S. melor	igena L.	M. char	M. charantia L.		C. sativus L.	
Parameters	Water source	Foliage	Fruit	Foliage	Fruit	Foliage	Fruit	Foliage	Fruit
	Waste water	1.9±0.1	1.8±0.1	2.1±0.2	2.0±0.1	2.4±0.2	2.2±0.1	2.1±0.2	2.0±0.1
Pb	Control t-value Sig (2 tailed)	0.1±0.03 75.7 .000	0.05±0.01 151.5 .000	0.23±0.04 125.6 .000	0.12±0.03 92.9 .000	0.26±0.06 632.0 .000	0.24±0.04 135.1 .000	0.17±0.06 215.1 .000	0.12±0.03 92.88 .000
	Waste water	$0.15 \pm 0.04$	0.13±0.03	0.20±0.03	$0.14 \pm 0.04$	0.25±0.05	$0.20 \pm 0.04$	$0.20 \pm 0.04$	0.16±0.04
Cd	Control t-value Sig (2 tailed)	0.05±0.02 17.32 .003	0.03±0.01 13.86 .005	0.07±0.01 22.0 .002	0.06±0.02 40.0 .001	0.09±0.02 17.76 .003	0.08±0.01 13.2 .006	0.06±0.01 43.0 .001	0.04±0.01 14.36 .005
	Waste water	0.73±0.04	0.70±0.05	1.01±0.1	0.91±0.06	0.89±0.05	0.85±0.06	0.86±0.06	0.80±0.05
Ni	Control t-value Sig (2 tailed)	0.21±0.03 77.0 .000	0.19±0.04 154.0 .000	0.36±0.06 20.55 .002	0.30±0.05 64.41 .000	0.22±0.02 35.74 0.001	0.21±0.03 36.66 0.001	0.28±0.04 39.46 0.001	0.25±0.04 95.26 0.000
	Waste water	0.86±0.07	0.80±0.05	1.26±0.06	1.13±0.06	$1.20 \pm 0.05$	$1.10 \pm 0.05$	$1.15 \pm 0.05$	$1.0 \pm 0.03$
Cu	Control t-value Sig (2 tailed)	0.20±0.05 75.59 .000	0.16±0.05 194.0 .000	0.45±0.05 105.7 .000	0.40±0.04 49.4 .000	0.33±0.02 120.57 .000	0.21±0.03 85.15 .000	0.25±0.02 113.0 .000	0.23±0.02 60.3 .000
	Waste water	0.16±0.03	0.11±0.03	0.23±0.03	0.20±0.03	0.24±0.05	0.12±0.03	0.23±0.03	0.18±0.04
Mn	Control t-value Sig (2 tailed)	0.10±0.02 16.0 .004	0.08±0.01 8.0 .015	0.15±0.03 22.0 0.002	0.12±0.04 13.86 0.005	0.20±0.03 3.46 .074	0.16±0.04 5.20 .035	0.12±0.03 34.0 .001	0.11±0.03 13.86 0.005
Zn	Waste water	1.26±0.07	1.20±0.04	1.46±0.06	1.23±0.03	1.53±0.07	1.40±0.05	1.60±0.07	1.50±0.05
	Control	0.95±0.06	0.91±0.03	$1.04 \pm 0.04$	0.97±0.05	1.13±0.05	$1.0 \pm 0.05$	$1.15 \pm 0.05$	$1.10 \pm 0.06$

Concentration of Proximate and Heavy Metals in Four Selected Vegetables Irrigated by Municipal Wastewater

	t-value	6.77	31.1	31.0	21.91	121.0	119.0	45.0	69.28
	Sig (2 tailed)	.021	.001	.001	.002	.000	.000	.000	.000
	Waste water	$1.90 \pm 0.06$	$1.50 \pm 0.05$	2.51±0.12	2.19±0.14	2.96±0.15	2.90±0.15	1.92±0.12	1.70±0.12
Fe	Control	$1.50 \pm 0.05$	$1.36 \pm 0.04$	$1.93 \pm 0.06$	$1.71 \pm 0.1$	2.70±0.12	2.46±0.13	$1.60 \pm 0.10$	$1.65 \pm 0.13$
ге	t-value	69.28	21.50	14.50	7.0	6.0	5.0	9.0	6.15
	Sig (2	.000	.002	.005	.024	.012	.015	.010	.026
	tailed)								

Sig = Significant value,  $\pm$  = Stander deviation

#### 3.4.3. Nickel (Ni) Content:

Foliage and eatable parts of waste watered samples confined grater Ni contents (0.73 - 1.01 mg/kg & 0.70 - 0.91 mg/kg) with respect to the pure watered vegetables (0.21 - 0.36 & 0.19 - 0.30 mg/kg), respectively. Among the investigated vegetables, *C. annuum* L. indicated lowest and *Solanum melongena* exhibited highest contents of Ni in both parts (leaves and edibles). Facts of paired-sample t-test at 95% confidence level for Ni contents showed that the difference was highly significant (0.001- 0.000) between fresh and wastewater irrigated vegetables (Tables 2). Present study also indicated that foliage of all the investigated vegetables contained more Ni contents as compared to the eatable parts (Tables 2).

## 3.4.4. Copper (Cu) Content:

The foliage and eatable parts showed more cu contents in the vegetables cultivated with municipal polluted water (0.86 -1.26 mg/kg & 0.80 – 1.13 mg/kg) as compared to fresh watered vegetables (0.20-0.45mg/kg & 0.16-0.40mg/kg), respectively. *S. melongena* L. exhibited highest contents of Cu in both parts (leaves & edibles) and *C. annuum* L. indicated lowest. Statistical data illustrated in Table 2 exhibited that Cu was significantly (0.000) high in waste watered vegetables as compared to fresh watered, while in all the samples it was within safe limits (73mg/kg) as defined by WHO standards (Table 3).

Table 3: Summary of heavy metals for safe limits (mg/kg) in leaves and other parts of plant.

Tuble of building of heavy h	letais for sur		gj mileuve	s and other	par to or pra			
Heavy metals	Pb	Cd	Ni	Cu	Mn	Zn	Fe	
Nontoxic limits (mg/kg)	0.30	0.10	67	73	500	100	425	

Source: World Health Organization guideline

#### 3.4.5. Manganese (Mn) Content:

The contents of Mn in Foliage and eatable parts of wastewater treated vegetables was in the ranged 0.16-0.24 mg/kg and 0.11-0.12mg/kg, while in fresh watered vegetable it was 0.10-0.20mg/kg and 0.08-0.16mg/kg, respectively. Among all the waste and fresh water treated vegetables, *M. charantia* L. exhibited highest accumulation of Mn and *C. annuum* L. showed least. Outcomes of paired-sample t-test at 95% confidence level for Mn contents revealed that the difference was highly to slightly significant (0.074-0.001) between fresh and waste watered vegetables (Tables 2).

## 3.4.6. Zinc (Zn) Content:

Foliage and eatable parts of waste watered vegetables established more Zn contents (1.26-1.60mg/kg and 1.20-1.50mg/kg) than the fresh water grown samples which ranged between 0.95-1.15mg/kg and0.91-1.10 mg/kg, respectively. Among all the examined vegetables *C. annuum* L. showed lowest contents of Zn and *C. sativus* L. highest. Statistical data specified that there was highly to slightly significant (0.000-0.021) variation between waste and fresh watered vegetables for Zn concentration in leaves and eatable (Table 2).

#### 3.4.7. Iron (Fe) Content:

The concentration of Iron (Fe) in Foliage and eatable parts was found 1.90 – 2.96mg/kg & 1.50 – 2.90mg/kg in waste watered vegetables, while the vegetables cultivated with freshwater Fe was in the ranged 1.50 - 2.70mg/kg

and 1.36 - 2.46mg/kg, respectively. *M. charantia* L. exhibited highest contents of Fe in both parts (leaves & edibles) and *C. annuum* L. indicated lowest. Consequence of paired-sample t-test at 95% confidence level for Fe contents revealed that the difference was highly to slightly significant (0.000-0.026) between fresh and waste watered vegetables (Tables 2).

## 4. Discussion:

Observations about moisture content in foliage and palatable fragments of diverse vegetables grown by polluted water reported from Rehman *et al.* (2013) was comparable by means of our results. Alike consequences were also described by Effiong *et al.* (2009) from the foliage of diverse vegetable plant species. In present study, the data illustrated in Table 1 also exhibited that the foliage exhibited less moistness as compared to eatable parts. Great vapor concentration from eatable parts in diverse vegetables was also testified by other researchers (Rehman *et al.*, 2013; Hanif *et al.*, 2006). Present results displayed fewer ash concentrations from edible parts with respect to foliage of identical vegetables. Results described from other researcher such as Hanif *et al.* (2006), Effiong *et al.* (2009) and Rehman *et al.* (2013); also back our results. Moreover, present study exposed greater contents of ash from the observation establish by Rehman *et al.* (2013) that might be for the reason of variation in ecological and soil circumstances of study sites and also due to dissimilar types of vegetables. Observations noticed by Effiong *et al.* (2009) in altered vegetables were comparable with our results for protein contents in all selected vegetables. But these contents were greater than results described by other investigators (Hanif *et al.*, 2006; Rehman *et al.*, 2013).

Higher amount of Pb was also found by other authors (Rehman et al., 2013) from polluted water cultivated vegetables. Data illustrated in Table 2 also indicated that Pb contents in all four species for waste watered vegetables were much higher than the harmless confines (0.3 mg/kg) as defined by WHO criterions and can carriage harmful properties for human health. Exceeding contents of pb was also reported by Muchuwti et al. (2006) from the harmless standards in vegetables cultivated with wastewater. They also found that regular irrigation with wastewater can result in more build-up of Pb in plants. In present study, all investigated vegetable irrigated with pure water were establish with in nontoxic boundaries and foliage showed more accumulation of Pb than their respective eatable parts. More accumulation of heavy metals in leaves than edible parts was also reported by Sawidis et al. (2001). Present study also exhibited that vegetable irrigated with fresh water showed Cd contents within harmless confines whereas in waste watered vegetables Cd were higher than the nontoxic level (0.1mg/kg) as mentioned from WHO standard in foliage and eatable parts. These finding were similar to those found in different vegetables (Rehman et al., 2013). Present observations as well exhibited that the foliage displayed greater concentration of Cd then eatable parts. Fytianos et al. (2001) reported more Cd contents in roots and leaves of vegetables as compared to other parts. They also found that foliage and roots store more Cd contents actively and they also noticed that the vegetables grown in industrial area confined more contents of Cd as compared to rural area. This study also showed more accumulation of Cu in foliage with respect to edible parts in all investigated samples moisturized in fresh and municipal wastewater. Results detected by Rehman et al. (2013) were also comparable to these consequences. They noticed that contents of Cu were grater in waste watered vegetables than freshwater samples and they also seen more Cu in foliage then eatable parts. Farooq et al. (2008) detected less then 10mg/kg Cu contents in waste watered vegetables, while Fytianos et al. (2001) observed non-significant variations of copper concentration from the vegetables cultivated in urban and rural sites. Results reported from other researchers (Fardous et al., 2010; Khan et al., 2012) are in favor our observations as they described the grater contents of Mn in waste watered vegetables as likened to pure watered grown samples. Data presented in this study showed fewer contents for Mn as compared to those reported by Ellen et al. (1990). In existing study all selected vegetables showed more Mn values in their leaves with respect to eatable parts but it was within harmless limits (500mg/kg) as designated by WHO (1996) standards (Table 3). Present results were comparable to those conveyed by Rehman et al. (2013). They indicated grater contents Zn in waste watered irrigated species than the fresh water grown vegetable. Present study indicated that all the four vegetables moisturized with pure and municipal polluted water were found within harmless limitations (100mg/kg) as described by the standards of WHO, (Table 3). Similar observation was also exhibited by Demirezen & Aksoy, (2006) in diverse vegetables. Similar results were also described by Ismail et al. (2005) for Mn, Fe, and Zn in fresh and polluted watered vegetables. This study also showed that all selected samples were found within harmless limits (425mg/kg) as defined by WHO standards (Table 3).

## 5. Conclusion:

This study concluded that heavy metals significantly accumulated in food crops due to nonstop wastewater irrigation in the study area. This study also accomplished that in waste watered vegetables the Pb and Cd were exceeding from

safe limits according to the standards defined by WHO, while other remaining toxic metals were in the allowable confines. Variation in proximate composition among varied vegetables was also noticed by this study. This variation in elements might be due to structures, integration abilities of different vegetables and environmental changes of the areas. Mostly foliage of all investigated vegetables portrayed grater contents metals than their eatable portion. Significant deduction of this research paper is that subsequently vegetables have a habit of fascinate and store toxic metals in their foliage and eatable portion, which are most consumable portion of the vegetables and also important component of human food. Therefore, it is strongly suggested all types of food crops should be avoided to grown in municipal as well as industrial polluted water.

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Concentration of Proximate and Heavy Metals in Four Selected Vegetables Irrigated by Municipal Wastewater



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## Effect of Drought and Waterlogging on Cotton crop in Dera Ghazi Khan

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## Abstract

Cotton (Gossipium hirsutum L.) is an important fiber crop. Cotton plays a key role in the economic and also in the social affairs of the world. Cotton is a soft fiber. Cotton may be exported to earn foreign exchange; or it may provide the raw material for textile production for domestic markets. Especially in developing countries cotton is a cash crop for millions of farmers worldwide. The experiment was conducted in the warehouse of Department of Botany, Ghazi University Dera Ghazi Khan Punjab, Pakistan. Dera Ghazi Khan is located at 30'03" N and 70'38" E. The overall climate of the city is dry with little rainfall. The winter is relatively cold, and the climate is hot during the remaining part of the year, but it is very hot in summer. Seed of cotton variety 555 were collected from the local market of Dera Ghazi Khan. Earthen pots were used have 12 cm in height and 10 cm in width. The experiment was conducted during summer to study the effect of drought and waterlogging on cotton. The pots were used have 12 cm in height and 10 cm width 25 pots were used for experiment. Each pot was filled loamy soil (3kg). The seeds were sown in pots at the depth of 2cm. 5 seeds of cotton were sown in each pot to achieve the best density after primitive growth 4 plants were selected to growth. Pots containing plants were arranged in a (RCBD) design in a wire house. Overall germination in this trial experiment of cotton species under the treatments of drought showed significant effects at the different levels of vegetative growth. The trends of drought and water logging effects showed that all the vegetative growth parameters studied during experiment simultaneously decrease significant folds than the control. Consequences of waterlogging impart adverse changes in different characteristics of crop plants. Some varieties however are resistant to waterlogging but susceptible varieties undergo vast range of damages. A major portion of country's economy base upon cash crops, waterlogging can decline the overall yield of crops ultimately leading to financial loss. Our study can be the used as the basic tool for analyzing the physiological, morphological and anatomical and biochemical attributes of plants under waterlogging.

Keywords: Cotton; Waterlogging; Growth; Dera Ghazi Khan; Drought conditions

## 1. Introduction:

Cotton (*Gossipium hirsutum* L.) grows around the seeds of the cotton plant. It is a shrub that is native to tropical as well as subtropical regions of the world, like Americas, India, and Africa (Zachary, 2007). Among 50 species recognized in the dicotyledonous genus *Gossipium*, belonging to Malvaceae family. In which about 45 are diploids. These are divided into three geographical groups and corresponding subgenera *viz.*, Surtia, Houzingenia

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and Gossipium. Five species are tetraploids included in one subgenus Karpas (Fryxell, 1984; Wendel & Cronn 2003; Cronn & Wendel, 2004).

Cotton is a major fiber crop and sixth largest source of vegetable oil in the world (Alishah *et al.*, 2008). It is estimated that the crop was planted on 2.5 percent of the world's arable land area. Cotton is gown in rain fed and irrigated areas of world. Pakistan is primarily an agricultural country and prosperity of people depends fundamentally upon the successful cultivation of crops like wheat, cotton, rice, maize, and sugarcane (Anonymous, 2005). Agriculture is one of the largest sectors of economy. It contributes 21% to GDP and employing 44% of the workforce in Pakistan (Government of Pakistan, 2007-08). Cotton plays a fundamental role in the agriculturally based economy of Pakistan. It is grown on an area of about 3.1 million hectares with production of 12.4 million bales in Pakistan (Anonymous, 2006-2007). Pakistan is the fourth largest cotton producing country of the world after China, India and USA (GoP, Cotton Statistical Bulletin, 2006). Cotton is commonly known as "white gold". It is grown in agricultural plains of Punjab and Sindh (Ijaz, 2009). Southern Punjab is the leading area in the production of cotton crop (so called as "Cotton Belt") and the cotton growing areas in Sindh comprising of the left bank of Indus accounts for only 19%, whereas 0.72% area lies in rest of the country (Agricultural Statistics of Pakistan, 2007-08). Cotton production is limited by different biotic and abiotic factors. Drought, or water-deficit, is a major abiotic stress that limits fiber development in cotton. It can also reduce its yield if it occurs during the reproductive phase (Selote & Chopra, 2004). Therefore, increasing crop adaptation to drought stress would be the most economical approach to improve agricultural productivity and to reduce agricultural use of freshwater resources (Xiong et al., 2006). Specifically for cotton leaves, past research has indicated that water deficit stress results in decreased water potential and osmotic adjustment (Wullschleger & Oosterhuis, 1990), lower photosynthetic rates (Pettigrew, 2004), while respiration rates have a biphasial response (Pallas et al., 1967). Leaf expansion in several species has been shown to be sensitive to water stress (Hsiao, 1973; Masle & Passioura, 1987).

Water is one of the key factors in crop production. Although water is the most abundant molecule on the earth surface, the availability of water strongly restricts terrestrial plant production (Pospisilova et al., 2000). Environmental stress limits the overlay productivity of world plant production to %50 of its potential (Boyer, 1982). Global climatic trends may accentuate this problem (LeHouerou, 1996). Adequate supply of water is a prerequisite for optimum plant growth and satisfactory yield in every crop. Water-deficit stresses occur in about 70% of arable land around the world (LeHouerou, 1996) and have been shown to have an effect on every aspect of plant growth (Kramer, 1983). In some crops observations proved that Water stress during early squaring results in pollen sterility in wheat, oats, barley, corn, and rice due to perturbations in carbohydrate metabolism (Saini & Aspinall, 1981; Saini, 1997). Water stress during flowering resulted in reductions in pollen viability, pollination and fertilization in rice, corn, beans, and chickpea due to floral abnormalities (Hsiao, 1982; O'Toole & Namuco, 1983; Westgate & Peterson, 1993). The favorable nutrition and climatic conditions (sunlight, water and temperature) normally results in excessive vegetative growth in cotton cultivars, which can enhance pests and diseases control problems and boll rot, as well as affect mechanical harvest and favor fruit abscission (Oosterhuis, 2001). Successful cotton production totally depends upon the availability of irrigation water either from canal or tube well. Irrigated agriculture is facing acute competition for low cost and high-quality water Howell (2001). Drought stress influences leaf water content, photosynthesis, and water-use efficiency (Egilla et al., 2005). In Cotton, cell expansion, division and differentiation are the first functions to be affected by water-deficit stress, followed by reductions in stomatal conductance (Ackerson et al., 1977). As a result, photosynthetic rates, plant height and leaf area are reduced while rate of squaring and node production decline resulting ultimately in yield reduction (Pettigrew, 2004). Water logging is a phenomenon that strongly influences the distribution of plant species and crop production. According to a FAO 2007 report, 20-30 million hectares of irrigated land area was affected by soil water logging as a result of poor soil drainage, intensive irrigation and highly variable weather patterns. This in turn affects crop production in many parts of the world (Setter & Waters, 2003). Water logging is a worldwide phenomenon that affects crop yield in agricultural regions. It is a major constraint to cotton production in developing countries such as India, Pakistan and China (Pang et al., 2004) and the annual agricultural production losses due to water logging in Australia are \$A180 million (Price, 1993). Flood water fills soil pores, that reducing oxygen availability and the diffusion of dissolved  $O_2$  in stagnant water is also slow that only a thin layer of soil near the surface contains oxygen (Taiz & Zeiger, 2010). In water logging situation ground water is too high that it does not allow convenient agricultural activities (Sharma & Swarup, 1988).

## 1.1. Objectives:

• To assess the effect of drought and water logging on cotton in the earthen pot experiments in the wire net house of Department of Botany, Ghazi University, Dera Ghazi Khan.

Effect of Drought and Waterlogging on Cotton crop in Dera Ghazi Khan

To measure the different parameters of waterlogging and drought conditions of cotton in Dera Ghazi Khan.

## 2. Materials and Methods:

#### 2.1. Study Area:

The experiment was conducted in the warehouse of Department of Botany, Ghazi University, Dera Ghazi Khan Punjab, Pakistan. Dera Ghazi Khan is located at 30'03" N and 70'38" E. The overall climate of the city is dry with little rainfall. The winter is relatively cold, and the climate is hot during the remaining part of the year, but it is very hot in summer. The temperature during summer is usually about 115 °F (46 °C), while during winter season the temperature is as low as 40 °F (4 °C).

## 2.2. Seed Collection:

Seed of cotton variety 555 were collected from the local market of Dera Ghazi Khan. Earthen pots were used have 12 cm in height and 10 cm in width. The experiment was conducted during summer to study the effect of drought and waterlogging on cotton.

## 2.3. Soil Preparation:

A dried, surround sieved and analysed loamy soil was collected from the field of Botanical Garden in Ghazi University Dera Ghazi Khan. The soil saturated to determine the water holding capacity of soil (Rhoades, 1982).

## 2.4. Growth Experiment:

The pots were used have 12 cm in height and 10 cm width 25 pots were used for experiment. Each pot was filled loamy soil (3 Kg). The seeds were sown in pots at the depth of 2cm. 5 seeds of cotton were sown in each pot to achieve the best density after primitive growth 4 plants were selected to growth. Pots containing plants were arranged in a (RCBD) design in a wire house.

#### 2.5. Treatments:

Plants were divided after planting into drought-treatment, waterlogging-treatment and watered-control groups. After germination of two-week treatment was started. The treatments were used in the experiments given below.

Experiment No. 1:	T0: Normal (control)	T1: One cycle drought	T2: Two cycle drought
Experiment No. 2:	T0: Normal (control)	T1: One cycle Waterlogging	T2: Two cycle Waterlogging

Each treatment has 5 replicates, and 25 pots were maintained for all treatment. Plants were thinned after two weeks of germination. Respective treatments were started after the seedling emergence. The drought-treated plants (one cycle) were not watered for 7 days. While drought-treated (two cycle) plants were not watered for 14 days. Similarly waterlogging-treatment plants of one cycle well-watered for 7 days and two cycle plants flooded for 14 days. At the end of the treatments, control, drought-treated and waterlogging-treated plants were sampled. Also, after each harvest remaining drought-treated and waterlogging-treated plants were keep in normal condition for a 10-days recovery period. This method of treatments was applied for each harvest.

#### 2.6. Growth Parameters:

The different growth parameters including Shoot Length (cm), Root Length (cm), Shoot Fresh weight (g), Shoot Dry weight (g), Root Fresh weight (g), Root Dry weight (g), No of Leaf, Leaf area (cm<sup>2</sup>), No of Boll, Boll Diameter (cm<sup>2</sup>) were studied at various stages of the experiment. However, shoot height was measured from base of the stem. Just above the soil surface. Roots were separated from shoot and measured from shoot to its apex, using centimeter rule. Root length of each pot was recorded with the help of meter rod from base to terminated leaf of plant. Shoot were separated from root and then immediately weighing using Fan electric weighing balance. Shoot were dried in an oven at 80 c° for 48 hours. The weight was then determined by using electric Fan weighing balance. Fresh root was immediately weighing using an electric weighing balance. Roots were dried in an oven at 80 c° for 48 hours. The weight was then determined by using electric weight balance in grams. After first harvesting same concentration of treatments was continued up to further 45 days. Same procedure was used to take second and third harvest. Number of bolls counted when third harvest was conducted. Boll diameter was measure by using venire caliper. Five bolls were taken from each plant and measured the diameters then their mean value was taken. Number of leaves also counted for each harvest. Leaf area was obtained by measuring the length of leaf and calculate following formula Otusanya *et al.* (2007) as shown below.

## $LA = 0.5 (L \times W)$

Where L is the length and W is the maximum width measured for each leaf on each plant

#### 2.7. Harvesting Stages:

After one month of treatment plants were harvested from each pot was carefully up rooted and washed thoroughly with water to remove soil particles on the surface of the plants parts and blotted to surface dry. The plants samples were collected for growth analysis. One plant from each treatment with 5 replicates were harvested and analyzed for various growth parameters. This treatment was continued on remaining plants for one month. After two months second harvest was taken and following studies was performed and the same procedures as like in harvest first were adopted for the measurements of growth parameters. After the second harvest, treatment was continued up to the yield harvest. All measurements of growth parameters were adopted in this harvest as like in the last, but number of boll and boll diameter were also measured.

## 2.8. Statistical Analysis:

Data collected was subjected to analysis of variance using SPSS Software Statistics 8.1. The mean values were recorded at 0.05 % probability level.

## 3. Results:

Different vegetative growth parameters regarding to drought treatments were analysed. Overall germination in this trial experiment of cotton species under the treatments of drought showed significant effects at the different levels of vegetative growth. The trends of drought effects in Table 1 showed that all the vegetative growth parameters studied during experiment simultaneously decrease significant folds than the control.

Table 1: One-way analysis of variance (unstacked) among all cotton growth parameters of drought treatments.

 Source	Degree of Freedom	Mean Square	<b>F-ration</b>	P-values	
 Factor	13	4181.14	371.56	0.0000	
Error	616	11.25			
Total	629				

#### 3.1. Effect of Drought Treatments on Growth parameters of Cotton:

The shoot growth of cotton species was noteworthy response in various treatments of drought. In the response of treatments, the shoot growth of the particular plant individual in the pots was not decrease. From the one-way analysis of variance (Table 2) showed that the P<0.5543 means the non-significant differences were found in the growth of shoot of Cotton species in connection to different levels of drought conditions.

Table 2: Effect of drought treatments on different growth parameters of Cotton crop.

Growth Parameters	Mean Square	F-ratio	P-Values
Shoot Length	60.830	0.60	0.5543
Root Length	32.8580	2.63	0.0837

Effect of Drought and Waterlogging on Cotton crop in Dera Ghazi Khan

Fresh Shoot Weight	24.3389	3.15	0.0533
Dry Shoot Weight	7.52289	4.02	0.0252
Fresh Root Weight	0.24089	1.27	0.2909
Dry Root Weight	0.09317	1.19	0.3136
Number of Leaves	2.9556	0.17	0.8401
Leaf Area	19.6551	3.57	0.0371
Number of Boll	3.35556	0.48	0.6211
Boll Diameter	0.00070	0.00	0.9992

The shoot growth of cotton species was noteworthy response in various treatments of drought. In the response of treatments, the shoot growth of the particular plant individual in the pots was not decrease. From the one-way analysis of variance showed that the P<0.5543 means the non-significant differences were found in the growth of shoot of Cotton species in connection to different levels of drought conditions. The roots of cotton species were showed insignificant response in variable treatments of drought and waterlogging. From the one-way analysis of variance (P<0.0837) showed that the insignificant differences were found in the growth of roots of cotton species into different levels of drought. Shoot fresh weight of cotton species was showed strikingly sensitive response in various treatments of drought. The obtained mass of plants of cotton was showed sensitivity in the drought condition. The one-way analysis of variances (P<0.0533) showed the significant differences were found in the fresh weight of shoot of cotton which were grown in the different levels of drought treatments. The shoot dry weight of cotton species was showed noticeably sensitive response in various treatments of drought. The obtained dry mass of plants of cotton was showed highly sensitivity in the drought condition. The one-way analysis of variance showed that the strong significant differences were found in the dry weight of shoot of cotton species which were grown in the different levels of drought treatments in the earthen pots. Root fresh weight of cotton did not show significant response against the drought treatments. From the one-way analysis of variance, the value of (P<0.2909) showed that there was no significant response of treatments on root fresh weight of cotton.

Data showed that root dry weight of cotton was not affected by treatments. The obtained dry mass of plants of cotton did not show sensitivity in the drought condition. The one-way analysis of variance showed that the insignificant differences were found in the dry weight of root of cotton which was grown in the drought-treatment. he effects of drought on number of leaves were not significant. The number and size of the leaves of cotton were not show sensitivity in the different treatments of drought. The one-way analysis of variance showed that the insignificant differences were found in the number of leaves of cotton species which were grown under the treatments of drought. During this trial experiment it was observed that the leaf lamina of cotton species was reduced due to drought treatments. Statistical study suggested the significant difference (P < 0.0371) between control plants under the drought stress. The result of one-way analysis of variance for number of bolls is given in Table 2 suggested that in cotton, number of bolls was not influenced by the effect of drought. The given results showed that insignificant differences were found between boll diameter and treatments. Data of analysis of variances describe that boll diameter was not affected under drought treatments (Table 2).

## 3.2. Effect of Waterlogging Treatments on Growth parameters of Cotton:

Overall germination in this trial experiment of cotton species under the treatments of waterlogging showed strong significant effects at the various levels of vegetative growth. The trends of waterlogging affect showed that all the vegetative growth parameters studied during experiment simultaneously decrease significant folds than the control (Table 3).

Source	Degree of Freedom	Mean Square	<b>F-ration</b>	P-values
Factor	13	3598.43	149.44	0.0000
Error	616	24.08		
Total	629			

Table 1: One-way analysis of variance (unstacked) among all cotton growth parameters of waterlogging treatments.

The shoot growth of cotton species was noteworthy response in various treatments of waterlogging. In the response of treatments, the shoot growth of the particular plant individual in the pots was significant decrease and stunted growth than the growth of control plants. From the one-way analysis of variance showed that the P<0.0009

means the strong significant differences were found in the growth of shoot of cotton species in connection to different levels of waterlogging conditions. The roots of cotton species were showed highly significant response in variable treatments of waterlogging. From the one-way analysis of variance (P<0.0001) showed that the significant differences were found in the growth of roots of cotton species into different levels of flooding. Shoot fresh weight of cotton species was showed strikingly sensitive response in various treatments of waterlogging. The obtained mass of plants of cotton was showed highly sensitivity in the waterlogging condition. The one-way analysis of variances (P<0.0050) showed the strong significant differences were found in the fresh weight of shoot of cotton which were grown in the different levels of waterlogging treatments. The shoot dry weight of cotton species was showed noticeably sensitivity in the waterlogging. The obtained dry mass of plants of cotton was showed highly sensitivity in the one-way analysis of variance (P<0.0000) showed that the strong significant differences were found in the fresh weight of cotton species was showed noticeably sensitivity in the waterlogging condition. The obtained dry mass of plants of cotton was showed highly sensitivity in the waterlogging. The obtained dry mass of plants of cotton was showed highly sensitivity in the waterlogging condition. The one-way analysis of variance (P<0.0000) showed that the strong significant differences were found in the dry weight of shoot of species which were grown in the different levels of waterlogging condition. The one-way analysis of variance (P<0.0000) showed that the strong significant differences were found in the dry weight of shoot of species which were grown in the different levels of waterlogging treatments in the earthen pots (Table 4).

<b>Growth Parameters</b>	Mean Square	F-ratio	P-Values
Shoot Length	995.928	8.28	0.0009
Root Length	184.165	12.45	0.0001
Fresh Shoot Weight	121.758	6.02	0.0050
Dry Shoot Weight	141.955	20.78	0.0000
Fresh Root Weight	35.5882	15.14	0.0000
Dry Root Weight	14.9609	18.26	0.0000
Number of Leaves	286.422	8.28	0.0009
Leaf Area	323.170	13.12	0.0000
Number of Boll	48.4667	3.06	0.0572
Boll Diameter	18.4747	3.17	0.0523

Table 4: Effect of waterlogging treatments on different growth parameters of Cotton crop.

Root fresh weight of cotton showed significant response against the waterlogging treatments. From the oneway analysis of variance, the value of (P<0.0000) showed that there was also highly significant response of treatments on root fresh weight of cotton. Data showed that root dry weight of cotton was affected by treatments. The obtained dry mass of plants of cotton shows sensitivity in the waterlogging condition. The one-way analysis of variance showed that the significant differences were found in the dry weight of root of cotton which was grown in the waterlogging treatments. The effect of waterlogging on number of leaves was significant. The number and size of the leaves of cotton sp. showed sensitivity in the different treatments of waterlogging. The one-way analysis of variance showed that the significant differences were found in the number of leaves of cotton species which were grown under the treatments of waterlogging. During this trial experiment it was observed that the leaf lamina of cotton species was reduced due to waterlogging treatments. Statistical study described the significant difference (P < 0.0000) between control plants under the waterlogging stress. The results of one-way analysis of variance for number of bolls are given in Table 4 suggested that in cotton, number of bolls was influenced by the effect of waterlogging. The statistical data (p<0.0523) showed that boll diameter of cotton also effected under the different treatments of waterlogging (Table 4).

#### 4. Discussion:

A few features of this experiment were noteworthy. The 4-5 days required for plants in the first flooding period to begin responding was much longer than had been expected based on controlled environment work. These results were unexpected given that atmospheric evaporative demand was quite high (average daily pan evaporation was 9.8 ram). When the visible plant response occurred, it did so with a distinct break about 2 m up from position 4. At this position, the water table was 0.32 m below the soil surface with 45% of the root system above the water table. With capillary rise, a larger portion of the root system should have been exposed to nearly saturated conditions. Another feature of the response was the lack of uniformity in the plants at any particular position. Thus, on day 8 only a few plants were severely wilted, most others appeared to be wilted while others seemed to be unaffected. The exact cause of this effect was not determined, and we can only surmise that it was due to individual plant variability caused by both genetic and phenotypic differences and other differences associated with special variability within the facility. The other noteworthy feature was the non-repeatability of the two flooding events. Presumably some acclimatization and conditioning occurred following the first flooding which better enabled the plants to cope with the second flooding event. Observed changes in root distribution may be involved. During the first flooding period, the decrease in growth of leaves was not consistently associated with decreased potential.

Our observations suggest that the initial wilt symptoms were not caused by poor plant water status as measured with the pressure chamber and that only when plants were severely wilted did osmotic and water potential decrease (on day 8 of the first flooding event). Partial closure of stomata on and after day 5 of the first flooding event was sufficient to cause an increase in foliage temperature (Reicosky et al., 1985). Thus, the relationship between foliage temperature and potential, (Idso *et al.*, 1982) may not operate when increases in foliage temperature are caused by waterlogging and drought stress. For the second flooding event the reduction in leaf growth again began about 2 days after flooding and was apparently not mediated by decreased potential. The reason for the reduced growth may be associated with the rapidly declining nutrient levels where petiole N, in particular, reached deficiency levels at the end of each flooding period. The interaction between N deficiency and water deficit stress physiology has been studied in the drought situation (Radin & Parker, 1979) and some of their findings are relevant here. They concluded that stomata of N-deficient plants were more sensitive to stress. The coincidence in time of decreased leaf growth and low petiole nitrate levels suggests that the interacting control mechanisms discussed by Radin & Parker (1979) were also operative where plant stress was induced as the result of waterlogging and drought stress. Consideration of the above data suggests that the primary response of cotton plants subjected to waterlogging and drought stresses in the field is not immediately mediated through effects on plant water status. This is in contrast to the general conclusion reached by Cannell (1977) and may reflect differences in the response of different species to waterlogging and drought stress. If water uptake is mainly a passive process (i.e., not requiring 02) then this will continue despite impairment of the active uptake functions of the root. However, this impairment of active functions will very likely affect nutrient uptake and root hormone production more readily than water uptake. Thus, the present results are consistent with the premise that waterlogging, and drought stresses primarily cause a direct reduction in active {energy requiring} root processes such as nutrient uptake resulting in decreased leaf growth and a delayed effect on plant water status. Even though the plant measurements indicated that some stress occurred (mainly during the first flooding event) its subsequent effect on plant yield appears to have been beneficial. This response in cotton is not unknown (Hearn, 1980) although there is some confusion about the extent of the effect. Those experiments which do report yield increases as the result of drought stress indicate that the stress occurred prior to flowering. In the present experiment, the first flooding event with measurable imposed plant stress was prior to the main flowering period which subsequently occurred when water was non-limiting and additional nitrogen fertilizer was supplied.

## 5. Conclusion:

The basic concept of using a sloping plot to impose a variable water table depth was successful and provided a range of soil water and oxygen levels where cotton plants had various portions of their root system under waterlogged conditions. Two flood events in the growing season showed similar soil water and oxygen profiles with the plant response more dramatic in the first flooding. Soil oxygen partial pressures decreased to low values within two days of the water-table being imposed, but none showed complete oxygen depletion. About 55% of the root system needed to be under the water-table before visual effects were observed in the plant canopy. Unacclimatized plants in the first flooding exhibited a reduction in leaf growth rates and eventually exhibited decreased leaf water potentials and visible wilt symptoms. The first flood event altered the root distribution to such an extent that plant water status in the second flooding was not affected. The observed stress and decreased leaf growth in both flood events appeared to be associated with a decrease in N uptake during flooding. The interactions of nutrient stress and other stresses experienced by the plant were not clear. Seed cotton yield was largest where the intermittent water table during the two flood events was between 0.1 and 0.3 m below the soil surface. Should these stress effects occur prior to the major flowering period small reductions in vegetative growth may result in the promotion of reproductive growth. These results demonstrate plant tenacity and the ability to acclimatize to waterlogging events to the extent that subsequent waterlogging exposure of previously waterlogged plants may have little detrimental effect. The variable water table depth facility enables study of plants subjected to a continuum of conditions ranging from complete inundation to a complete absence of a water-table within the root zone and may prove useful as an aid to screening plant genotypes for their tolerance to rapidly imposed waterlogging events. Consequences of waterlogging and drought stresses impart adverse changes in different characteristics of crop plants. Some varieties however are resistant to waterlogging but susceptible varieties undergo vast range of damages. A major portion of country's economy base upon cash crops, waterlogging and drought stresses both can decline the overall yield of crops ultimately leading to financial loss. Our study can be the used as the basic tool for analyzing the physiological, morphological and anatomical and biochemical attributes of plants under waterlogging. Cotton do not grows well in waterlogged soils because it does not exhibit some constitutive internal features which make it resistant to waterlogging as like wheat, sorghum, maize, cotton and other plants do not show efficient growth under

waterlogging; On the other hand Rice grows well in waterlogged soils because it exhibit some constitutive internal features which make it resistant to waterlogging, moreover, some other resistant varieties have ability to grow in such a condition, because they develop certain type of modifications which help them to adapt waterlogging and drought stresses conditions.

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Effect of Drought and Waterlogging on Cotton crop in Dera Ghazi Khan

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## Effect of Coal Fly Ash on Seedling Growth, Productivity, Biochemical and Heavy Metals Uptake of *Zea mays* L. and Soil Characteristics in Semi-arid and Climatic Conditions of Quetta Valley

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## Abstract

In this study, the chemical composition of Fly Ash (FA) released from coal based thermal power plants, was characterized its macro and micronutrients to improve the chemical and physical properties of soil. With this concept, a field experiment was conducted to find out the efficiency of Fly Ash (FA) for the growth of *Zea mays* L. Six various fly ash treatments (0%, 20%, 40%, 60%, 80%, 100% w/w) were mixed with soil in pots and *Z. mays* seeds were sown. Different growth parameters and photosynthetic activity of plant were examined. Fly Ash (FA) treated with soil caused significant improvement in soil quality and germination percentage of *Z. mays* seeds. The plant growth, chlorophyll, and yield content enhanced with 0% to 40% fly ash (FA), being optimal at 60%. From 80% onwards, the examined parameters tended to reduce. Hence, fly ash (FA) can be utilized as a substrate or as a soil improving material for the growth of plants, leading to the sustainable utilization of solid waste material.

Keywords: Pot Culture Experiment; Fly Ash (FA); Growth; Yield Parameters; Nutrients

## 1. Introduction:

Fly ash (FA) is a resultant of waste produced from the combustion of coal in Brick kiln and thermal power plants. In Pakistan and India millions of tons of fly ash is generated annually and projections also show that the production including both fly ash and bottom ash and being increasing day by day (Pandey *et al.*, 2009; Pandey & Singh, 2010). A huge amount of fly ash is creating environmental problems due to improper utilization or disposal. The management of this huge amount of solid waste, at both regional and global level is a prime concern for the present and coming future (Ahmaruzzaman, 2010; Kishore *et al.*, 2010). The elemental composition (both nutrient and toxic elements) varies due to types and sources of used coal (Camberato*et al.*, 1997). Combustion of bituminous or lignite coal for power and heat production is associated with release of byproducts such as ash, bottom ash, boiler

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slag, etc., which in general are referred to as "coal combustion byproducts" (Asokan *et al.*, 2005). Fly ash is sandy spherical amorphous iron-aluminum minerals. Physical, chemical and mineralogical characteristics of fly ash depend on the quality of raw materials, combustion method, and the power plant performance efficiency (Bhisham *et al.*, 2013).

The accumulation of fly ash is a global environmental problem, since the ash land is also covering a huge territory, the ash is difficult to transport because of its airborne nature; light particles from ash deposits are carried away by the wind contaminating nearby reservoirs and lands. The ash harms the health of the local population due to the penetration of the particles into the organism through the respiratory system (Lee *et al.*, 2006). The world has gained huge experience in the use of fly ash. FA consists of plant macronutrients Na, K, P and Fe and micro-nutrients Co, B, Zn, Cu and Mn. The elements Pb, Ni, Cr, Cd and a few more also occur abundantly and have the potential to cause contamination/toxicity (Fytianos *et al.*, 2001). Further, plant micro-nutrients at high concentrations can cause toxicity (Miller *et al.*, 2000). Fly ash use in agriculture is mainly based on its limiting potential and supply of nutrients which promote growth of plants and alleviate the condition of nutrient deficiency in soils (Singh *et al.*, 2008, Pandey *et al.*, 2009, Singh & Agrawal, 2010). Fly ash can be used as a liming agent not only in mono but also in di-cotyledons plants for better crop yields (Ahmed *et al.*, 1986; Sarangi & Mishra, 1998; Singh & Siddiqui, 2003). Several researchers have already proposed that low dose of fly ash amendments improves the physicochemical properties of the soil such as pH, texture, water holding capacity, electrical conductivity etc. (Sajwan *et al.*, 2003; Siddique, 2004; Lee *et al.*, 2006; Kishore *et al.*, 2010).

Enriching soil with fly ash increased the plants growth and yield of vegetables oils and cereal crops like tomato, potato, cabbage, pea, wheat, mustard, oats and sunflower (Mittra *et al.*, 2005; Saxena *et al.*, 2005). (Khandakar *et al.*, 1990) reported high yield of rice, soya bean and black gram in fly ash amended soil. (Rajiv & Prakash, 2012) reported that black gram grown on fly ash amended soils resulted in a considerable increase in carotenoid content of leaves though chlorophyll a and b content were not greatly affected. (Pandey *et al.*, 1994) reported that sunflower plants treated with fly ash exhibited improved growth. High concentrations of metals (loid) and organic pollutants in fly ash wastes are released into soil, air, and water presenting a global threat to the surrounding environment and human health. Harsh conditions prevailing on fly ash are unfavorable mechanical composition and pH, high concentrations of Soluble salts, lack of nitrogen and phosphorous, reduced number of microorganisms and fungus, toxic concentrations of As, Au, Ag, B, Cu, Cd, Cr, Hg, Mn, Mo, Ni, Pb, Zn, and the presence of PAHs and PCBs. There is no report on growth, yield or leaf metabolism and heavy metal uptake in maize plant due to FA amendment.

#### 1.1. Objectives:

- To study the impact of varied levels of fly ash amendments (FAA) on the growth and yield
- To examine the grain/ seed quality of high yielding, bio-chemical and its soil characteristics

## 2. Materials and Methods:

#### 2.1. Physio-chemical Properties of Fly Ash (FA):

Fly Ash (FA) in an unweather condition (sample-lots less than 30 days old) obtained from Brick kiln industries of Quetta, Balochistan-Pakistan. Before the use of fly ash in experiment it was analyzed for its sand, silt, clay composition, electrical conductivity (EC), moisture content, organic matter (OM) and PH characteristic by using hydro meter and pH meter calibrated with standard buffer solutions. The chemical composition (%) of fly ash including Cl, OM, NO<sub>3</sub>, PO<sub>4</sub>, Na, K, Ca and its trace elements (mg/kg) such as Cd, Cu, Fe, Mn, Mg, Ni, Pb, Zn were analyzed with an absorption spectrophotometer (Model AA1475 at IMMT, Bhubaneswar).

## 2.2. Experimental Field Design:

Experimental fields were prepared on randomized block design with 3 replications for each fly ash amendments (FAA). Fields were prepared by ploughing to a depth of 1ft and the cultivation was done by following standard agronomic practices. Fly ash was uniformly mixed in soils as 0%, 20%, 40%, 60%, 80% and 100% (w/w) and designated as T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub> and T<sub>6</sub> respectively. After complete dryness, the soil was homogeneously mixed with previously air-dried fly ash as per selected ratio. For 20% FAA, 40 kg of fly ash were mixed with 160 kg of air-dried soil, for 40% FAA, 80 kg FA were mixed with 120 kg of soil and for 60% FAA, 120 kg of FA were mixed with 60 kg of soil. For 80% FAA, 160 kg of fly ash were mixed with 40 Kg soil sample. On average, the weight of total soil for

the experiment plot of 1m<sup>2</sup>area was found to be 200 kg. The fly ash mixed soil was then properly poured back into the respective experimental plots. Seeds of maize were collected from Quetta Agricultural Centre, Balochistan and used as test sample in the present experiment.

#### 2.3. Growth Response and Chlorophyll Estimation:

The pH values show that soils of Dera Ghazi Khan District are slightly alkaline, with no significant difference between one site and another as the zone and site main factors were not significant in overall analysis of variance. The pH is a negative logarithm of Hydrogen ion concentration. The soil of Dera Ghazi Khan was neutral to slightly alkaline from upland area to low land area. The differential differences among the sites were responsible for the occurrence of significant interaction between sites and zones in the overall analysis.

Chlorophyll a = (12.7 × A663 – 2.69 × A645) × V/1000× W Chlorophyll b = (22.9 × A645 – 4.68 × A663) × V/1000× W Total Chlorophyll = (8.02 × A663 + 20.21 × A645) × V/ 1000 × W

Estimation of carotenoid was done using the formula of (Kirk and Allen, 1965) and expressed in absorbance units as Acar 480 per gram fresh weight.

Acar/ 480 per leaf segment = A480+ 0.114.A663 -0.638 × A645) V/ 1000 × W

Where A480, A645 and A663 are the absorbance of the acetone extract at 480 nm, 645 nm and 663 nm, respectively. **V=** Volume of acetone extract, **W=** Weight of leaf (gram fresh weight)

## 2.4. Analysis of Heavy Metals:

For heavy metal analysis, 1g of oven dried and grinded seeds were digested with tri-acid mixture (HNO<sub>3</sub>:H<sub>2</sub>SO<sub>4</sub>: HClO<sub>4</sub> in the ratio 5:1:1) till transparent color appeared (Allen, 1974). The digested samples were then filtered through Whatman no. 42 filter paper and the volume was maintained up to 50 ml in volumetric flask deionized water. The concentrations of different heavy metals i.e., Zn, Pb, Cd, Cr etc. were determined using Atomic Absorption Spectrophotometer (AAS). For evaluating the transport behaviors of assessed heavy metals in seeds of maize and Bio-Concentration Factors (BCF) were calculated by using the following formula.

$$BCF = \frac{Concentration of Heavy Metal in the Plant Parts}{Concentration of Heavy Metal in the Initial Soil}$$

## 3. Results:

#### 3.1. Physio-chemical Properties:

During study, various parameters of soil and fly ash were examined and represented in table 1. The pH of soil was alkaline-acidic 7.8 while in case of fly ash noted about 7.1. Similarly, electrical conductivity of soil 1.24 ds/cm which was higher than fly ash 1.18ds/cm and moisture contents was ranged from 9.5 to 9.7 respectively. The soil was loamy sand in which the sand was 84.6%, followed by clay 14.05%, silt 1.35%, and chloride of soil examined 98% correspondingly. In addition, in soil the organic matters were noted higher than fly ash 0.8% and 0.3%. The nitrate content of normal field soil was 0.004%, while it was below detection limit in fly ash 0.0015%. Similarly, higher values of sodium and potassium were detected in soil 3ppm, 143ppm as compared to fly ash 2ppm, 36ppm, whereas the phosphate was detected higher in fly ash 0.12ppm than soil 0.1ppm, and in soil calcium was noted 0.210 while in case of fly ash observed 0.435, respectively (Table 1).

S. No.	Parameters	Soil	Fly Ash (FA)
1	pH (Alkaline-acidic)	7.8	7.1
2	Electrical conductivity (EC) ds/cm	1.24	1.18
3	Moisture contents	9.5	9.7
4	Sand %	84.6	-
5	Silt %	1.35	-
6	Clay %	14.05	-
7	Texture class	Loamy sand	-
8	Chloride (%) (meq/L)	98	-
9	Organic Matters (OM) (%)	0.8	0.3
10	Nitrate (N03-N) %	0.004	0.0015
11	Phosphate (ppm)	0.1	0.12
12	Na (Exchangeable) ppm	3	2
13	K (Exchangeable) ppm	143	36
14	Ca	0.210	0.435

Table 1: Physio-chemical properties of soil and fly ash used for experiment

#### 3.2. Element Contents:

The elements composition of soil and fly ash (FA) are provided in Table 2. As it can clearly be seen from the table, the element contents of soil and fly ash particles were mainly composed of (Cd, Cu, Fe, Mn, Mg, Ni, Pb, Zn). The concentration of elements ordered from the highest to lowest as Fe, Mn, Mg, Zn, Cu, Ni, Pb, and Cd in fly ash, however there were a significance difference between element contents of soil and fly ash. For instance, fly ash has higher concentration of Mn, Mg, Ni, Cu, and Zn, whereas soil has higher concentration of Fe. The amount of Cd and Pb in the fly ash higher than soil.

S. No.	Parameters	Soil	Fly Ash (FA)	
1	Cd	0.002	0.007	
2	Cu	0.013536019	0.058220017	
3	Fe	0.963	0.742	
4	Mn	0.3511535	0.687818	
5	Mg	0.124	0.406	
6	Ni	0.005331	0.017098	
7	Pb	0.002	0.010	
8	Zn	0.042	0.052	

3.3. Shoot Height:

The shoot height of *Z. mays* showed an increasing trend at 0% fly ash (FA). In case of fly ash effect the shoot, height was increasing at 20% fly ash. Effect of fly ash amendment on shoot height was observed maximum in  $T_2$  as recorded on 10, 15, 20, 25, and 30 days in Table 3. The highest shoot height was examining in  $T_2$  26.4, followed by  $T_3$  24.5,  $T_5$  22.5,  $T_4$  22.5, and  $T_6$  17.8 at 30 days.

Table 3: Effect of different concentration of coal ash fly % (w/w) on growth rate (Shoot height (cm)) of maize seedling.

Treatments –	Days After Sowing							
Treatments –	5 Days	10 Days	15 Days	20 Days	25 Days	30 Days	LSD 0.05	
T <sub>1</sub> 0%	0	6.2	11.4	17.3	23.6	29.3	NS	
T <sub>2</sub> 20%	0	6.1	12.5	16.2	21.1	26.4	NS	
T <sub>3</sub> 40%	0	5.0	9.3	14.1	19.6	24.5	NS	
T <sub>4</sub> 60%	0	0	5.3	10.0	15.3	19.2	NS	
T <sub>5</sub> 80%	0	4.7	10.0	10.8	18.9	22.5	NS	
T6 100%	0	4.2	7.1	10.5	14.4	17.8	NS	
<i>p</i> ≤0.05	***	***	***	***	***	***		

## 3.4. Germination Percentage:

Germination is a prime plant growth process; the effect of various concentration of fly ash blended with soil on the germination of *Z. mays* as presented in Table 4. Germination of *Z. mays* starts after 5 days of sowing in all the treatments, it might be due to hard coat of seed. The maximum germination percentage 68% was recorded at 80% FA (T<sub>5</sub>), followed by  $T_6$  (65%),  $T_2$  (62%),  $T_1$  (57),  $T_4$  (37%), while least was noted in  $T_3$  (25%) at 20, 25, and 30 days respectively.

Table 4: Germination Percentage and survival of *Z. mays* seedling under the influence of different percentages (w/w) of coal fly ash.

Treatmonte	Germination (%)	Harvest Day After Germination of Seedlings						
Treatments	Germination (%)	Day 5	Day 10	Day 15	Day 20	Day 25	Day 30	P <u>&lt;</u> 0.05
T1 0%	57	0	1	4	5	5	5	NS
T2 20%	62	0	3	4	5	5	5	NS
T3 40%	25	0	1	2	2	2	2	NS
T4 60%	37	0	0	2	3	4	3	NS
T <sub>5</sub> 80%	68	0	1	3	6	7	7	NS
T <sub>6</sub> 100%	65	0	3	5	5	5	5	NS
p≤0.05/ LSD0.05	***	***	***	***	***	***	***	

3.5. Number and Area of Leaves:

Effect of fly ash amendment was studied with respect to number of leaves and area of leaves. From result it was showed that the number of leaves per plant at 20 DAS, 25DAS, and 30 DAS were higher as compared to other days after sowing (DAS). The maximum number of leaves per plant was recorded at  $T_1$  followed by  $T_2$ ,  $T_3$ ,  $T_5$ ,  $T_6$ , and the minimum was noted in  $T_4$  respectively in Table 5.

Table 5: Effects of different ratio of coal fly ash % age (w/w) No of leaves of Z. mays.

	Coal Fly Ash Percentage on Leaves Days After Sowing						
Treatments	5 days	10 days	15 days	20 days	25 days	30 days	_ p≤0.05/ LSD 0.05
T1 0%	0	2	4	6	7	7	NS
T2 20%	0	1	3	5	6	7	NS
T3 40%	0	1	2	4	5	6	NS
T4 60%	0	0	1	3	4	4	NS
T5 80%	0	2	2	4	4	5	NS
T <sub>6</sub> 100%	0	1	2	3	3	4	NS
p≤0.05	***	***	***	***	***	***	

Similarly, the maximum area of leaves was observed at 30 DAS as compared to remaining DAS in Table 6.

Table 6: Effects of different ratio of coal fly ash % age (W/W) on leaves area of Z. mays.

Treatmonte	Coal F	p≤0.05/ LSD					
Treatments	5 days	10 days	15 days	20 days	25 days	30 days	0.05
$T_1 0\%$	0	45	95	125	209	267	NS
T <sub>2</sub> 20%	0	38	105	137	197	254	NS
T3 40%	0	24	116	145	203	287	NS
T4 60%	0	20	126	138	199	264	NS
T5 80%	0	37	85	109	184	247	NS
T6100%	0	0	36	54	97	105	NS
p≤0.05	***	***	***	***	***	***	

The highest leaf area was recorded  $T_3$ , which was followed by  $T_4$ ,  $T_1$ ,  $T_2$ ,  $T_5$ , and lowest leaf area was recorded in  $T_6$  (fly ash) in table6. In addition, after three replication the average area of leaves in which the highest leaf area showed by soil control 151.15 cm<sup>2</sup>, while the lowest leaf area exhibited by control fly ash 4.16 cm<sup>2</sup> in Table 7.

Effect of Coal Fly Ash on Seedling Growth Productivity, Biochemical and Heavy Metals Uptake of Zea mays

Treatment (%)	<b>Replication 1</b>	<b>Replication 2</b>	<b>Replication 3</b>	Average leaves area cm <sup>2</sup>
Control soil	153.05	151.26	149.14	151.15
Control coal	3.63	4.62	4.24	4.16
20%	121.44	95.62	114.31	110.31
40%	113.59	123.32	99.91	112.27
60%	105.72	94.16	92.42	97.43
80%	78.04	84.29	76.66	79.66

Table 7: Effects of different ratio of coal fly ash on leaves area of Z. mays.

#### 3.6. Heavy Metals Content:

Accumulation of metals in plants vary from species to species and also within the various parts of a plant. The application of fly ash alters positively the elements concentration in *Z. mays* plants, however it enhanced elements numerically as compared to control. The accumulation of various metals such as Cu, Fe, Mn, Ni, Zn in fly ash treated plots were relatively higher as enhanced the percentage of fly ash, as a result the accumulation content of studied elements was higher at 60%, 80%, and 100% (fly ash) over control respectively in Table 8. Similarly, the result of the present study indicated the application of fly ash significantly increased the content of Cd and Pb at 60%, 80% and 100% (fly ash) over control.

Table 8: Heavy metals ( $\mu$ g/g) uptakes by *Z. mays* at different coal fly ash application ratio/percentage.

Elements	Control (0%)	20%	40%	60%	80%	100%	<i>P</i> ≤0.05/LSD <sub>0.05</sub>
Cd	BDL	BDL	0.0017	-0.0015	0.0172	0.00706	NS
Cu	0.0021	0.0042	0.0073	0.0211	0.0236	0.034	NS
Fe	0.2320	0.1765	0.1752	2.6189	2.25292	2.61248	NS
Mn	0.0091	0.0261	0.0631	0.1351	0.3140	0.672	NS
Mg	0.1470	0.9725	0.1583	0.2100	0.5421	0.617	NS
Ni	0.0034	0.0037	0.0062	0.0232	0.9542	0.132	NS
Pb	BDL	BDL	0.0919	0.09197	0.16862	0.3065	NS
Zn	0.0192	0.038	0.0434	0.0721	0.0780	0.084	NS

Where BDL: Below Detectable Level/ Limit

#### 3.7. Chlorophyll and Carotenoid Content:

Chlorophyl and carotenoids are the most essential biochemicals content which are used as the capability of the plant growth. The content of Chl. a, Chl. b, total chlorophyl and carotenoids were enhanced little by the application of fly ash. The effect of fly ash on the photosynthetic pigments of *Z. mays* leaves at 15 DAS is presented in Table 9. At 15 DAS Chl a content of detached leaves showed an increased value in the range of 0.61 to 0.76 mg g<sup>-1</sup>. The maximum Chl. a value content was observed at 100% (T<sub>6</sub>), while the minimum was examined at 80% (T<sub>5</sub>). Chl. b contents increased progressively from 1.02 to 1.20 mg g<sup>-1</sup>. The highest Chl. b content was recorded at 20% (T<sub>2</sub>), whereas the lowest was reported in 40% (T<sub>3</sub>). In addition, the total chlorophyl content was ranged from 1.71 to 1.9 mg g<sup>-1</sup>, the maximum was indicated by 100% (T<sub>6</sub>), while the minimum was showed by 40% (T<sub>3</sub>). Similarly, the carotenoids value ranged from 1.07 to 1.41 mg g<sup>-1</sup>, the highest noted at 20% (T<sub>2</sub>), whereas the least was noted at 100% (T<sub>6</sub>) respectively in Table 9. In our study, the enhancement of fly ash does not affect the levels of pigments Chl. a, Chl. b, total chlorophyl, and carotenoids, however over all the most effected one was the 20% (T<sub>2</sub>) which little increased the pigments contents of *Z. mays* leaves.

Table 9: Effect of coal fly ash % age (w/w) on Chl. a, Chl. b, total chl. and carotenoids contents of leaves of Z. mays.

Treatmonte	15 Days After Sowing						
Treatments -	Chl. a	Chl. b	Total chl.	Carotenoids			
T1 0%	0.70	1.10	1.8	1.29			
T <sub>2</sub> 20%	0.68	1.20	1.88	1.41			
T3 40%	0.69	1.02	1.71	1.21			
T4 60%	0.73	1.14	1.87	1.26			
T <sub>5</sub> 80%	0.61	1.20	1.81	1.30			

125

 T <sub>6</sub> 100%	0.76	1.14	1.9	1.07
p≤0.05	***	***	***	***

#### 3.8. Yield Parameters:

Effect of fly ash (FA) amendment on yield of *Z. mays* such as diameter of cone, length of cone, no of seeds, and weight of 1000 seeds of various treatments are presented in Table 10. The highest diameter of cone 12.6cm, highest length of cone 11.8cm, highest no of seeds 132, and highest weight of 1000 seeds 152gm was recorded in 0% ( $T_1$ ). However, at 20% ( $T_2$ ), showed secondly dominated value as compared to other treatments of fly ash (FA) in Table 10.

Table 10: Effects of coal fly ash % age (W/W) on yield parameters of Z. mays	Table 10: Effects of coal fly	v ash % age (W/V	V) on vield pa	rameters of Z. mays.
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Treatments	Diameter of Cone (cm)	Length of Cone (cm)	No of Seeds/ Cone	Weight of 1000 Seeds (gm)
T1 0%	12.6	11.8	132	151
T <sub>2</sub> 20%	11.9	11.0	117	133
T <sub>3</sub> 40%	10.6	8.8	111	123
T4 60%	9.1	9.3	107	117
T5 80%	11.0	11.5	45	84
T <sub>6</sub> 100%	0	0	0	0
p≤0.05	***	***	***	***

#### 4. Discussion:

Fly ash, when added to soil, was generally beneficial for the growth of Z. mays from control to 40%, above which it had deleterious effects. In nature the coal fly ash is generally alkaline and Gray in colour. From the results it was noted that the pH of soil was alkaline-acidic 7.8 while in case of fly ash noted about 7.1 and electrical conductivity of soil 1.24 ds/cm which was higher than fly ash 1.18ds/cm. Similar observation from coal fly as was also reported by (Mahale et al., 2012). Addition of fly ash to the soil will increase the conductivity of soil. (Hodgson et al., 1982) reported that the alkaline pH of fly ash may be due to the presence of Ca, Na, and Mg, along with other trace metals. CaO is a major constituent of the fly ash that forms Ca  $(OH)_2$  with water and thus attributes to alkalinity. Acidic soil can be neutralized by the addition of fly ash because it contains hydroxide and carbonate salts, which have the ability to neutralize the soil (Pathan et al., 2003). According to (Khan & Khan, 1996) the increase in soil pH might be due to the neutralization of H<sup>+</sup> by alkali salts and also due to solubilization of basic metallic oxides of fly ash in soil. Similar result was reported by (Kalara et al., 2003) in trends of higher pH, EC and WHC in fly ash (FA) soil at their experiments with maize, wheat, mustard, and mung beans. The present results regarding moisture content, organic matter (OM), chloride, nitrate, phosphate, Na, K and Ca in soil and coal fly ash were examined respectively. The fly ash has less organic matter (0.3%) compared to soil (0.8%). All the metals present in soil are also present in the fly ash. The nitrate, sodium, and potassium were found higher amount in soil but the content phosphate, and calcium were found higher amount in fly ash than soil. Fly ash contains all the important metals needed for plant growth and metabolism. Observations reported by (Mahale et al., 2012) are similar to our results. The availability of plant nutrients in fly ash amended soil samples were positively various from the control. In addition, fly ash has higher concentration of Mn, Mg, Ni, Cu, and Zn, whereas soil has higher concentration of Fe. The better growth and yield were apparently due to presence of utilizable plant nutrients viz., Fe, Mn, Mg, Zn, Cu, Ca, P, Ketc. in fly ash. The nutrients from fly ash have been reported previously as being beneficial to plants through soil application or foliar dusting (Mishra & Shukla, 1986). The amount of Cd and Pb in the fly ash higher than soil. The physicochemical properties and the heavy metal contents of fly ash depend on the parent coal composition from which it is produced and, on its coal, combustion conditions (Dhadse et al., 2008). It is difficult to generalize the composition of ashes due to the varying nature of coal. (Gatima et al., 2005) observed that the coal fly ash contains approximately 95-99% oxides of Si, Al, Fe, and Ca, and about 0.5 to 3.5% of Na, P, K, and S. (Bhanarkar et al., 2008) reported that the fly ash consists of minute glass-like particles of 0.01 to 100 mm size.

In the present study, no visible injury symptoms were observed in any of the treatments during the growth and development, however with enhanced the amounts of fly ash, there is gradual retardation in growth parameters such as shoot height, germination percentage, number of leaves and leaves area. Soil blended with 80%, and 100% fly ash (FA) display a negative effect, however it shows better growth when soil amended with 20%, and control during experiment. The retarded plant growth, as reflected by reduce in shoot height, germination percentage,

Effect of Coal Fly Ash on Seedling Growth Productivity, Biochemical and Heavy Metals Uptake of Zea mays

number of leaves and leaves area, may be coupled with undesirable chemical properties of the FA (Mishra and Shukla, 1986, Wong and Wong, 1989). Other works had also displayed that the high alkalinity influence the availability of nutrients, mainly phosphorus, which affects the plant growth adversely (Jala & Goyal, 2006).

Accumulation of metals in plants vary from species to species and also within the various parts of a plant. The application of fly ash alters positively the elements concentration in *Z. mays* plants, however it enhanced elements numerically as compared to control. The accumulation of various metals such as Cu, Fe, Mn, Ni, Zn in fly ash treated plots were relatively higher as enhanced the percentage of fly ash, as a result the accumulation content of studied elements was higher at 60%, 80%, and 100% (fly ash) over control respectively. (Brun *et al.*, 2001) observed that accumulation of Cu in maize was same in acidic and calcareous soils, while the accumulation of Zn was quite reverse. Similarly, the result of the present study indicated the application of fly ash significantly increased the content of Cd and Pb at 60%, 80% and 100% (fly ash) over control. Similar results of significant increase in uptake of nutrients were observed in okra (Yeledhalli & Ravi, 2008), Sudan grass and oats (Srivastava & Chhonkar, 2000). However, (Rautaray *et al.*, 2003) reported less concentration of Cd and Ni in rice grain but, high Cd content in residual mustard. Similar results were reported by (Singh *et al.*, 2008). Accumulation of Cd decreased over years and the values were within the permissible limit (Jena, 2013). Several studies conducted so far showed that long term use of fly ash in agricultural fields can modify the soil health as well as heavy metal uptake too and also influence the growth, crop quality and physiology (Pandey & Sing, 2010).

Chlorophyl and carotenoids are the most essential biochemicals content which are used as the capability of the plant growth. In present study, at 15 DAS Chl. a, Chl. b, total chlorophyl, and carotenoids content of detached leaves showed decrease value with increasing concentration of flay ash (FA) as compared to that of control. (Krupa & Baszynski, 1995) reported that the accumulation of heavy metals leads to inhibition of chlorophyll formation.

Srivastava & Chhonkar, (2000) described that fly ash play an important role in mobilization of nutrients and there by leading to better availability of nutrients facilitating uptake by plants resulting in better growth and dry matter production. The effect of fly ash (FA) amendment on yield of *Z. mays* from our study concluded that highest diameter of cone, length of cone, no of seeds, and weight of 1000 seeds were exhibited when plants treated with control, and 20% application of fly ash (FA) respectively. This result showed the findings of (Niaz *et al.*, 2008) on *Eclipta albe*. The current study displayed that the fly ash could be beneficial in improving the soil quality and plant yield, but the most suitable treatment for improved the *Z. mays* yield control to 20% (T<sub>2</sub>) fly ash with soil as its maximum crop yield.

Fly ash application in soil improved the growth of *Z. mays* up to certain treatments, and after that the concentration of fly ash caused deleterious effects on the growth of plant. In current study, control to 40% fly ash levels proved to be optimally useful for the growth of plant. The observed responses of the plants are also supported by other workers, like; (Pathan *et al.*, 2003) on *Cynodon dactylon*; (Bharia *et al.*, 2000) on green gram; (Hisamuddin & Singh, 2007) on *Pisum sativum*; (Parveen *et al.*, 2006) on *Mentha citrate*. Such results showed that the fly ash concentration for better growth of plant varied from plant to plant. High levels of fly ash have caused toxic effects on the growth of plants. (Rai *et al.*, 2000; Kumar *et al.*, 2001) reported that the fly ash used as fertilizer to improve the fertility of soil, especially for barren lands.

#### 5. Conclusion:

Thus, fly ash was beneficial for the cultivation of *Z. mays* L. Its application at 20% to 40% can enhanced the growth and yield of *Z. mays*. It is also important to examine the possible accumulation of toxic substances (present in fly ash) in soil and especially in the edible parts of the plants. However, the fly ash varied widely in its physical and chemical composition, therefore the mode of use in agriculture is different and defends on the characteristics of soil or soil type. The findings of such studies may help in planning future strategies for the utilization of fly ash.

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Effect of Coal Fly Ash on Seedling Growth Productivity, Biochemical and Heavy Metals Uptake of Zea mays

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# Analytical Study of the Historical Contribution of Muslims in Horticulture During 800-1250 AD

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## Abstract

In Islamic history, horticulture had a prominent place in the Muslim era. The Arabs planted gardens in Medina and Taif. In this sense, gardens were an important source of food and trade. Leaving Arabia, when Islam reached Persia and Syria and Egypt and Andalusia, Muslim kings and rulers planted gardens. They not only encouraged farmers but also took personal interest. The Arabs planted fruit trees abundantly in mountainous and desert areas. And when the canals were dug, fruit trees were planted along their banks. Palm groves were plentiful in the Hijaz and later planted in Iraq and Syria. During the Umayyad period, the fruit trade spread throughout Arabia. Similarly, when Muslims arrived in Andalusia, gardening was a common industry. From Abdur Rehman to the end, the rulers were fond of horticulture, thanks to which the cultivation of new plants was promoted, and Muslim botanists played a vital role in the field of horticulture.

Keywords: Arabs; Muslims; History; Horticulture; Contribution; Syria; Andalusia

## 1. Introduction:

It is a common thing in modern times that the scientific revolution in Europe has led to an economic revolution in the world. Europeans not only forgot the role of Muslim scientists, but also turned a blind eye to the role played by Muslims in a bright past. With the exception of a few Western Orientalists, most played a partisan role. But history is not for anyone to forget everything. The pages of history bear witness to the enlightened role of Muslims in any sphere of life. In the field of science, where he laid the foundations of science, agriculture, irrigation and horticulture remained an important subject of his attention. The Muslims paid special attention to horticulture and laid the foundation for the cultivation of many new fruit trees, such as the cultivation of olives, which was an important achievement of the Muslims (Hitti, 1937). Olive produced largely in Andalusia, and it was produced in large quantity. People of Andalusia not only benefited from this fruit, but the entire Europe benefited from this through the efforts of the Muslims of Andalusia. Orchards were planted everywhere in Andalusia and many fruit nuts or pens were imported from various foreign countries, such as pomegranates from Syria (Maqri, 1921)

Literature is very important component of historical research. Literature on this topic is mostly fragment and it is not easy to touch all relevant data regarding this topic. But there is an attempt to consult the most relevant

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literature i.e., History of Muslims in Spain, History of Arabs, Moors in Spain, Tareekh-e-Islam, Nafah-ul-Teeb, Tamadun Arab, Khilafaat Andalusia i.e., are remarkable.

#### 1.1. Objectives:

- To explains the role of Muslims in the field of horticulture
- To strengthen this field with their research efforts and experiences
- To explore the analytical attempt at the efforts of Muslims in the field of horticulture

## 2. Materials and Methods/ Methodology:

This is an analytical study and basically the research is related to the historical perspective and the data is based on qualitative approach. Therefore, historic method of research has been used in this research with the help of documentary sources based on primary and secondary sources i.e., books, articles and the other available historical sources related to this research.

#### 3. Discussion:

Even in the early days of Islam, there was a special focus on gardens and gardening was an excellent source of income. Even before the advent of Islam, Medina and Zaif were famous for their gardens. There were many orchards of date palms and vineyards. The cultivation and protection of orchards was also ensured during the spread of Islam and the rule of the world. During the conquest of Khyber, the destruction of orchards was strictly forbidden, and caution was given not to damage fruit trees. Similarly, when the armies left, the Rightly Guided Caliphs always advised them not to destroy fruit trees and fields (Qureshi, 1993).

During the Prophet's time, many of the Companions not only had gardens but also traded in dates. During the conquest of Iran and Syria, the Companions engaged in farming and gardening in the conquered areas. But in the Umayyad period, not only did horticulture flourish on a regular basis, but production began on a large scale by improving the irrigation system. In the time of Banu Abbas, the time of Harun al-Rashid was the golden age of this series when there was an abundance of gardens. Promoted gardening from Baghdad to the borders of the Islamic Empire (Solat, 1997).

While other disciplines flourished with the establishment of the House of Wisdom in the time of Mamun, the interest of medical experts in plants highlighted the medical importance of plants. The beginnings of development in horticulture during the Abbasid period were plundered by the Mongols but re-ignited by the Muslims in Spain. Abd al-Rahman made Andalusia the head of the gardens. He invited all kinds of plants from all over the world and planted them in Andalusia. Similarly, from the time of the rule until the fall of Granada, unfortunately, Andalusia was the centre of horticulture (Akbar, 1986).

Islam promoted tree as the Quran is the source of various sciences and explains all the sciences. It also mentions the sciences which we call science and botanical science is one of them. Allah Says in the Holy Quran "It is Allah Who sends down for you rain from the sky which you drink, from which you make the trees grow and graze your animals. Indeed, these are signs for a people who reflect" (Al-Quran, 27: 59-60). In the Holy Quran, Allah Almighty has made different tastes of trees and shrubs as His sign. Some are double-stemmed, and some are single-stemmed. They are all irrigated with the same water, and we give some of them superiority over others in taste. Surely in all these there are signs for those who use their intellect. In another place, Allah Almighty has declared the trees as one of His blessings. He said: Then We caused to grow in it gardens of abundant water. It was not in your power to make their trees grow. Is there another god with Allah? no! Rather, they have turned away from the path" (Al-Quran 13: 04). Hazrat Muhammad (PBUH) has declared tree planting as a charity for the believers in order to promote tree planting. He said: Charity will be counted by him (Sahih Bukhari). He gave so much importance to tree planting that he ordered to continue this process till the Day of Resurrection. The Prophet (PBUH) said: He may not stand without planting. In his teachings, not only are there rules for planting trees, but there are also clear rules for protecting trees by planting them.

Whoever plants a tree, then protects and watches over it until the tree bears fruit, it will be a cause of charity with Allah for him. Unnecessary cutting of trees, especially fruit trees, has been declared immoral and disgusting. It is narrated on the authority of 'Abdullah ibn Habash that the Prophet (peace and blessings of Allaah be upon him)

Whoever cuts down a berry tree, Allah will turn his head upside down in Hell.

# (Iqbal, 2010) شى البي مَتْكَاتِيْبَمَ عن عقر الشجر، فانه عصمة للدواب في الجدب- .

Acacia or berry tree is common in Arabia. The Prophet (peace and blessings of Allaah be upon him) said about the berry tree: Whoever cuts down the berry tree, Allaah will cast him face down in Hell (Sayuti, 1972). Hazrat Salman appeared in the service of the Persian Messenger of Allah and told me that there is a barren land of Judaism. The Holy Prophet (peace and blessings of Allaah be upon him) ordered the Companions to go and lose 360 hours. The Prophet (peace and blessings of Allaah be upon him) planted palms with his blessed hands and the Companions gave him water. When he planted the last palm tree and looked behind him, all the palm trees were young and had gone too high to give dates. He was surprised to see this; he became a Muslim and he also freed Hazrat Salman Farsi. The palm groves shown in this picture are in the hands of the Holy Prophet. Famous companion of the Holy Prophet "Sabit ibn al-Dahdah ibn Naeem ibn Ghanim ibn Ayas." He is one of the Companions. He had two palm groves, out of which 600 palm trees in the big garden, "Thabit bin Al-Dahdah bin Naeem bin Ghanim bin Ayas". Therefore, he is called "Blue Ansari". It is well known that he was a Companion. You had two palm groves, the largest of which had 600 palm trees. The tree is a great blessing of Allah Almighty, with which there is companionship even in this world and the very identity of Paradise is with the gardens and trees. There are also real needs. The life of beasts is due to forests and trees. Ever since deforestation began, these beasts have become extinct, while trees are also a source of human oxygen, shade, fruit and cooling. According to researchers, trees help humans breathe as tree provides oxygen.

After the Prophet era, Muslims had a credit to promote the discipline of horticulture from their early period at Arabia and they shifted this field into the entire Islamic world from Arabia to its surrounding areas. The grapes and dates were the favorite fruits. Due to poverty Arabs concentrated on dates even during the war days. In the modern world it is assumed that Muslims contributed to Islamic theology, and they have no share in other discipline of life. But on the pages of history, it is preserved that Muslims were the patronage and custodian of agriculture and this field horticulture (Ibn-e-Khaldon, 1985)

Agriculture also flourished in the early days of the Abbasi Caliphate. Although it remained in the hands of the ancient inhabitants. But with the attention of the caliphs, it improved. The barren fields and barren villages were reclaimed and rehabilitated. The government focused on the Euphrates Valley, the most fertile area in the entire empire. The old Euphrates canals were re-dug, or new canals were built. The area of Khurasan was also fertile. It was one of the highest paying provinces.

Ports such as Baghdad, Basra, Cairo and Alexandria became centers of maritime trade. From the time of Mansoor, Muslim traders started going to China. Similarly, to the west, they traveled to Morocco and Andalusia. They carried dates, sugar, cotton woolen garments, steel tools and glassware from the Arabian Peninsula and imported spices, camphor, silk and ivory, ebony from the Far East. At night the old canals were re-dug, or new canals were built. The area of Khorasan was also fertile. It was one of the most tributary provinces. According to Arab geographers, the outskirts of Bukhara had become a real garden. Flowers were also widely grown for commercial purposes. In Damascus, Shiraz, Firozabad and Persia, etc., the perfume industry of rose, lotus and violet were developing. During the Abbasid period, the empire expanded, and civilization flourished. As a result, trade flourished. Ports became centers of maritime trade. Traders used to go to China on one side and Andalusia on the other side with the products of the Arab regions and return to the Arab regions with their goods there. Similarly, various industries also developed. Factories for carpets, curtains, floors, apparel, jewelry, paper, soap and glass were established in various cities. Minerals were extracted from different parts of the country. Attention was also paid to agriculture. Barren lands were made cultivable. Canals were dug for irrigation. A variety of fruits, flowers and salads were grown.

The Muslims developed this sector in all the areas where there was scope for agriculture and planted more trees in large numbers. One of the reasons for attachment to trees was the command of the Prophet. After arranging irrigation, special attention was paid to the cultivation of olives, dates, fruits, oranges, lemons, figs, almonds, pomegranates, bananas, apples, walnuts, oak, peaches, peaches, wheat and gram in the Islamic Empire along with cereals. It was the work of Muslims in the world. Muslims also planted many fragrant plants including sunflower, clove, sandalwood, Zaffron (Poole, 1886). The Muslims allowed the cultivable lands in their conquered areas to be cultivated by their former owners and also built roads and bridges for their convenience. In addition, they reduced the amount of revenue and gave them the freedom to trade. As a result, as production increased, people became more prosperous, and more and more people began to be attracted to this sector (Nasir, 1984). According to Thatcher, Muslim botanists studied the best time to sow seeds after learning about their movements in plants. They also improved the seeds through transplanting. They knew eight ways to transplant. In all the big cities and towns there were agricultural madrassas where the information obtained was passed on to the farmers. Biologists and

botanists were aware of the harmful effects of sun exposure on small plants and how to protect fruits from harmful pests. He continued about Muslims in these words "Muslims did a great job in gardening. They knew how to transplant, and they knew how to grow new varieties of flowers and fruits. He began to cultivate as many trees and plants in the East as he did in the West and wrote scientific pamphlets on agriculture." For a long time, the olive oil industry continued to flourish here (Oliver, 1900).

Ameer Ali Syed with the reference of Hitti, "In the field of natural history especially botany, pure and applied, as in that of astronomy and mathematics, the western Muslims (of Spain) enriched the world by their research. They made accurate observations on the sexual difference of various plants." According to Professor Arnold, the process of Muslims from all over the world traveling to Mecca and Medina for Hajj and pilgrimage has made significant progress in biological science. Al-Ghafiki and Al-Idreesi traveled from Andalusia (Spain) to Africa, collecting information on hundreds of plants and compiling books. Ibn al-Awwam compiled a book containing the properties and conditions of 585 plants and put botany on the path of development. On the subject of botany, Al-Dinuri wrote a six-volume "Kitab-e-Nabat" which is considered to be the first comprehensive and comprehensive Encyclopedia Botanica in the scientific world. This book was written at a time when people were ignorant of knowledge and Arabic translation of Greek books had not even begun (Ali, 1922).

The ruler of Islamic Spain, Abd al-Rahman I, founded an agricultural research institute in Cordrdoba called the "Hadiqa Botany of Medicine", which provided opportunities to establish not only botany but also medicine. There were also research doors in medical sciences. Thus, Andalusian botanists have rightly discovered the existence of sex differences in plants. In this discovery, where he was assisted by the empirical research done in the "Hadith of Botanical Medicine", the command of Allah, the Lord of Glory, "All things have been created in pairs" (Boun, 1976).

The study of botany in Andalusia was started by the Muslims in their early days. Research on medicinal plants was an integral part of the development of medicine. So, Abd al-Rahman I set up an agricultural research farm in Cordoba called Hadiqa Botanical Medicine, where physicians and botanists had the opportunity to study the properties of plants, their breeding and their effects. Abd al-Rahman I took special interest in the patronage of botany and imported seeds and cuttings of plants and trees that were not available in Andalusia from distant lands. He therefore sent official delegations not only to the continent of Africa but also to most of the Asian countries to help in the search and production of rare plants, trees and herbs. Abdullah bin Abdul Aziz Al-Bakri compiled the properties of the trees and plants of Andalusia under the name of 'Kitab Ayan Al-Nabat wa Al-Shajariyat Al-Andalusiya'. Ibn al-Rumiyah, a botanist, traveled to most of Africa and Asia, except Andalusia, and studied the plants and herbs found during that time from a purely botanical point of view (Hitti, 1937).

Abd al-Rahman, who was fond of gardening, He planted strange trees and plants which were not present in Andalusia from outside in his vast garden. He used to spend a lot of his time in this garden. It is said, when Abdul Rahman formed a strong government in Andalusia, he planted an air garden west of Cordoba. And named Risafah, he adorned the garden with wonderful plants with the help of experts. Plants from many countries were sought for this garden. From Syria and Iraq, plant beautiful and delicious fruit plants, seeds and nuts of different plants. Geographers were also called in to help. This garden was taken care of very carefully. Experts look at the condition of each plant in the garden on a daily basis. The result was that in a few years these plants converted into trees and began to produce excellent crops, after which Cordoba had abundant fruit (Ibn-e-Aseer, 1986). Abdur Rehman's sister Umm Asbagh used to send him fruits from Syria as gifts. While she sent some pomegranate was flowing, Abdul Rehman planted a pomegranate.

#### 4. Conclusion:

Islam is the religion of nature and the source of human welfare. It highlights every positive aspect of life that contributes to the well-being of humanity. Trees are an important steppingstone to human employment and development. Islam declared tree planting as charity. From the time of Prophethood, horticulture started to flourish and then it continued to grow gradually. Muslim botanists played a key role in horticulture during the Abbasid period, focusing on the growth of trees. And as the seasons began to be planted, geographers also suggested suitable areas in this regard. The sources of irrigation began to play a role. Then came the era of Abdul Rahman and the horticulture industry flourished. In the history of Andalusia, the Muslims paid the most attention to gardening, which resulted in Andalusia gaining a prominent place in the world for gardens. Summarizing this discussion, we can say that Muslims, like other sciences, played a central role in bringing horticulture to its peak. Ignoring their historical role would make the history of horticulture incomplete.

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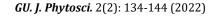
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# A Systematic Review on Synthesis of Nanoparticles and Their Applications

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## Abstract

Nanobiotechnology indicates the manufacturing and application of resources on a nanometric scale by convention. The maximum size of nanoparticles up to 100 nanometers has been indicated in the literature. Nanoparticles are used in various fields of life including medical to many subdivisions of industrial production like solar power and oxide fuel cells for energy storage. The nanoparticles are also widely used in the Sterilization, biological classification, and treatment of some cancers. Due to their unique properties, anti-corrosion activity, high oxidation resistance, and high thermal conductivity, nanoparticles have proven very significant in the last decade. Nanoparticles can be synthesized by chemical and biological methods. There are different types of metallic nanoparticles that have major industrial applications including gold, silver, alloys, and magnetism. The main objectives of this review were to describe nanoparticles, with specific reference to their mechanisms and types of biosynthesis. The literature review showed that various combinations of plants and nanomaterials were utilized to reduce the polluted environment.

Keywords: Nanoparticles; Elements; Sterilization; Oxidation resistance; Magnetism

## 1. Introduction:

Nanotechnology is a broad field in the 21st century and has consumed a huge effect on the economy, industry, and way of life of the world's population (Scott *et al.*, 2018) and these particles solved the somatic, biochemical, and organic properties of substances measured at the nanometric scale (1-100 nm) and their impact on human interests (Zhao *et al.*, 2017). As reported by USEPA (The United States Environmental Protection Agency), nanomaterials are elements with a minimum particle size of approximately 1-100 nm. It can control and produce substances of such size, thereby promoting the progress of advanced and unique properties. They can be used to solve numerous common technologies and problems. China and other developing countries have made quick improvements in the design of agrochemicals based on nanobiotechnology, and its uses in this field are expected to be realized in the upcoming 510 years. However, its achievement is influenced by numerous aspects, including the

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market mandate for yield limitations, eco-friendly welfares, threat valuation, and other organization strategies in the context of economics and technology (Roshitha et al., 2019).

Nanobiotechnology denotes the term used to manufacture, manipulate, represent, and apply structures by controlling the nature, and size of the nanoscale (Matinise et al., 2017). The field of nanobiotechnology is the maximum energetic research field in materials science, and nanoparticle (NP) synthesis is an important alternative worldwide. Considering specific faces, viz. size (1-100 nm), nature and structure, nanoparticles show new or improved characteristics (Chen et al., 15). Nanoparticles can be summarized in inorganic and organic NPs. Inorganic NPs include semiconductor nanoparticles (such as Zinc Oxide, Zinc sulfide, Cadmium sulfide), metal Nanoparticles (like Gold, Silver, Copper, Aluminum), and magnetic NPs (such as cobalt, iron, nickel), whereas organic NPs include carbon NPs (Such as buckminsterfullerene, artificial atoms, carbon nanotubes).

## 1.1. Objectives:

- To assess the synthesis of the nanoparticle by chemical method and green synthesis method.
- To examine the characterization of Nanoparticles
- To study the applications of foliar nanoparticle spray as a possible remedy or to study plant response to various toxic chemicals.
- To find the impact of nanoparticles on plants and humans

#### 2. Review of Relevant Literature:

#### 2.1. Nano Biotechnology:

Many technologies have been increased agricultural yield to developed and reduce the environment associated with farming production. This technology has the potential to protect water and land resources with equal inputs, or the desire to increase production and ultimately protect the environment. However, it will be very important to support them, because they may not reap commercial gains and may also cause the gap between developed and developing countries to widen. Therefore, their common and moral impact must be considered. However, what must be considered is the efficiency in certain areas, although these cannot solve the present issues related to food production and dispense in the world. Therefore, when considering the ability to use these new technologies, developing countries have been actively engaged in the research and development of these technologies (Ditta et al., 2015). Nanobiotechnology is the branch of science that uses nanoscale materials, ranging from 1 nanometer to 1,000 nanometers (Stern et al., 2008). The range of biomolecules is the maximum size of these NPs, like antibodies, membrane receptors, and enzymes (Faraji et al., 2009). Nanobiotechnology has a comprehensive range of uses and is related to biomedical sciences, materials sciences, and scientific sciences. Nanoparticle research is an important part of nanoscience and nanobiotechnology.

#### 2.2. The Absorption and Uptake of Nanoparticle (NPs) by Plants:

The absorption of NPs by plants is affected through numerous factors, which are associated with the properties of the nanoparticles themselves, as well as the physiological functions of plants and the interaction between nanomaterials and nanoparticles. The properties of nanoparticles will greatly influence their behaviours, so plants can absorb them. Size appears to be one of the major limitations in penetrating plant tissue, and it has been reported about the maximum size at which plants permit NPs (Nanoparticles) to move and store in the cells, generally 4050 nm as the size exclusion limit. (Latef et al., 2018). In addition, the type of NPs and their chemical composition are other factors that affect absorption (López et al., 2008). In many cases, morphology has also been shown to be the deciding factors (Bar et al., 2009). Surface and coating functionalization of nanomaterials can vary considerably the absorption and accumulation characteristics of plants (Yedurkar et al., 2017).

Shin et al. (2019) reported by the physiological functions in plant species may be different, and this difference will lead to changes in the absorption of nanoparticles. These works show how the crop species that belong to different plant families and are showing to magnetic nanoparticles of CO, TiO, or gold exhibit different patterns of absorption and accumulation by plants. But the applications are also important in determining how plants can efficiently take up nanomaterials. The roots exclusively absorb nutrients and water, while the leaves develop for gas exchange and have a dense cuticle (Nazligul *et al.*, 2020). They interact with the other components of the environment, and when they are assimilated by plants, they will affect their characteristics. For example, the

135

occurrence of organic matter like humic acid in the soil can improve the stability of the nanomaterials, thus increasing the bioavailability of the nanomaterials, while salt ions can produce precipitates and cause the opposite effect (Wang *et al., 2016*). In addition, the occurrence of organisms like bacteria and fungi will affect the absorption of NPs by plants, mostly because these microorganisms have established a symbiotic relationship with plants, like mycorrhizal fungi (Matussin *et al., 2020*).

### 2.3. NPs Movements in Plants:

Once NPs penetrate plants, they can penetrate tissues in two ways: apoplasts and sympathetic nerves. The cell membrane is transported to the outside of the plasma membrane that penetrates the extracellular space, in the cell walls of adjacent cells, and the xylem vasculature (Chandra et al., 2015) while sympathetic nerve transportation contains the water movement and matter among the cytoplasm. The attached cells pass through special structures named cytoplasmic nodules (Jahan et al., 2018) and the sieve tubes. Apoplasts pathway is significant for the circular change in plant tissues and permits NPs to influence in the central cylinder of roots and tissues of vascular to greater upward change in the aerial roots (Liu et al., 2019). Confidential after the essential chamber, the nanoparticles can move through the xylem along with moisture to the above-ground part (Ishak et al., 2019). However, reaching the xylem through the root resources junction the barrier of the apoplasts pathway, the Casparia zone, which necessarily follows a simple pathway through endoderm cells (Yan et al., 2015). Some nanomaterials can stop and accumulate in the Caspian Sea (Rehana et al., 2017). Another important symbiotic transport is possible, using screening factors in the phloem, and allowing delivery to non-photosynthetic tissues and organs (Matussin et al., 2020). In foliar applications, NPs must overcome the wall created by the stratum corneum by lipophilic or hydrophilic pathways (Vickers et al., 2017). Lipophilicity involves the diffusion of wax through the stratum corneum, while the hydrophilic pathway is achieved through the polar water pores in the epidermis and stomata (Urban et al., 2021). Since the diameter of the epidermal pores is estimated to be about 2 nm (Sarkar et al., 2020), the stomatal pathway seems to be the maximum likelihood path for NPs penetration, and the exclusion of size is limited to more than 10 nm (Kalita et al., 2016). The pathways of NPs movements through plants are important because they can indicate which portions of the plant can be reached and anywhere, they can close and store. For example, if one type of nanoparticle is transported primarily through the xylem moderately than the phloem, it is likely that they primarily travel from the base to the shoots and leaves moderately than down, so they should be applied to the roots to achieve good distribution in the factory. On the other hand, if the nanoparticles have good mobility through the phloem, they should be applied by foliar spray. Most agricultural products (like fresh vegetables, fruits, meat, eggs, milk and dairy products, various food processes, nutritional foods & medicines) are unpreserved. Nanotechnology study helps maintain cleanness, quality, and protection.

#### 2.4. Green Nanobiotechnology:

For the sustainable development of all regions of the world, it is imperative to find cheap, secure, and renewable sources of energy. Green nanotechnology is developed to obtain flexible and well-organized energy in solar cell form and has extended a target of steamy countries. However, the application of crystal photovoltaic plates is unrefined and overpriced. Most industrialized countries are focusing on the enlargement of photovoltaic plates, storing energy, and other nanotechnologies to improve thermal energy-saving methods. Commercial viability is a key feature in the development of these energetic and photocatalytic materials. By properly approaching this element, we will be able to develop further concepts. Jennings & Cliffel in Vanderbilt University has made sustainable technological breakthroughs, exploring the application of photosynthetic units of protein derivative from leafy plants to directly save solar energy for electricity (Ciesielski et al., 2010), for approximately 1 year. The maximum expensive element of the structure is the microscope plate at the bottom of the cell. Harnessing solar energy determination be a great success in the service of humanity, and it may continue and intensify in the years to come. Nanotechnology can also be used to preserve biomass in oils, biochemical intermediates, compounds and specialty foodstuffs (as well as catalysts) to reduce manufacturing costs while maintaining efficiency and economy. These nanostructured compounds have a large area of surface per unit volume and can also precisely control the arrangement, structural function, and further significant properties of the catalyst (Davis et al., 2008). The significant improvements in the rate of seeds germination by the penetration of nanomaterials into the seeds have been reported in the literature. The impacts of NPs on the plants can be resulted positive or negative (Monica *et al.*, 2009). The amount of plant toxicity may be depending on the type of nanomaterial and its potential applications. Suppose that silica nanoparticles labeled with fluorescent isothiocyanate (FTIC) and light strong cadmium selenide (Cdse) of quantum dots have been verified as biomarkers for their ability to promote the germination seed. It has been found that silica nanoparticles labeled with FTIC could induce germination of rice seeds, while quantum dots stopped germinating (Nair *et al.*, 2011).

#### 2.5. Plant Protection and Production:

Nanoparticles have been increased the dispersion of pesticides and moisture (i.e. reduce organic loss of solvents) and harmful migration of pesticides (Bergeson et al., 2010a). Nanoparticles, as well as biomaterials, show beneficial quality, like hardness, absorbency, crystal, required for the training nanoparticles (Bordes et al., 2009). Nanomaterial furthermore provides a large of area-specific increases for objective (Khatami *et al.*, 2018). Emulsion, nano-capsulation, and nano-packaging are some of the newly discussed nanoparticle delivery techniques (Bergeson et al., 2010b). The stability of the expression is an important aspect of the nanoscale. (Liu et al., 2019) The use of polymeric balance, like acrylic acid, butyl acrylic acid, PVP (polyvinyl pyrrolidone), and PVOH (polyvinyl alcohol) produces biocides bacteria Stable nano-bacteria. Rapid nano palletization techniques are used to prepare biposiline particles at 60 to 200 nm. By using this type of technology for commercial purposes, it is necessary to consider the stability of the polymers over time. Although it is inadequate in agriculture (Anjali et al., 2010), it has been reported that it stabilized with a larval of natural cleansing surfactants by the extract of natural plants. Further studies of this class are required to investigate the application of natural stabilized nano-biocide conceptualization to the protection of agricultural crops. Another field of avant-garde research may involve the development of nanomaterials that can be used protective layer for a passive release of traditional pesticides and fertilizers (Corradini *et al.*, 2010). Suppose that, examined the possibility of using NPs of highly biodegradable antimicrobial matter to make the NPK (Nitrogen, Phosphorous, Potassium) fertilizer released slowly (Yan et al., 2005) develop Kaolin Nanitizer, which can be used as an adhesive and as a coating to control the discharge of fertilizers. Many of these studies are summarized (Ghosh et al., 2020). First, the Nano clay material provides an interaction surface of a high aspect ratio to encapsulate "pesticides", such as "fertilizer, growth promoter of plants and pesticides" (Ghosh et al., 2020).

#### 2.6. Excess Insecticide Detection:

The Food and Drug Administration (FDA) has been describing 1,045 chemical residues from insecticide excess (FDA, 2005). NPs-based on the application of nano sensors can detect pesticide residues (Khatami *et al.*, 2018; Hondred *et al.*, 2019). Such as in situ sample collection, laboratory solid-phase extraction, sample analysis, and comparison of the obtained spectral peaks with reference values for pesticide residue determination. Currently, the United States Department of Agriculture (USDA) implements a GC/LCMS-based one or more component method for residue assessment of "Organic phosphate, OCPs (organochlorines pesticides), carbamate insecticides, triazines, triazoles, pyrethroids, new nicotines and strobilurins (USD, 2010). The PDA (Pesticide Data Program) investigates more than ten thousand samples per year to control the Insecticide excess is related to the production of agriculture (USDAPDA, 2008). In addition, (Van *et al.*, 2011) have studied the biosensors of enzyme-based for the residue's detection of organo-chlorines, organo-phosphates, and carbamate. The amount of detecting material applications of copper, mercury, Ti, Platinum & nano structured Pb dioxide was used to stop the enzyme in a detection substrate and improve the sensitivity of a sensor.

Several problems are also indicated in literature for using the nanomaterial-based detection such as:

- ✓ Availability of sensitive nanomaterials in the residues of pesticides at normal depth.
- Simplicity of instrumentation and sensor technologies
- Trace level detection required consistency and repeatability,
- ✓ In addition, the large number of pesticides used in the production of agriculture, the applicability based on nanomaterials sensors in the detection of residue pesticides may limited (Van *et al.*, 2011; Duan *et al.*, 2016).
- Randomized trials cannot be performed with all pesticides in a sample (Van *et al.*, 2011; Duan *et al.*, 2016), reported that it is necessary to develop technology and to selectively and stably detect nanomaterials for incorporation of chemical compounds (amino acid, enzymes) among nanomaterials. In a first step, regulators could apply nano sensors for the detection of important residues which are extremely harmful to human health. Smart nanomaterials can also be used as an alternative to pesticide detection sensors, smart nanomaterials and nano pesticide (Bergeson *et al.*, 2010b) as pesticide supplies and indicator sensors can eliminate the essential for sensors to detection of pesticide residues in the soil. Nanomaterials can release materials slowly and selectively and can also indicate a lack of nutrients in the soil (such as color change)

and can be used for a progressive early warning system for farmers to determine the application of rate and frequency.

#### 2.7. Plant-Pathogen Detection:

Smart field systems detect, locate and report pathogens, then apply pesticides and fertilizers as needed before symptoms appear (Bergeson *et al.*, 2010b). Instinctive, NPs can be applied as quick identifying tools to detect bacterial, viral, and fungal diseases of crops (Chartuprayoon *et al.*, 2010), thus it has been currently surveying in its initial step. Nanoparticle-based sensors can provide better detection parameters for the detection of disease-causing viruses in plants (Yao *et al.*, 2009). NPs can be modified directly to detect pathogens or applied as an analytical tool for the detection of compounds that indicate disease. Nanochips chips containing fluorescent oligonucleotide entrap enter through hybridization which can be detected (López *et al.*, 2016). The above-mentioned nanochips are named as their reactivity and selectivity for detection of each nucleotide replace in viruses and bacteria (Yao *et al.*, 2009; López *et al.*, 2016) by using silica NPs. The fluorescence is conjugated through antibodies for the detection of *Xanthomonas axonopodis PV. Vesicatoria* is caused by bacteria. Spots on the plants of the Solanaceae family, showing the potential of nanoparticles to detection of a disease (Singh *et al.*, 2010) using a yellow nano immunosensor, the Surface of Plasmon Resonance (SPR) can be used for detect diseases for certification of seed and established a phytosanitary system. The study on the field application of nano sensors for the detection of pathogens is of great value for the rapid diagnosis and management of a disease.

#### 2.8. Major Important Nanoparticles (NPs):

#### 2.8.1. Gold:

Gold (Au) nanoparticle is used to identify protein interactions. They are used as a laboratory monitoring of the DNA fingerprints to detect the detection of DNA antibiotics and aminoglycoside (such as streptomycin, gentamicin and neomycin) in samples. Nano-rod gold is used to identify cancer stem cells benefiting from identification of the cancer treatment, and types of bacteria (Tomar *et al.*, 2013).

#### 2.8.2. Alloy:

Nanoparticles showed different structural properties in bulk samples (Mohl *et al.*, 2011). The alloy NPs characteristics are affected in these two metals and showed further benefits than normal metal NPs (Jain *et al.*, 2017).

### 2.8.3. Magnetic:

Magnetic nanoparticles such as magnetic fields and magnetite are called biocompatibility. It has been energetically studied for the treatment of cancer, cell alignment, and operation; for Induction of a drug, Gene Delivery, Analysis of DNA and Nuclear magnetic resonance imaging (NMRI), Magnetic Resonance Tomography (MRT) (Bakshi *et al.*, 2014).

### 2.8.4. Silver:

Silver nanoparticles are the most important because they have antimicrobial effects on the viruses, bacteria and eukaryotic microbes (Gladkova *et al.*, 2013) are used in Neem (Shankar *et al.*, 2004) Pepper (Stern *et al.*, 2008), and Papaya (Rajput *et al.*, 2017).

#### 2.8.5. Copper:

Copper and copper oxide nanoparticles are very important because copper is the most important modern technology type and is easy to use (Zafar *et al.*, 2017). It can be used as an active electrode material as well as active electrode materials such as ammonia and carbon monoxide, hydrocarbon and ammonia nitrogen phenol and carbon monoxide, hydrocarbons and phenols, as well as carriers of catalysts and catalysts (Patil *et al.*, 2012).

#### 2.8.6. Inorganic:

In modern material science, the unique physical characteristics of inorganic nanoparticles in biotechnologies, particularly in biotechnology, play an important role. Based on the two factors of these two inorganic nanoparticles, they have specific material properties which are mainly subdivided with optical, magnetic, electrons, and catalytic properties. The new physical properties are mainly related to them because their size is close to the nano size (Xiong *et al.*, 2017).

#### 2.9. Synthesis and Biosynthesis of Nanoparticles:

Nanoparticles can be made by using chemicals and biologically. With the help of chemicals, the synthesis is toxic because many chemicals are presented. The method of nanoparticles preparation with the help of using microorganisms to prepare nanoparticles (Zhang *et al.*, 2014) enzyme (Rao *et al.*, 2016) and using plants extract (Hulkoti *et al.*, 2014). The mechanism for microorganism biosynthesis of nanoparticles is a kind of ecological equipment. Other microorganisms, prokaryotes, and eukaryotes are used to synthesize metal nanoparticles. Metal oxides nanoparticles such as gold, platinum, zirconium, palladium, iron and cadmium. Microorganisms comprise bacteria, actin festivals, mushrooms, and birds. The NPs synthesis as a function of the position of the nanoparticles has been intracellular or extracellular (Bordes *et al.*, 2009).

#### 2.10. Metal NPs Synthesis in Normal Medium:

The metal oxide NPs (Nanoparticles) synthesis has been playing an important role because it is necessary to maintain a simple and fast synthesis and a cell culture and an environment of various types of nanoparticles (Ghosh *et al.*, 2020). Nanoparticle synthesis mechanisms are in principle the same for microorganisms and plants. Metal salts containing metal ions are reduced to the reactor by the first reducing agent. The atom obtained is then cut into a small group and pushes with particles. Shankar et al. (2004) reported that the presence of secondary proteins and metabolites in the assignment of geranium sheets and the presence of second metabolites and tarpons reduce the silver and oxidized ions in the carbonyl group. The analysis of transcriptional infrared spectroscopy (FTIR) of this study (FTIR) showed that the ester group C = 0 acts as a reducing agent and a protein and participates in the surface coating of the gold nanoparticles synthesis of the metallic NPs by the "green" method. Biomass or extracts from various plants have been used successfully as reducing agents (Duhan *et al.*, 2017). The application of several helpful plants for the integration of silver NPs has been reported. Table 1 compiles a list of some plants that have been reported to be used in nanoparticle synthesis.

#### 2.11. Proposed Mechanism for Nanoparticle Synthesis:

Despite a large number of reports on the use of biological pathways for the synthesis of nanoparticles, there are data available to understand the exact mechanism. The common explanation is the involvement of proteins as enzymes and cofactors with redox potential and acting as an electron shuttle, playing an important role in the metal reduction.

S. No	Name of Plant	Nanoparticle (NPs)	Size in nm	References			
Leaves Extracts							
1.	Argemone maxicana	Silver	30	(Singh <i>et al.,</i> 2010)			
2.	Acalypha indica	Silver	20-30	(Krishnaraj <i>et al.,</i> 2010)			
3.	Mangifera indica	Silver	20	(Hulkoti <i>et al.,</i> 2014)			
4.	Cassia fistula	Silver	50-60	(Lin <i>et al.,</i> 2010)			
5.	Doipyros kaki	Platinum	2-12	(Song <i>et al.,</i> 2010)			
6.	Murraya koenigii	Gold & Silver	10-25	(Christensen et al., 2011)			
7.	Aloe vera	Gold & Silver	15.2	(Chandran <i>et al.,</i> 2006)			
8.	Eucalyptus camaldulensis	Gold	5.5 - 7.5	(Ramezani <i>et al.,</i> 2008)			
9.	Magnolia Kobus	Copper	50-250	(Hosseini <i>et al.,</i> 2014)			
10	T. procumbens	Copper	2000-5000	(Gopalakrishnan <i>et al.,</i> 2012)			
11.	Cassia fistula	Zinc	5-15	(Suresh <i>et al.,</i> 2015)			

Table 1: Synthesis of Nanoparticles reported in the Literature.

12.	Ixora coccinea	Zinc	145.1	(Yedurkar <i>et al.,</i> 2016)			
Fruit Extract							
13.	Emblica officinalis	Gold & Silver	10-20&15-25	(Ankamwar <i>et al.,</i> 2005)			
14.	Musa paradisica	Palladium	50	(Bankar <i>et al.,</i> 2010)			
15.	Citrus aurantifolia	Zinc	50-200	(Samat <i>et al.,</i> 2013)			
16.	Sambucus nigra	Silver	20-80	(Malik <i>et al.,</i> 2014)			
Flower Extract							
17.	Nyctanthes arbortristis	Gold	19.8	(Das <i>et al.,</i> 2011)			
18.	Trifolium pratense	Zinc	60-70	(Dobrucka <i>et al.,</i> 2016)			

The method was composed of transfer ions with microbial cells if enzymes are present to form nanoparticles. It is linked to the size of the reduced nanoparticles outside the cell and the nanoparticles formed in the vivo are small. The size limitations can be associated with the body nuclear forming particles (Narayanan *et al.,* 2010).

#### 2.12. Extracellular synthesis method of NPs by Mushroom:

Compared to the intracellular synthesis of NPs in the cells, the cells are more used and do not replace the components of the cell adjacent to the cells. In most cases, the fungus is known to produce nanoparticles outside the cells because it has been a large number of unknown components that participate in the reduction and reach in the NPs (Narayanan *et al.*, 2010). In addition to extracellular forms, unicellular and multicellular organisms produce inorganic materials (Saif *et al.*, 2016). The ability of microorganisms such as bacteria and fungi to control the synthesis of metal nanoparticles is being used to search for new materials. Because of their tolerance to metals and their ability to bioaccumulate, fungi occupy a central place in the study of the biophysics of metal nanoparticles (80). Microorganisms are potential nano factories for the ecological and economic synthesis of various metal nanoparticles (such as silver, gold, copper) and metal oxides (such as zinc oxide, copper oxide, etc.) These nanostructures can exist in different shapes and forms, such as nanotubes, nanomaterials, nanorods, and more.

#### 2.13. Use of Nanoparticles (NPs):

#### 2.13.1. Food:

Nanofood is a term used to describe foods added using artificial nanotechnology, tools, or nano artificial materials during culture, production, of processing, or packaging. The development of nano fuels has several objectives. These measures include improving food security, nutrition, and taste and reducing production and consumption costs. In addition, Nano Hoods has many advantages, including health additives, longer distribution due to new flavors, and new varieties of flavor. The uses of nanobiotechnology in food vary from food processing to food packaging to improve the use of nutrients due to food processing in agricultural applications.

#### 2.13.2. Gene Delivery:

Gene delivery is an important technology that can efficiently introduce target genes and express encoded proteins in the appropriate host or host cell. Today, there are different major types of gene delivery systems, mainly using viral vectors, retroviruses, and adenoviruses, nucleic acid electrochemistry, and nucleic acid transduction (Koteswaramma *et al.*, 2018).

### 2.13.3. Cancer Treatment:

There are currently various nanoparticle systems in biomedicine finite research that focus on the treatment of cancer. Many concerns including quantum points and some innovative natural nanoparticles, including quantum points and some natural nanoparticles in precious metal (mainly gold and silver) and of some magnetic oxides (including Fe3O4 magnetic) (Golesta *et al.*, 2018). A single optical conversion process can be used to allow sensitive therapeutic agents for cancer treatment (Sharma *et al.*, 2018).

### 2.14. Use of Nanobiotechnology by remove Heavy Metals from Water:

Heavy metals such as arsenic, cadmium, and mercury have a serious effect on water. Sources of heavy metals in the country are domestic and agricultural pollution. Copper and zinc are considered essential elements as well as in animals, but high levels of these two elements in the human body will affect the kidneys, anemia, and toxicity. If the arsenic content in water is high, it will cause melanosis, keratosis, and hyperpigmentation in humans. Cadmium affects human bones and kidneys when its amount in water is high. Cadmium has an acute effect on children. A large number of NPs have been prepared and applications to remove heavy metal contaminants from wastewater due to their high stoma value and the large specific surface area.

#### 2.15. Characterization of Nanoparticles:

Many different techniques are used to characterize nanoparticles by XRD (X-ray diffraction) (Bruker D8 Advance, Germany), SEM (JEOL, JSM5910). UV-V is a spectrophotometer (Perkin Elmer UV/Visible Lambda25). Fourier Transform Infra-Red Spectrophotometer (FTIR) Brook, Tensioner 27.

#### 2.15.1. X-Rays Diffraction (XRD):

The XRD analysis of the nanoparticles was performed by XRD (Bruker D Advance, Germany) using Cu and Ka radiation within the wavelength of 26°A. Use a current of 30mA and a power of 40KV.UV/Visible spectroscopy. Observe the light absorption spectra of nanoparticles and coatings using an ultraviolet/visible spectrophotometer (Perklin Elmer UV / VI Slambda 25). Spectroscopy was carried out at room temperature. The wavelength range is 200,800 nm.

#### 2.15.2. Fourier Transform Infra-Red Spectrophotometer (FTIR):

The copper oxide nanoparticle powder sample is loaded into the FTIR spectrometer (Bruker, Tensor 27), the scanning range is 620686 cm1, and the resolution is 4 cm1. FTIR enables interface analysis to study the surface absorption of functional groups on nanoparticles. One advantage of FTIR is that it allows the user to analyze a layer of nanoparticles.

## 3. Discussion:

In the agricultural field, nanotechnology has been used to increase crop yields and improve quality by improving agricultural systems. The emergence of man-made nanomaterials and their role in sustainable agriculture has greatly changed global agriculture thanks to novelty, rapid growth, and huge forecasts to meet the global demand for food. In sustainable agriculture, protecting the environment from pollution is the key objective of trade, and nanomaterials provide a guarantee for better management and protection of plant production inputs. The potential of nanomaterials has fueled a new green revolution to reduce agricultural risks. However, there is still a large gap in our understanding of the absorption capacity, allowable limits, and ecotoxicity of different nanomaterials (Govindarajan et al., 2016; Esfanddarani et al., 2018). Therefore, further research is urgently needed to clarify the behavior and fate of modified agricultural inputs and their interactions with biological macromolecules in living systems and the environment. Peaks at 32.8°, 35.9°, 39.1°, 46.3°, 49.1°, 52.9°, 58.7°, 66.6°, 68.3°, 72°, 6<sup>th</sup>, and 75.5° can be assigned to (111), (1), (1)), (112), (202), (020), (202), (311), (113) and (400) corresponds to the monoclinic phase values of the nanoparticles reported in the literature (Xiong et al., 2017) and to the corresponding "JCPDS" (Joint Committee on Powder Diffraction Standards Tag No. 45 (0937). The particle size obtained from the broadening of the XRD (X-ray diffraction) line is about 18.34 nm. The FTIR spectrum is the measurement sample Interpretation of the IR spectrum plot of the absorption of IR radiation versus wavelength involves the correlation of the absorption band (vibration band) with the sample compound. In this way, the biomolecules present in plant extracts can be identified, and these biomolecules are responsible for minimizing and stabilizing the green synthesis of these biomolecular NPs.

# 4. Conclusion:

Due to their excellent properties, NPs have become important in several fields in upcoming years, like energy, health, environment, agriculture, etc. NPs (Nanoparticles) technology has unlimited potential for transforming unwell soluble, absorbed, and unstable bioactive substances into promising release agents. Nanobiotechnology is a powerful tool to improve agriculture. However, it exhibits different behavior from many of its counterparts, producing nontoxicity effects based on completely different parameters rather than dose versus mass. Rather, characteristics like aggregation, morphology of substance, concentration of nanoparticles, surface of variation, and size determine the toxicity of nanomaterials, which determine the different biochemical effects they can cause for plants. Whether its origin is a product for a specific agricultural purpose, and it can be released into the environment through inadequate treatment of waste containing nanomaterials, it is essential for recognizing the exact toxic special effects of the NPs harm that can cause nanostructured systems in plant organisms. Therefore, it is significant to develop effective regulations to prevent its entry, protect other plant species that interact with certain NPs (Nanoparticles) with its adverse effects beneficial to its development.

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# Nanobiotechnology: As Emerging Field in Agriculture

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### Abstract

Food in the world is a challenge because of the limited resources available to cater to the world's rising population. Governments, organizations, and researchers are making various efforts to reduce the shortfall of the human food web. Before the industrial revolution, agriculture provided the foundation for growth in around 90 nations. Although nanotechnology has found industrial uses, its usage in agriculture is considerably more recent. In this study, we will look into nanotechnology applications in fields, such as crop cultivation, protection, and disease monitoring. The United States has the most research budget for nano research, trailed by German and Japanese, while China has the most papers and the most patents. Carbon-based nano-fertilizers, nano-pesticides, and nano-herbicides improve sprouting, chlorophyll content, nutritional utilization efficiency, and plant development, therefore increasing food output. Nanobiotechnology plays important role in pest control and management by the formation of nanocomposites, Fe<sub>3</sub>O<sub>4</sub>, and Au nanoparticles (NPs). Nanotechnology offers promising uses in genomic transfer via nanoparticles. It could be utilized to carry genome and certain desirable compounds into phytoconstituents, hence preventing insect invasion.

Keywords: Nanotechnology; Agriculture; Nano-fertilizer, ; Pest Ciontrol; Nanoparticles (NPs)

### 1. Introduction:

The advent of nanotechnology has become a hallmark of the present era after the mid-1700s Industrialization, the 1940s War, the 1960s Agro Renaissance, the 1980s IT Revolution, and the Biotech Revolution of the 1990s. Nanotechnology is a multidisciplinary scientific approach that entails designing, inventing, and implementing materials and devices at the molecular level on a nanoscale level (Fakruddin, 2012). The term "nanomaterial" originates from the term "nano," which means "tiny" in the Greek. Generally, nanomaterials have dimensions of 1 to 100 nanometers (Rai & Ingle, 2012). It is used to measure, manipulate, and manufacture things on the nanolevel (nm). Nanometers are length units that correspond to a billionth of a meter, according to the SI standard (Systeme Internationale d'Unite, in French), (Mongillo, 2007; Can *et al.*, 2011).

These advanced polymers typically have unique compositions, sizes, shapes, surface areas, conductivities, or surface chemistry that allow them to exhibit unique properties, and they have been used in a variety of applications including textiles, electronics, engineering, and medical (Smith *et al.*, 2007). Nanotechnological advances are being

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made to understand and control matter at the nanolevel range of 1 to 100 nanometers, where enhanced physicochemical and biological characteristics are enabling novel applications (Vyom *et al., 2012*).

Nanoscience and nanotechnology are cutting-edge scientific breakthroughs from the twenty-first century. According to Stat Nano, from 2000 to 2019, the Academic databases (WoS) indexed over 1.8 million nanotechnology publications. It has enormous potential for developing advanced materials with improved functionality. Diagnostic equipment, imaging, athletics, theragnostic, telecommunications, medicines, ecological clean-up, aesthetics and serums (USEPA, 2007; NCPI, 2011), agribusiness, fabrics, and nutrition are just a few of the areas where nanotechbased products are being used (Caruthers *et al.*, 2007; Sharon *et al.*, 2010; Sastry *et al.*, 2011). Nanotechnologies will have a greater impact on the global economy in the future as more items using nanotechnologies shift from research and development to manufacturing and commerce. The classic nanoparticle induction technique, which involves nuclear, chemical, and particulate processing in pressure or a solvent mixture, is expensive (Mandal *et al.*, 2006; Anandan *et al.*, 2008) and inefficient in terms of material and energy usage (Pugazhenthiran *et al.*, 2009). Many nanoparticles were synthesized from fungus and are now widely employed in different disciplines both domestically and overseas (Kashyap *et al.*, 2013).

A wide range of essential areas of food manufacturing will be influenced by nanotechnology in the future, from the development of novel foods and ingredients to food safety (Pathakoti *et al.*, 2017). As a result of nanotechnology, genetically modified crops, livestock, and fisheries input, chemical pesticides, and precision agricultural technologies are predicted to be developed more efficiently. To increase crop yield, precision farming involves monitoring environmental factors and taking precise actions (Chen & Yada, 2011). There are a variety of pre-harvest and post-processing modifications that impact the food's biological and biochemical makeup and composition. This means that advances in biochemistry and biology nanotechnology might potentially have an impact on the edible industry (Sozer & Kokini, 2009; Jain *et al.*, 2016). The detection and quantification of contaminants in foods must be simplified, quicker, more sensitive, and less expensive. Sensors and systems enabled by nanotechnology have been utilized to identify food pollutants more quickly and non-invasively in the past decade, thanks to significant breakthroughs in nanoscience.

#### 1.1. Objectives:

- To explore the latest status of nanobiotechnology and its applications
- To aware the researchers for new technology of NPs in next generation
- To measure the benefits and approaches of Nanobiotechnology in coming era

### 2. Review of Relevant Literature:

#### 2.1. Application of Nanobiotechnology in Agriculture:

The conventional farming industry employs agrochemicals for the development and protection of its crops, including fertilizers, pesticides, fungicides, insecticides, and herbicides. Their excessive use, on the other hand, is harming the ecology by introducing poisons into the ground and surface water (Mukhopadhyay, 2014). Nanobiotechnology, on the other hand, has the potential to tackle agricultural problems and reduce chemical misuse.

#### 2.2. Nano-Fertilizers:

Fertilizers are chemical agents that provide sustenance to plants (Bottoms & Emerson, 2013). Since the early 1950s, agrochemicals have been used and have become entirely compatible with fertilizer application. Inorganic fertilizers misuse, on the other hand, has increased water pollution, diminished soil fertility, and induced eutrophication. Inorganic fertilizers can cause environmental damage, while micro fertilizers have the capacity to mitigate this. Because they include nutrients and growth-promoting chemicals contained in nano polymers, chelates, or emulsions, nano fertilizers can promote plant development. Water pollution is diminished, soil toxicity is reduced, nutrient release is delayed and constant, production output (yield) is greater, photosynthesis is improved, fertilizer impact period (effect period) is extended, and soil nutrients are raised (Naderi & Danesh-Shahkari, 2013; Mehrazar *et al.*, 2015).

Many scientific studies have documented successful applications of metallic nano fertilizers in promoting plant productivity. The spinach cultivars (Varamin 88 and Viroflayand) treated with iron-chelated nano fertilizer at 4 kg ha-1 caused improvements in a wet weight of 58% and 47% as well as canopy index and floral organs (Moghadam

*et al.*, 2012). Shahrekizad *et al.*, 2015 showed that the Sunflower iron availability was similarly boosted by Fe<sub>3</sub>O<sub>4</sub> nanoparticles. When Cu NPs were applied to wheat, there was an increase in leaf area, chlorophyll content, fresh and dry weight, and root dry weight (Hafeez *et al.*, 2015). Liu & Lal (2014) found Plant development (32.7%) and harvest index (20.3%) are stimulated with synthetic apatite NP application respectively. Compared to the traditional urea fertilizers, nanozeourea boosted the amount of crude protein in maize plants grown in humus soil by 26% and in drained soil by 36% (Manikandan & Subramanian, *2016*). By enhancing phosphorus and nitrogen levels in cucumber plants, SiO2 nano-fertilizer induced growth and increased yield (Yassen *et al.*, 2017).

### 2.3. Nano-Pesticides:

Crop production is threatened by plant pests and must be controlled rigorously. Classical pest management Methodologies use a variety of synthetic chemicals. Furthermore, the overuse of chemicals leads to disturbance in organisms' systems, the ecology, and plant capacity to fix nitrogen (Ghormade et al., 2011; Elrahman & Mostafa, 2015; Bhattacharyya et al., 2016). Nanopesticides provide a mechanism to regulate pesticide distribution while also achieving higher benefits with a lower chemical dosage. The first kind of nano pesticides is supposed to be active substances that are designed, such as metallic or natural nanomaterials e.g., Ag and Cu (Guilger et al., 2019; Bratovcic, 2020) or metallic oxide nanoparticles e.g., ZnO, CuO, MnO<sub>2</sub>, SiO<sub>2</sub>, TiO<sub>2</sub> (Elmer *et al.*, 2018; Asiri *et al.*, 2020; Shukla et al., 2020). The use of nano pesticides improves a pesticide's efficiency and longevity while reducing the quantity of active chemicals present and minimizing or eliminating any possible risks. Copper-based nanoinsecticides in agricultural production systems may have unexpected repercussions on non-target soil microbial communities due to their broad antibacterial spectrum In the presence and absence of an edaphic organism, Peixoto et al., 2021 evaluated the influence of a conventional Cu  $(OH)_{2}$ - nano-pesticide on the activity, morphology, and richness of microbial communities over three months, at daily and annual agricultural application dosages (the isopod *Porcellionids pruinosus*). The use of nano-pesticides caused significant changes in the structure of both bacterial and fungal communities, especially throughout the treatment period. On a physiological level, populations exposed to nanoscale insecticide showed a large increase in microbial carbon consumption capacity and a considerable decrease in β-glucosidase activity. According to investigation the Copper (II) hydroxide nano pesticides influence the soil microbe's population, potentially altering its ecological niche. Researchers are now focusing on the application of silica nanoparticles in agriculture. Crop growth and yield can be improved by using nano-silica-based fertilizers, insecticides, and herbicides. Silica nanoparticles are simple to incorporate into crops. It has the potential to alter the plant's metabolic functions (Rajiv et al., 2020).

#### 2.4. Nano-Herbicides:

Weeds are undesirable plants that impair crop output and must be removed from agricultural grounds as soon as possible. Although most conventional herbicides target the vegetative plant components, they ignore the bulbs and root systems that can grow into a new plant under the right conditions (Ali *et al.*, 2014). Herbicides are barely had potent in unirrigated agroecosystem because they lack moisture (Subramanian & Tarafdar, 2011). The useage of nanoscale herbicides might be an efficient technique of weed control. The Nanoparticle's herbicide provides a number of advantages over traditional herbicides, including enhanced water solubility, reduced plant resistance, and lower toxicity. NPs of poly ( $\varepsilon$  caprolactone) containing the herbicide atrazine were applied to both target (*Brassica* sp.) and non-target crops (*Zea mays*). An off-target plant (*Zea mays*) was not harmed by encapsulated pesticide. Due to their enhanced mobility of atrazine, NPs are more effective as an herbicide against Brassica species (Pereira *et al.*, 2004). Nanoherbicides based on chitosan/tripolyphosphate NPs is also being developed. With paraquat herbicides, chitosan/tripolyphosphate NPs are less damaging to crops and effective in weed control (Grillo *et al.*, 2014). An atrazine-loaded nano capsule treatment of 2000 g ha<sup>-1</sup> inhibited growth parameter in *B. pilosa* as well as the functioning of the photosystem II (Sousa *et al.*, 2018).

#### 2.5. Nano-Fungicides:

In contrast to viruses and bacteria, fungus are the most common plant pathogens. *Aspergillus, Fusarium,* and *Phytophthora* species are examples of molds causing the disease that can be used as nanomaterials in the creation of nanoparticles. (Yadav *et al.,* 2015). In the cure of wide range of Phyto disease, silver nanoparticles could be used as an effective antifungal agent. On a petri dish, an invitro test was performed. They treated the silver nanoparticles with 18 plant pathogenic fungi on agar extract (malt, potato dextrose and cornmeal agar). To assess the antifungal

action of silver nanoparticles against infections, fungal inhibition was computed. The findings demonstrated that silver nanoparticles had antifungal properties at various doses against these phytopathogens (Kim *et al.*, 2012). Gold Nanoparticles were also demonstrated to be beneficial against Bipolaris sorokiniana infection in *Triticum aestivum* (Mishra *et al.*, 2014). Antifungal activity of Ag NPs has also been identified against Candida species (Kim *et al.*, 2009; Panacek *et al.*, 2009), Bipolaris sorokiniana, and Magnaporthe grisea (Jo *et al.*, 2009), as well as powdery mildews (Lamsal *et al.*, 2011). Furthermore, when employed contrarily *A. alternata* and *Botrytis cinerea*, Ag and Cu NPs inhibited fungal hyphae development to the greatest extent at a dosage of 15 mg L<sup>1</sup> (Ouda, 2014). Park *et al.*, 2006 examined the effectiveness of nanoscale silica silver in controlling phytopathogenic microorganisms. *Rhizoctonia solani, Botrytis cinerea, Magnaporthe grisea, Colletotrichum gloeosporioides*, and *Pythium ultimate* were tested for antifungal activity using silica silver nanoparticles. Powdery mildew tested in the field for antifungal activity. The results suggest that silver and silicon dioxide are much less costly than commercial fungicides because they are nontoxic and safe for human health. This nano-formulation is very useful in the treatment of various fungal plant diseases (Bhattacharyya *et al.*, 2016).

#### 2.6. Pest Control:

Pesticides, insecticides, and fungicides are now the quickest and least expensive way to manage infestations. Biocontrol approaches are also now prohibitively costly. Uncontrolled pesticide usage led to series of issues notably severe impacts on human, pollinator, and household pets, and even the emission of such chemical into the topsoil and groundwater, and its unintended consequences on ecologies. Pesticides, insecticides, and fungicides are now the most efficient and cost-effective way to manage pests and diseases. A possible answer for this challenge might be the smart utilization of chemicals on the nanoscale. These ingredients are incorporated into the plant portion that has been damaged by disease or insect. Pesticide efficiency for pesticide application at a lower dosage. Nanomaterials for the transport of psychoactive chemical and medication molecules would be at the forefront in the near future for the treatment of overall plant pathogenic ailments. There are several nano materials, such as nanocomposites, iron oxide (IO) nano particles, and AuNPs that may be easily produced and utilized to create novel formulations such as nanopesticides, nano-insecticides, and nano-insect repellants (Barik et al., 2008; Owolade et al., 2008; Gajbhiye et al., 2009; Goswami et al., 2010). The pharmacokinetic characteristics of these nanoparticles might vary depending on their size, shape, and surface functionalization. They can also be utilized to change the kinetic characteristics of drug release, resulting in more prolonged drug release with less frequent dosing (Sharon et al., 2010). Nanotechnology has promise for genome transfer via nanoparticles. It might be utilized to deliver Genes and other desired substances into phytoconstituent to protect host plants from insect infestations (Torney, 2009). Validamycin (pesticide)-loaded porous hollow silicon nanoparticles (PHSNs) can be employed as an effective pesticide carrier method for constrained release.

#### 3. Conclusion:

Nanobiotechnology involves modifying organisms using nanotechnology, allowing integration of anthropogenic and natural components. Nanobiotechnology has a variety of uses in agriculture, the food industry, and other areas. This is really an innovative agro invention that can offer sequenced alternatives to traditional agricultural practices in the form of nanoagro composite. Nanoparticle based herbicides, pesticides, fertilizers, fungicides and pesticides are recommended to boost agricultural output in an environmentally acceptable manner. The role of nanobiotechnology has also been shown to be useful in diagnosing pathogens and detecting pesticide residues. New insect-resistant cultivars might be developed via nanoparticle-mediated gene transfer. Nanotechnology can therefore give green and eco-friendly options for increasing food productivity and insect pest management without damaging the environment.

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