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Synthesis of Biochar-Based Composites to Evaluate Morphology of Wheat Seedling

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ABSTRACT: *Recently, biochar has emerged as an effective nutritive source that plays a noteworthy contribution in improving the growth of various crops. Current research signifies the role of Biochar-composite in the growth of wheat seedlings. In this study, the carbonaceous biomass synthesized from garden waste was impregnated with macro- and micro-nutrients. The effectiveness of this synthesized Biochar-composite (GW-B) was studied by using wheat (*Triticum aestivum*) as an experimental crop. The GW-C was applied to wheat seeds grown in petri plates and pots by supplementing them with different concentration of prepared biochar. Application of biochar composites improved the growth of wheat seedlings in both petri plates and pots in addition to enhancement in morphological attributes. Higher concentration of biochar-composites improved the morphology of wheat seedlings in a dose-dependent manner e.g. shoot fresh weight, shoot dry weight, root fresh weight, root dry weight, number of leaves per plant, plant height, leaf area and root length. Biochar-composite at concentration of 0.8 enhanced morphology of supplemented wheat seedlings sown in petri plates at equidistance manner over 5 layers of Whatman filter paper (no. 1). Biochar-composite of 0.03 g enhanced the morphological attribute of wheat seedling that were sown in equidistance manner in pot experiment. It was found that biochar amended soil improved the growth and morpho-physiological attributes of wheat seedlings, thereby enhancing the yield of crop.*

Key words: *Biochar, composites, wheat seedlings, nutritive source.*

INTRODUCTION

Biochar congregated from horticulture or boscage residue, usage as a soil alteration such as carbon. When alteration happens into the soil, biochar better the soil fertility, and increased the development of farming (Agegnehu et al., 2016; Agbna et al., 2017; Werner et al., 2018; Adli, 2019; Das et al., 2020).

Enhance the bio-oil temperature proceed to the considerable carbonized raw-material in which enhance the carbon content, acquired a favorable product must be assembled in temperature reactor, increase the temperature or time period of this procedure at the eventual temperature. Bio-oil boost the biochar usage in which extensively content of carbon or inexpensive development at the same period applicable for their utilization. Biochar utilized in surrounding areas, or their valid features and their quality (Butnan et al., 2015).

Recently, biochar usage as a catalyst, soil alteration, fuel cell, impurity sorption, and activated charcoal. The charcoal is also known as biochar when utilized as a soil alternation for the strengthened of the soil condition. Biochar usage improved the productive of fuel and advantageous to both farming and surrounding areas. The utilization of biochar, associate with inanimate fertilizer or living alteration, upgrade the effectiveness of fertilizers and development of soil agriculture (Ahmed et al., 2016; Amin et al., 2016; Al-Wabel et al., 2018; Braghiroli et al., 2018).

According to Blackwell et al. (2015) biochar mineral complex has a great influence on mycorrhizal association.

The major use of Biochar is it utilize as a renewable energy source (Ozcimen and Karaosmanoglu, 2004). Carbonization substance can be burned or co-burned by adjoining power plants. Biochar differentiate with bio fuels hold minimum quantity of notable chlorine, and vapors affect the depletion of boiler productivity and enhance the discharge of inorganic particles. The chlorine altered consequences from the attentiveness of the pyrolysis operation. Throughout pyrolysis, the chlorine holds the bio energy is free in gaseous shape and proceed to the environment. These consequences alter the composition of substances and the processes of bio-energy. Inorganic combination establishes the admirable particles enhance the slurry manufacturing in combustion boilers. This issue vanishes the short time of carbonization of a combustible, which is significantly necessary for the production of bio-energy combustion. Accordingly, Biochar is a main element in decreasing this drawback. Similarly, combustible is replacement to standard natural gases. Biochar-mediated sorption of organic impurity may be built on the concept of electrostatic interconnections with polar and non-polar category. Sorption of inorganic impurities, involvement the ion of massive metals, by the utilization of biochar is distinguished by four procedures:

➤ Cation and anion exchangers, ➤ Precipitation, ➤ Anionic metal attractiveness, ➤ Cationic metal attractiveness.

The current research planned to see the role of Biochar-composite in the growth of wheat seedlings.

MATERIALS AND METHOD

Biochar Composite Synthesis

The steam activation was utilized for the production of charcoal. This charcoal was activated in the cooking-pot at a medium temperature in the absence of oxygen for 24 days. The charcoal which was prepared in the cooking pot was remained stay hot and it required a sufficient time i.e. 30-40 minutes to cool down. Transfer the charcoal in the mortar and pestle and grind up into a very fine powder. After that, the powder was transferred into the clean bowl. Powder was dried and used to prepare the solution. Add the required nutrients to the powder in a small quantity, stirring it with rod under the magnetic stirrer hot plate until the mixture was dissolved. Separate the filtrate and residue by vacuum suction pump. The residue was dried at 100°C for 24 hours in an oven. The actual yield of charcoal was 100 grams and the residue was 87.55 grams.

Slow release fertilizer was synthesized by using biochar as a support material. A homogenous suspension was achieved by adding 100 grams of charcoal in 250 ml distilled water. Afterwards, following solutions were added in

reaction flask and allowed to stir at room temperature for 30min;

- 1). 50ml solution of 0.003 moles of sodium nitrate
- 2). 0.06 moles of sodium phosphate
- 3). 0.0003 moles of sodium chloride
- 4). 0.0001 moles of calcium chloride
- 5). 0.0002 moles of manganese nitrate
- 6). 0.0003 moles of iron sulfate
- 7). 0.0003 moles of magnesium chloride
- 8). 0.03 moles of potassium dihydrogen phosphate

Later on, the contents were filtered by vacuum filtration technique and followed by drying at 100°C for 24 hours in oven. Final product was grinded and stored in air-tight jar. Four Biochar treatments were used for the present study and labelled as B1= 0.01g, B2= 0.02 g, B3= 0.03 g and B4= 0.04 g.

RESULTS

Biochar composites have significant effect on growth and morphology of wheat seedling as compared to untreated ones in petri dish. Application of biochar composites improved the growth of wheat seedlings in both petri plates and pots. At concentration of 0.8 enhanced morphology of supplemented wheat seedlings sown in petri plates

Petri plate

A significant increase in plants height was observed in plants supplemented with B4 treatment as compared to B3, B2 and B1. The increase in plant height and various other parameters such as shoot fresh weight, shoot dry weight, root fresh weight and

root dry weight obeyed the order i.e. (B4) > (B3) > (B2) > (B1) > (control) (Fig 1-5). Highest value of root dry weight was observed in B4 treatment. B2 and B3 treatments showed no significant differences (Fig 5). B4 treatment showed 1.5 folds' increase in root dry weight as compared to control conditions.

Number of leaves increased with increasing concentrations of biochar. The

number of leaves in B1 and B2 did not differ significantly (Fig 6). The number of leaves in B4 treatment showed 1.6 folds increase as compared to wheat seedlings grown in controlled conditions. Both B3 and B4 treatments showed significant increase in number of leaves as compared to control conditions (Fig. 6).

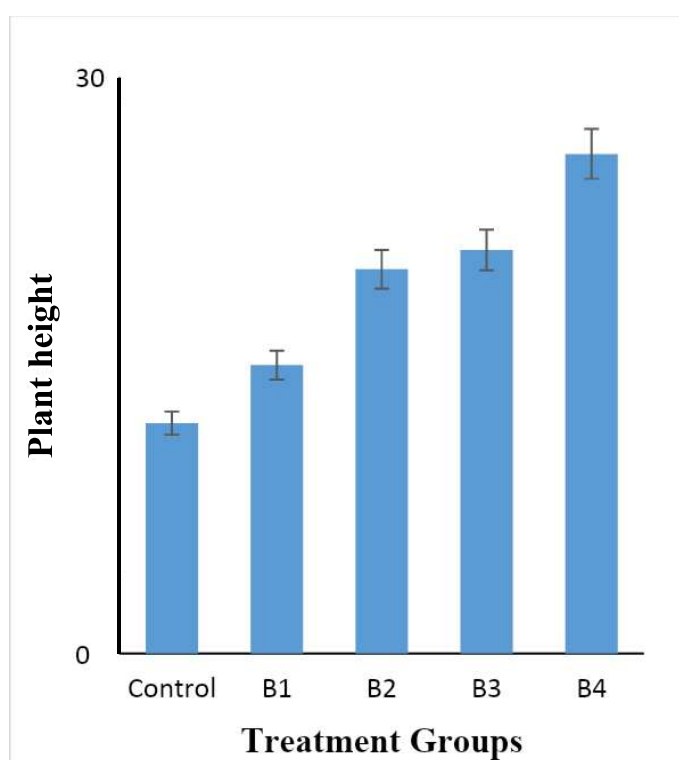


Fig. 1: Plant height against different experimental groups

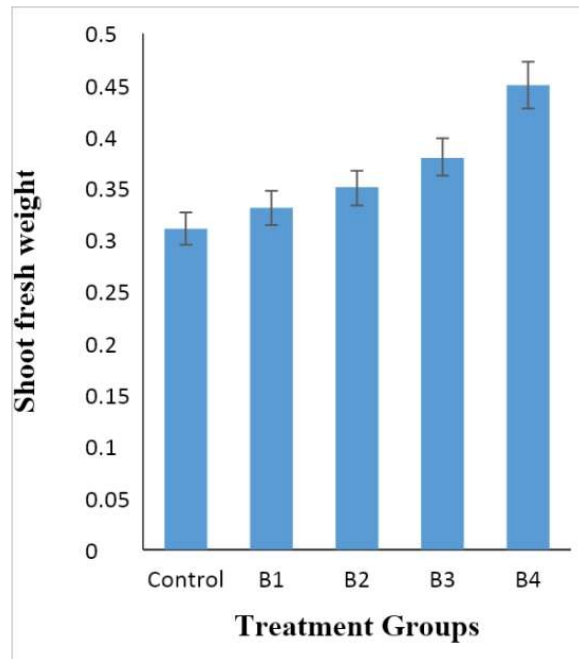


Fig. 2: Shoot fresh weight against different experimental groups

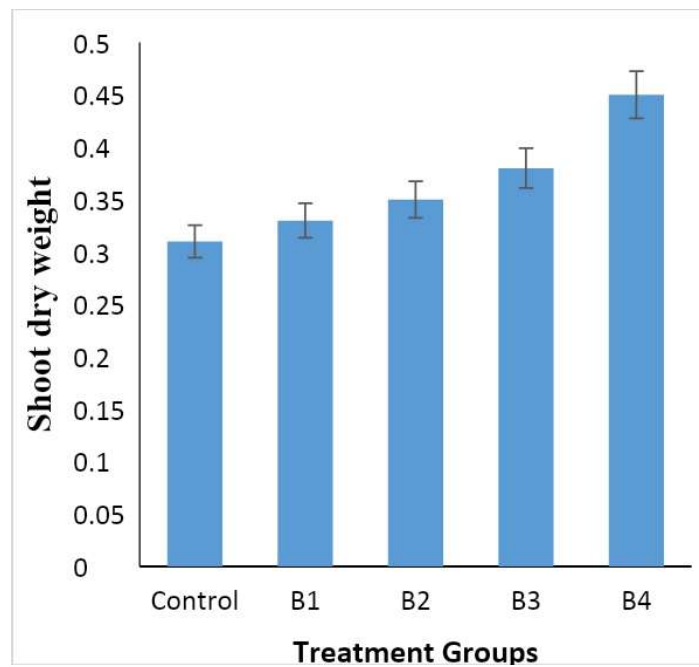


Fig. 3: Shoot dry weight against different experimental groups

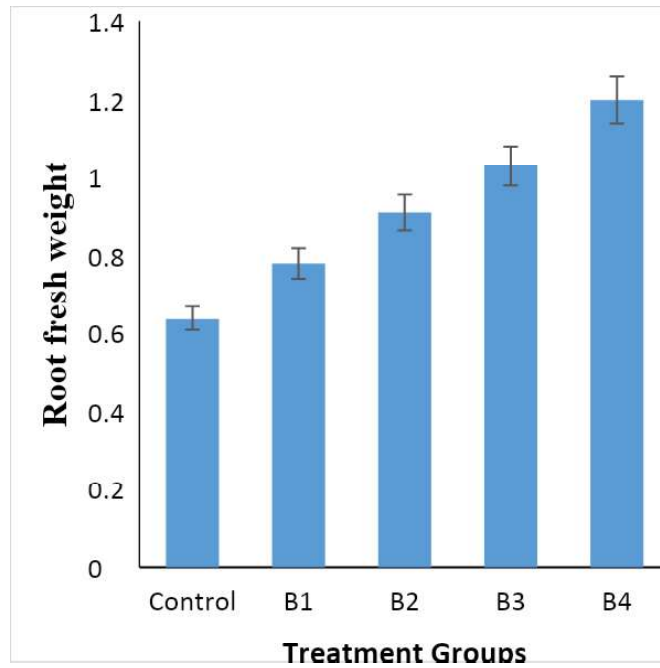


Fig. 4: Root fresh weight against different experimental groups

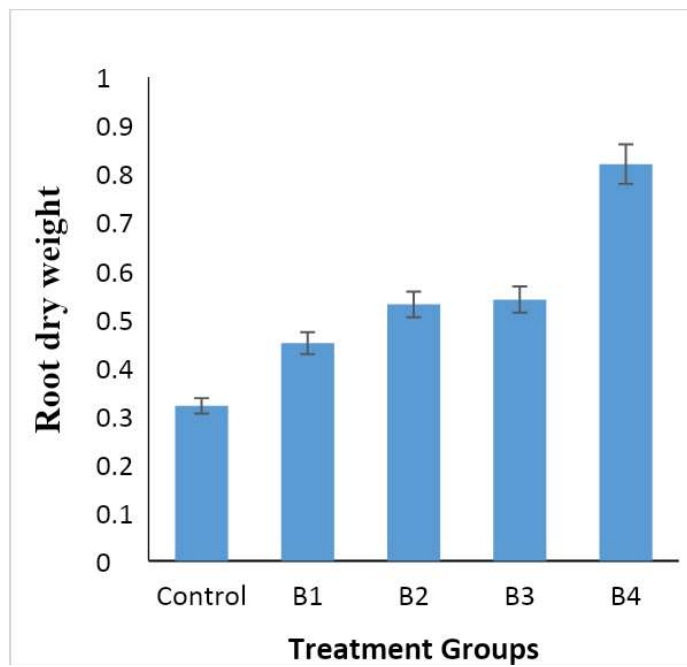


Fig. 5: Root dry weight against different experimental groups

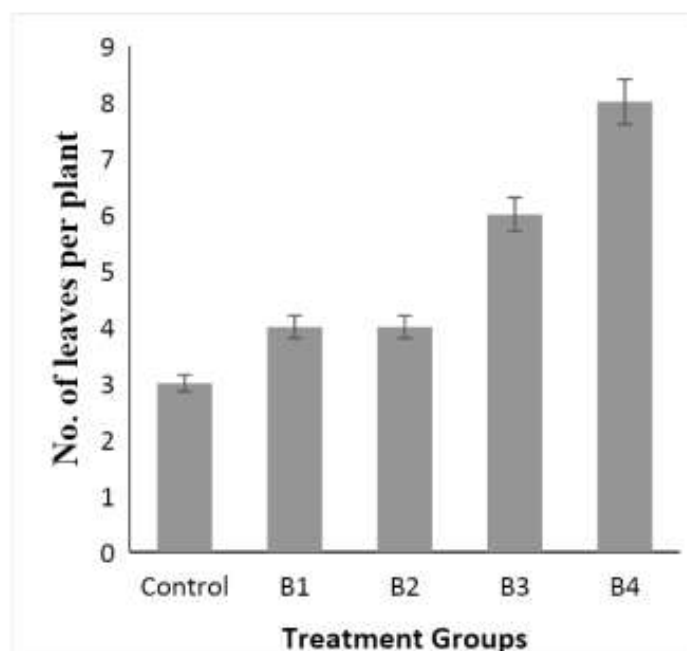


Fig. 6: No. of leaves per plant against different experimental groups

Seed tray

Biochar effect on seedling growth diverse with soil feature. Biochar composite may not be suitable as soil modification, significantly if they hold excessive amount of nutrients. It may inhibit the seed germination and assessing the presence of toxic compounds at the concentration of B4. Carbonaceous biomass may hold higher concentration of nutrients that could effect on seed germination. Biochar significantly increased the shoot dry weight of B3 at 2.7cm and B2 slightly increased at the length of 2.6cm (Fig. 7). In (Fig. 8) B3 has highest peak in plant height of 42cm. While B2 is having second highest value of plant height at 43cm. However, B1 and B4 has almost similar values. In (Fig. 9) Biochar composites remarkably increases the leaf area of B3 of wheat plant at 1.3cm. However, B2 is comparatively less

than B3 at 1.2cm. Biochar composites impressively increases the root dry weight of wheat plant B3 at 46cm. While B2 is more likely similar to B3 as there is only slight difference with dry weight of 45cm (Fig. 10). Root length increased by adding biochar in the seed tray. Highest value of root length was observed in B3 treatment. However, the least root length of the plant attains at 11 in control plant which is having a deficiency of nutrient. The other concentration i.e. B1, B2, and B4 slightly increase the root length with each other at 14, 19 and 21cm (Fig. 11).

Biochar composite increases the length of B3 at 37cm of wheat plant. However, B2 and B4 are little less in root length with B3 as B2 root length of wheat plant is at 33cm while B4 root length of wheat plant is at 35cm. B1 is having root length of wheat plant of 21cm. In all, control plant is having smallest root

length of wheat plant at 8cm. On the other hand, B3 is having largest root length in this biochar at 37cm (Fig. 11). B3 has highest value of number of values of

wheat plant while control plant and B4 having same quantity of number of leaves and both have smallest number of leaves of wheat plant comparatively (Fig. 12).

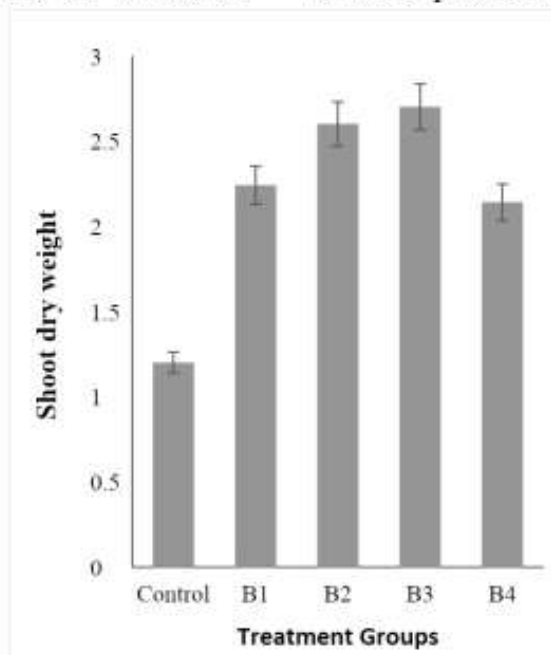


Fig. 7: Shoot dry weight against different experimental groups

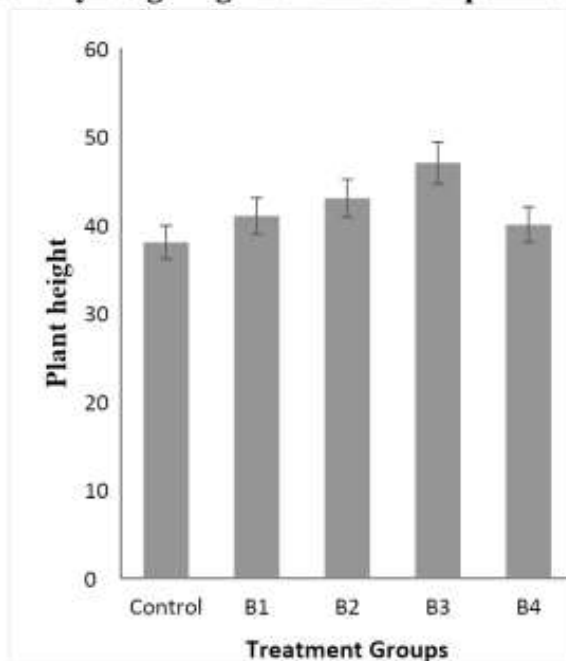


Fig. 8: Plant weight against different experimental groups

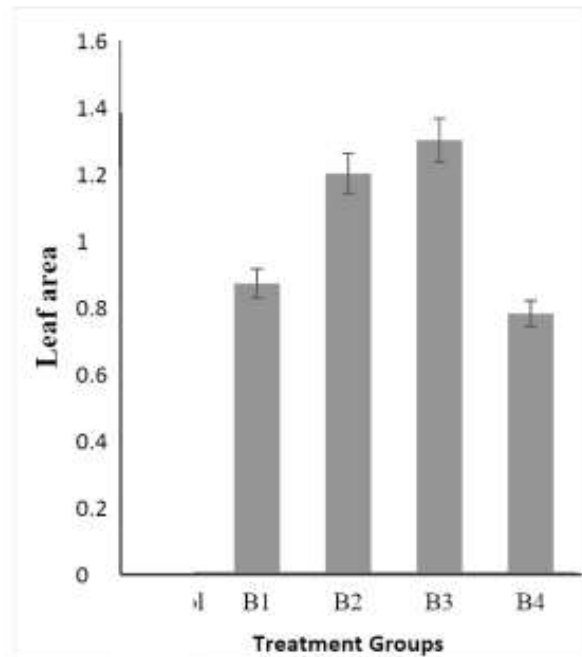


Fig. 9: Leaf area against different experimental groups

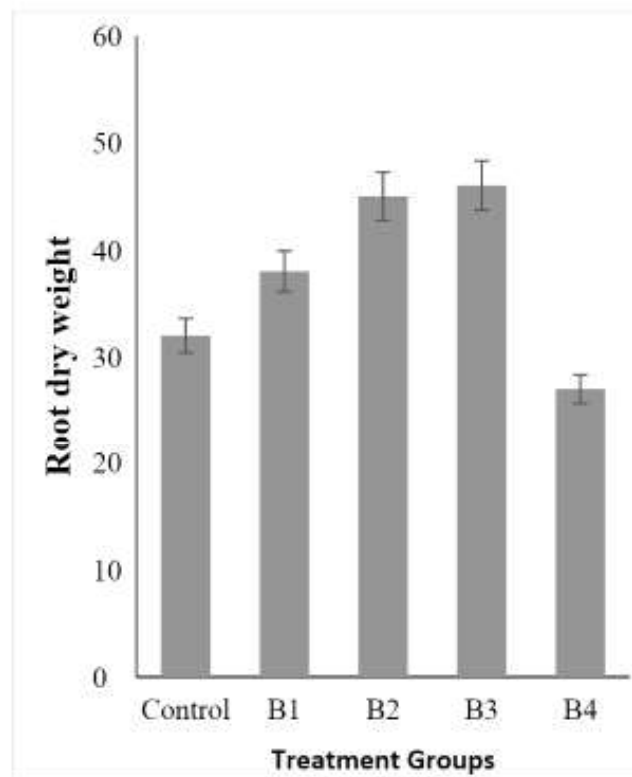


Fig. 10: Root dry weight against different experimental groups

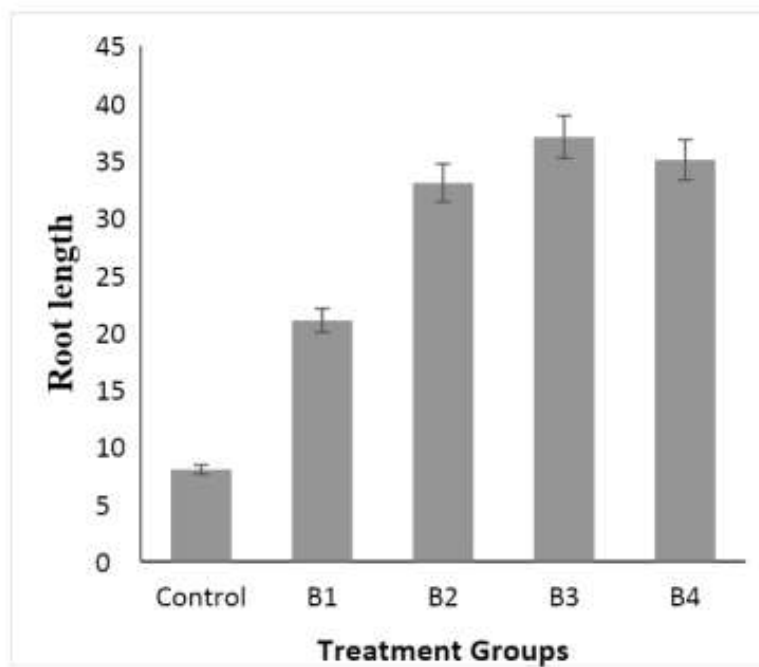


Fig. 11: Root length against different experimental groups

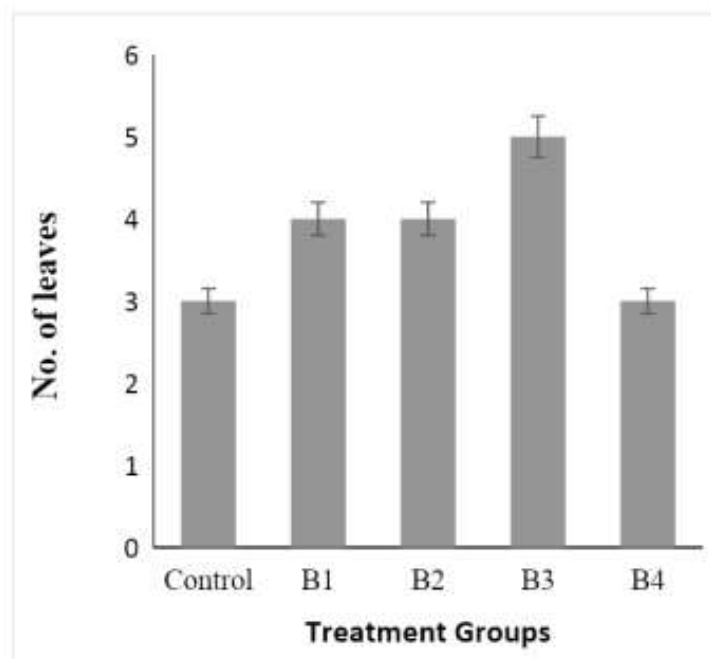


Fig. 12: No. of leaves against different experimental groups

DISCUSSION

Biochar significantly enhance the plant growth, shoot and root dry weight under moisture levels as compared to untreated ones and at lower straw rate. Biochar effectively enhance the plant growth at the highest rate of their application (Mohawesh et al., 2018). Kasak et al. (2018) showed two possible reasons, improved bacterial inhabitants and little amounts of biochar substances enhance plant evolution (hormesis).

Biochar synthesized from coir in composted green manure improved root and shoot length, plant height, root fresh and dry weight of *Calathea insignis* compared with the controlled plant (Huang et al., 2019). After biochar combination, increased plant growth lead to improved nutrients accessibility and increase water retention. Pumpkin (*Cucurbita pepo*) and muskmelon (*Cucumis melo*) increased plant height by 50% sugarcane biochar composite compared to controlled plant. (Yu et al., 2019) also revealed that biochar composite with the rest being perlite enhance dry weight and evolution of Chinese cabbage. Biochar application mainly improved the plant evolution and crop than particular modifications without BC enhance dry weights of shoot and root. Application of biochar soil enhance the development of wheat seedling under the cadmium stressful state. Biochar increased the plant height due to development in soil properties (Rizwan et al., 2018). Enhancement in wheat development with BC application under

cadmium stress due to their lower concentrations in plant. In our study, biochar composite also enhances the dry and fresh weight of root and shoot. Plant height, number of leaves, and leaf area.

Biochar application has multiple effects on the soil chemistry, ultimately affecting the growth of the crops. The composition and application of biochar depends on the biotic and abiotic environmental factors. This carbonaceous material is nutritive and plays significant contribution in regaining lost nutrients from the soil. Different concentrations of biochar have marked influence on the growth of wheat seedlings. Soil pH have greater impact on the surface features of biochar that alters the adhering capacity of this complex material (Mohamed et al., 2017). Moreover, observed a strong correlation between the biochar levels and growth. Just like biochar efficiency in stimulating growth in numerous crops, our studies also depicted increase in growth attributes of wheat seedlings. Biochar amended soil enhances the crop productivity (Novak et al., 2016).

Biochar amended soil increases crop productivity in both acidic and neutral soil. The statistical analysis showed increase in crop productivity with the increasing concentrations of biochar (Cornelissen et al., 2018). Application of biochar composites have significant effect on the growth of wheat seedlings (Vijayaraghavan, 2019). During the current study, increasing concentrations of biochar enhanced the morphological attributes of wheat seedlings. Crop

improvement was noticed in plants supplemented with biochar amended soil due to increase of water holding capacity and nutritive capacity of soil (Abbas et al., 2018). Higher concentrations of biochar enhanced the formation of roots and reduces the leaching of nitrogen and phosphorous from the soil (Karhu et al., 2018). Furthermore, the present study elicited improvement in morphological parameters; a sign of efficient growth in biochar-amended soil.

The soil deficient in nutrients results in decrease in growth of agronomic crops. This nutrient deficiency can be mitigated by application of biochar in soil (Yu et al., 2019). Zheng et al. (2018) reported that biochar application along with fertilizer can enhance the growth of numerous crops. The application of biochar to soil have multiple effects on the growth of crops. It is important to understand the soil chemistry for better applied approach of biochar. In our study, biochar along with different nutritive compounds increased the growth of supplemented wheat seedlings.

CONCLUSION

Recently, biochar has emerged as an effective nutritive source that plays noteworthy contribution in improving growth of diverse crops. Biochar may be added in combination with different nutrients. Biochar-composites are effective, perpetual and eco-friendly additives that have potential to enhance the growth of different crops. Addition of

biochar augments the porosity of soil; thereby improving the soil properties. During the current study, biochar composites were added in Petri plates and pot to appraise its potential to enhance growth of wheat seedlings. Improvement of growth in wheat seedling on exposure to biochar composites was concentration-dependent. Optimum concentration of biochar-composite enhanced the morphology of supplemented wheat seedlings in Petri plates and pots. Different morphological parameters improved in wheat seedlings on exposure to biochar composites i.e., shoot fresh weight, shoot dry weight, root fresh weight, root dry weight, number of leaves per plant, plant height, leaf area and root length. It is concluded that biochar composites may be supplemented in soil to enhance the growth in different crops. Moreover, the utilization of this carbonaceous material is an operative and eco-friendly techniques that is effective for growth of wheat seedling.

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Bio-remedial Properties of *Moringa oleifera* Seed Powder against Contaminated Water

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ABSTRACT: *Urbanization and various human activities are adversely affecting freshwater resources. The lowering of ground water table every day and drained water contamination is an alarming threat to potable water for the Lahore city. So, there is need to improve water quality and bioremediation is found as very useful application for reclamation of polluted water. In the current study 4g seeds of Moringa oleifera were used for treatment of one liter sewerage/polluted water. The water samples were collected by completely randomized design (CRM) from six primary drains [Cantt. Drain (CNTD), Sattu Katla Drain (SKD), Lower Chotta Ravi Drain (LCRD), Upper Chotta Ravi Drain (UCRD), Shalimar Escape Channel Drain (SECD) and Shahdra Drain (SHD)] and four houses located within 500m distance around each drain. The houses were named as H1, H2, H3 and H4. Samples were examined for physico-chemical parameters including turbidity, pH, Ts, Salinity (%), TSS, TDS, COD, BOD and heavy metals (Pb, Cu, Cd and Ca) before and after adding Moringa oleifera. Turbidity decreased by 95% after treatment with Moringa oleifera Seed Powder. After treatment with Moringa seeds, there was an increase in values of pH, NaCl (%), TS (mg/l), TSS (mg/l), BOD, Electrical conductivity and TDS (ppm) of all collected samples. Moreover COD of all samples was reduced which is a good measure for water pollution. Mean levels of trace metals were found to be reduced after treatment with MOSP at all sites which strongly advocates for the use of this product for reclamation of polluted waters.*

Keywords: *Primary drains, Sewerage water, Moringa oleifera seeds, bioremediation, heavy metal*

INTRODUCTION

Depletion of the water resources such as streams, lakes, rivers and ground water occur due to industrialization, urbanization and other agricultural activities and anthropometric procedures. Agrochemical wastes, hospital wastes, toxic metals and pharmaceuticals contaminate the water reservoirs. The open drainage system in many areas of the country that are urbanized results in poor quality of water. Ground pollution spreads when the household wastes are allowed to flow in the river water which is recharging the groundwater. The function of natural influences and human actions affect the quality of ground water and surface water (Baxter et al., 2001).

A large number of municipal drains are present in Lahore which is the 8th most polluted urban area of Pakistan, and stands at the 54th position globally. Being the central hub of business, communities over-occupy Lahore and many industries with harmful waste encircle it. The weak enforcement of environmental laws has led to dumping of unchecked harmful industrial waste into municipal drains and other water resources of the city (Sanchez-Triana et al., 2014).

Trace metals contamination is a big threat at state and territory level. In all living organisms, trace metals are present and they play an important role. Essential metals can divert into toxic while non-essential elements also have fatal effects

on the body as they move in (Azimi et al., 2003). In sewerage drains trace metals such as zinc, copper, cobalt and lead are a big source of environmental pollutants and abundantly determined in the water (Malik et al., 2009). Sewage water being entered into the drinking water by infiltration is the most widespread danger to the human species. The polluted water causes a huge number of different kinds of diseases. The presence of fecal coliform bacteria is the main indicator for fecal pollution and is effective in testing of water resources (Anwar et al., 2010).

Bioremediation is a wastewater management technique that facilitates removal or neutralization of pollutants from a contaminated site. Medicinal plants have proved valuable as natural resources with pharmacological and nutritional properties aiding humans in treatment of wastewater (Lea, 2010). The bioremedial property of *Moringa oleifera* (also called as Saijan or drumstick) is time tested as its fruit and leaf are consumed as diet and the bark has healing properties as recorded in ancient medicine. *Moringa oleifera* seeds have also been reported to have bioremedial property (George et al., 2016).

Important medicinal properties of *Moringa oleifera* include antipyretic, antiepileptic, anti-inflammatory, antiulcerative, antihypertensive, cholesterol lowering, antioxidant, anti-diabetic, hepatoprotective, antibacterial and antifungal activities. In addition, *M.*

oleifera seed possesses water-purifying powers. They are known to be anti-helminthic, antibiotic, detoxifiers, immune builders and have been used to treat malaria and it can also be used as a less expensive bio-absorbent for the removal of heavy metals. In Sudan, powdered seeds of *M. oleifera* have been used in water purification (Jamil et al., 2007). Similarly Basra et al. (2014) also reported the sewerage water purification ability of *M. oleifera* seeds. They found its seed powder effective tool for reduction of electrical conductivity and pH maintenance.

Heavy metals are very common and harmful contaminant of water. Their removal is very important for better health of human beings. Many conventional methods for removal of water pollutants are ion exchange, reverse osmosis, solvent extraction, chemical precipitation and sedimentation. These methods are not cost effective and have disadvantages. In current study *M. oleifera* seeds are used as

an alternative to substitute the conventional water purification methods. In this study its effect is also monitored on different physic-chemical parameters like (Ts, TSS, pH, EC, COD, BOD and Salinity) of sewerage water collected from six primary drains of Lahore city.

MATERIALS AND METHODS

Sampling Sites

Water samples were collected from six primary drains as shown in (Fig. 1) and from 24 houses among them 04 were situated within 500m distance of sewerage drains as given below:

1. Cantt Drain (CNTD)
2. Sattu Katla Drain (SKD)
3. Lower Chotta Ravi Drain (LCRD)
4. Upper Chotta Ravi Drain (UCRD)
5. Shalimar Escape Channel Drain (SECD)
6. Shahdra Drain (SHD)

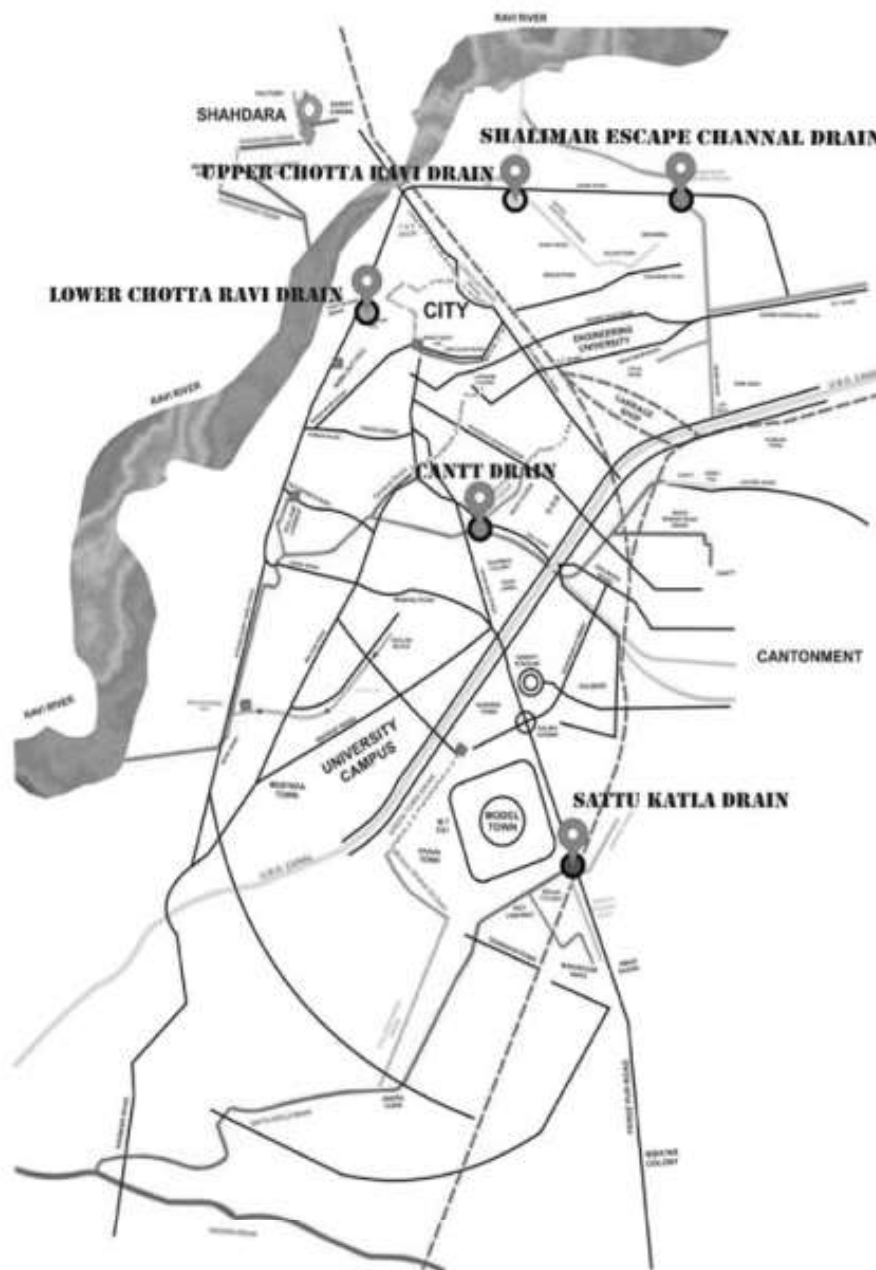


Fig. 1: Six primary drain sites of Lahore Credit: WASA, Lahore

Procurement and Preparation of *Moringa oleifera* Seed Powder (MOSP)

The fresh seedpods of *Moringa oleifera* were collected from Institute of Agricultural Sciences, University of the

Punjab, Lahore. The seeds were removed from the pod and grinded to make powder that was stored in clean plastic box. As per literature *Moringa oleifera* seed are harmless to human beings and safe treatment of waste water (Madrona et al.,

2012). Its seeds are possessing natural coagulation property that is parallel and even better than alum (Aluminium Sulphate) (Arnoldson et al., 2008).

Collection of Samples

A total of 02 liters water was collected in completely sterilized plastic bottles from selected sites CRM (Complete Randomized Method) used for sample collection. At every site sample water was divided into two bottles of 01 liter capacity marked as treated (with *Moringa oleifera* seed powder) and untreated (without *Moringa oleifera* seed powder). The sampled water was tested for followings:-

- i. Physico-chemical parameters
- ii. Removal of heavy metals

Preparation of Sample

In water sample labeled as treated 04 gms of MOSP was added while nothing was added in untreated water.

i. Physico-chemical Parameters

The color of treated and untreated sampled water was observed at sites. Color of treated water was observed after every 30mins till six hours after addition of MOSP. The turbidity, pH, electrical conductivity, salinity, Ts, TDS and TSS were measured with Bench top multi-meter, Germany after 48hours of treatment with MOSP. The BOD (biochemical oxygen demand) and COD (chemical oxygen demand) were measured according to Suhartini et al. (2013) and Forster, (2000).

ii. Heavy Metals Analysis

A total of four heavy metals Copper (Cu), Chromium (Cr), Cadmium (Cd), and Lead (Pb) were selected to test the bioremedial property of MOSP. The heavy metals analysis of above selected metals for both treated and untreated samples was performed by chemical digestion methods followed by (AAS) Atomic Absorption Spectrophotometry (Perkin-Elmer guide for atomic absorption, 1996) and (Rehman and Husnain, 2012).

RESULTS AND DISCUSSION

i. Effect of *Moringa oleifera* Seed Powder (MOSP) on Physico-Chemical Parameters of Sewerage Water

A slight increase in pH levels was observed in water samples of six selected primary drains treated with MOSP after 48 as shown in Table-1 but the results given by Shahzad et al. (2013) are opposite to current findings as they found significant decrease in pH levels of non-treated sewerage water. As a matter of fact, seeds of *Moringa oleifera* contain water solvable proteins that support coagulation and retain adsorption power at pH of 5-8 (Vieira et al., 2010). The amino acids existent in moringa seeds proteins take water protons and release hydroxyl group ions to make solution basic due to its buffering capacity (Ndabigengesere et al., 1995; Ndabigengesere and Narasiah, 1998). On the other hand Chushan et al. (2014), Shan et al. (2017) reported that MOSP has

not affected the pH of waste water while Delelegn et al., (2018) reported a slight decrease in pH.

As far as the turbidity is concerned a significant decrease has been found after 48 hours treatment of 1litre sewerage water with 4g of MOSP at every drain but the turbidity of CNTD reduced from 83 ± 0.07 ppm to 2.5 ± 0.06 ppm. The results of current research are in agreement with findings of Suhartini et al. (2013). According to them the proportions of turbidity in water reduced significantly with increase of contact time. Narender et al. (2019) also observed that *Moringa oleifera* seeds have ability to reduce the turbidity as natural coagulant.

Conversely, the conductivity, salinity and TDS values of SW after 48hrs treatment with MOSP were increased. For example the EC of SECD was 1132 ± 0.01 μ S before treatment and after treatment it increased up to 2943 ± 1.00 μ S as given in Table-1 same is the case of salinity and TDS. The maximum TDS was raised from 647 ± 0.03 to 1483 ± 0.03 at SKD and maximum increase of Salinity from 2.7 ± 0.00 to 6.1 ± 0.04 was recorded in SW samples of LCRD. The increase in three parameters of SW was also recorded by Shan et al. (2017). The results of current work were in contrast with findings of Amagloh and Benang (2009) as they had revealed the decrease in value of conductivity on rising of MO seed cake concentration. Conversely, the earlier report by Arnoldsson et al. (2004) showed that the TDS and conductivity values had not been influenced with *Moringa*

oleifera coagulant. Hence, the findings of the SW sample were alike to the results of (Arnoldsson et al., 2004). Chushan et al. (2014) also reported increase in values of Conductivity, TDS and Salinity from 347 ± 0.00 to 390 ± 0.00 μ S, 307 ± 0.00 to 400 ± 0.00 ppm and 353 ± 0.00 to 410 ± 0.00 respectively after treatment. Salinity can carry an electrical charge due to that water may generate electricity thus; salinity is directly proportional to the conductivity (Ellis, 2004). The MOSP treatment did not alter the values of Ts and TSS in tested SW samples. Generally the Total Solids (Ts) is equals to TSS (total suspended solids) and TDS (Total dissolve solids) and sewerage water is hub of all kinds of solids like fixed solids, settleable solids, total dissolve solids, total solids, total suspended solids and volatiles solids. The values recorded for Ts and TSS were high after 48hrs treatment of MOSP (Table 1). Over all a decreasing trend in COD (mg/l) values was observed at all selected sewerage drains which is contrary to Arnoldsson et al. (2004) as they reported a dramatic increase in COD values after treatment with *Moringa oleifera* seed cake. However these findings are in agreement with Bhuptawat et al. (2007) reporting 64% reduction in COD value after treatment.

The COD (chemical oxygen demand) concentration is an indirectly to determine quantity of organic compounds/pollutants in sample water. In current case the decrease in COD can

support the bioremediation property of *Moringa oleifera* seed for removal of organic water pollutants

In current research work the BOD levels in all SW samples were increased after treatment of MOSP while maximum increase was noticed at LCRD 127 ± 0.01 to 526 ± 0.12 (mg/l). BOD (Biochemical Oxygen Demand) is an important tool to access the effect of municipal wastewater on receiving environment (Irenosen et al., 2012), the increased values are basically due to municipal and industrial wastes. The natural/organic compound in MOSP may be the reason for increase of BOD. The site LCRD is considered the highest polluted site towing to abundant receiving of wastewater from all over the city. These results are not in agreement with and Sajidu et al. (2005) but the observations are similar to findings of (Chushan et al., 2014; Shan et al., 2011).

ii. Effect of *Moringa oleifera* Seed Powder (MOSP) on Heavy Metals Concentration of Selected Samples

The effect of *Moringa oleifera* Seed Powder was also recorded heavy metals concentration of selected 30 water samples. Among these samples 06 were of sewerage water from selected primary drains while 24 were from four

houses selected within 500m distance from every drain. 02 liter of samples were collected in two different bottles of 1000ml capacity. The water with 04gms MOSP were labeled as treated and water without MOSP were labeled as untreated. The values of selected heavy metals Cr, Cu, Cd, and Pb obtained by atomic absorption spectrophotometry is given below with discussion

Highest concentration 6.64 ± 0.10 ppm of Chromium was reported in SW sample collected from UCRD that decreased to 4.41 ± 0.05 ppm after 48hrs treatment of MOSP (Table 2). The values in table-2 are indicating that MOSP is effective in reduction of Cr levels in all 30 samples of water. The maximum reduction in concentration 0.17 ± 0.00 ppm of Chromium was found in H4 within 500m of SHD. The trend of Cr concentration was $6.64 > 3.41 > 2.94 > 2.56 > 1.94 > 1.83$ at UCRD, SHD, SECD, CNTD, LCRD and SKD respectively. Ravikumar and Sheeja, (2013) also reported 95 % reduction for copper >93 % for lead >76 % for cadmium >70 % for chromium in water samples treated with *Moringa oleifera* seeds followed by double filtration.

Table 1: Effect of (MOSP) on Physico-Chemical Parameters of Sewerage Water

Parameters	SKD		UCRD		CNTD		LCRD		SECD		SHD	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post
1 pH	7.0±0.09	7.45±0.03	6.95±2.02	8.23±0.07	7.12±0.00	8.93±0.03	7.21±0.09	7.65±0.13	7.3±0.12	8.99±0.01	6.91±0.02	7.21±0.00
2 Turbidity (NTU)	77.25±0.01	6.07±0.00	57±0.08	4±0.02	83±0.07	2.5±0.06	57±0.07	4±0.04	168±0.212	3±0.30	93±0.07	5±0.00
3 Electrical Conductivity (µS)	432±0.04	1156±0.02	1040±0.01	1745±0.07	1375±1.48	1509±0.06	237±0.03	421±0.01	1132±0.01	2943±1.00	116±0.00	272±0.00
4 NaCl%	0.8±0.07	2.1±0.03	1.8±0.03	3.3±0.00	2.3±0.00	5.8±0.01	2.7±0.00	6.1±0.04	2.1±0.42	5.5±1.41	3.4±0.21	5.7±0.11
5 Ts (mg/L)	62±2.00	240±0.17	53±0.08	90±0.08	89±0.01	160±0.03	64±0.07	101±0.03	130±0.13	210±0.0	86±0.00	116±0.07
6 TDS(ppm)	647±0.03	1483±0.03	617±0.0	1155±0.08	524±0.05	934±0.0	429±0.07	566±0.07	216±0.00	580±0.03	319±0.01	427±0.07
7 TSS(mg/L)	4.2±0.00	59±0.07	5.3±0.02	72.1±0.07	8.9±0.00	25±0.00	4.7±0.03	622±0.03	13±2.38	129±0.07	11±0.04	87±0.03
8 BOD(mg/L)	514±0.03	617±0.02	63.2±0.07	297±0.00	173.4±01.40	420±0.00	127±0.0	526±0.12	19±0.00	150±01.06	127±0.06	512±0.0
9 COD(mg/L)	30±0.00	16±0.00	30±1.06	11±0.00	20±2.00	18±0.08	19±0.00	7±0.00	10±0.00	15±0.00	21±0.00	11±0.02

Table 2: Effect of MOSP on Cr Concentration (PPM) in water samples of selected sites

Sites	SKD		UCRD		CNTD		LCRD		SECD		SHD	
	Untreated Cr PPM	Treated Cr PPM	Untreated Cr PPM	Treated Cr PPM	Untreated Cr PPM	Treated Cr PPM	Untreated Cr PPM	Treated Cr PPM	Untreated Cr PPM	Treated Cr PPM	Untreated Cr PPM	Treated Cr PPM
SW	1.83±0.00	0.97±0.12	6.64±0.10	4.41±0.05	1.90±0.00	1.23±0.02	1.87±0.04	1.12±1.00	2.94±0.00	1.19±0.04	3.41±0.06	2.10±0.03
H1	1.51±0.00	1.17±0.03	5.21±0.00	4.11±0.10	2.01±0.03	1.97±1.00	1.94±0.00	1.22±0.31	2.512±0.05	2.11±0.09	3.22±0.01	2.91±0.06
H2	1.72±0.00	1.07±0.01	4.11±0.08	3.87±0.00	2.47±0.05	1.81±0.09	1.22±0.00	1.01±0.03	2.12±0.02	2.01±0.00	3.10±0.00	2.56±0.09
H3	1.63±0.00	1.09±0.03	3.80±0.02	3.01±0.00	2.56±0.07	1.62±0.02	1.19±0.06	1.04±0.08	2.17±0.02	1.89±0.00	2.43±0.00	0.44±0.00
H4	1.45±0.00	1.01±0.08	2.32±0.00	1.02±0.15	2.23±0.08	1.80±0.03	1.24±0.09	1.19±0.09	1.87±0.00	1.56±0.02	1.83±0.10	0.17±0.00

Abbreviations: * SW: (sewage water), H1: (House-1), H2: (House-2), H3: (House-3), H4: (House-4)

** All housed were selected within 500m distances of Sewerage Drains

Table 3: Effect of MOSP on Cu Concentration in water samples of selected sites

Sites	SKD		UCRD		CNTD		LCRD		SECD		SHID	
	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM
SW	2.21±0.05	1.87±0.08	0.82±0.11	0.01±0.00	0.29±0.00	0.19±0.07	0.25±0.00	0.17±0.00	0.24±0.03	0.18±0.05	0.28±0.11	0.22±0.00
H1	2.01±0.09	1.12±0.06	0.81±0.21	0.06±0.00	0.25±0.03	0.21±0.05	0.29±0.01	0.19±0.04	0.31±0.00	0.29±0.00	0.50±0.02	0.31±0.01
H2	1.98±0.11	1.09±0.00	0.77±0.08	0.55±0.06	0.23±0.07	0.10±0.02	0.30±0.02	0.20±0.12	0.38±0.01	0.27±0.03	0.42±0.09	0.30±0.03
H3	1.71±0.06	1.01±0.00	0.65±0.06	0.09±0.03	0.19±0.12	0.14±0.01	0.30±0.02	0.19±0.04	0.49±0.15	0.31±0.03	0.65±0.02	0.41±0.09
H4	1.42±0.08	0.97±0.00	0.56±0.02	0.47±0.01	0.13±0.05	0.11±0.00	0.31±0.00	0.20±0.03	0.50±0.09	0.42±0.01	0.32±0.00	0.21±0.19

Abbreviations: * SW: (sewage water), H1: (House-1), H2: (House-2), H3: (House-3), H4: (House-4)

** All housed were selected within 500m distances of Sewerage Drains

Table 4: Effect of MOSP on Cd Concentration in water samples of selected sites

Sites	SKD		UCRD		CNTD		LCRD		SECD		SHID	
	Untreated Cd PPM	Treated Cd PPM	Untreated Cd PPM	Treated Cd PPM	Untreated CudPPM	Treated Cd PPM	Untreated Cd PPM	Treated Cd PPM	Untreated Cd PPM	Treated Cud PPM	Untreated Cd PPM	Treated Cud PPM
SW	1.83±0.13	0.97±0.03	6.64±0.10	4.41±0.02	1.94±0.00	1.23±0.03	1.87±0.02	1.12±0.13	2.94±0.08	1.19±0.02	3.41±0.08	2.10±0.02
H1	1.51±0.00	1.17±0.09	5.21±0.01	4.11±0.03	2.01±0.19	1.97±0.05	1.94±0.01	1.22±0.21	2.51±0.06	2.11±0.02	3.22±0.00	2.91±0.01
H2	1.72±0.23	1.07±0.01	4.11±0.01	3.87±0.00	2.47±0.06	1.81±0.05	1.22±0.03	1.01±0.00	2.12±0.04	2.01±0.05	3.10±0.06	2.56±0.00
H3	1.63±0.11	1.09±0.02	3.80±0.02	3.01±0.07	2.56±0.03	1.62±0.01	1.19±0.11	1.00±0.00	2.17±0.02	1.89±0.07	2.43±0.05	2.44±0.00
H4	1.45±0.00	1.01±0.08	2.32±0.03	1.02±0.06	2.23±0.00	1.89±0.01	1.24±0.00	1.19±0.00	1.08±0.01	1.56±0.06	1.83±0.03	1.00±0.10

Abbreviations: * SW (sewerage water), H1: (House-1), H2: (House-2), H3: (House-3), H4: (House-4)

** All housed were selected within 500m distances of Sewerage Drains

Table 5: Effect of MOSP on Pb Concentration in water samples of selected sites

Sites	SKD		UCRD		CNTD		LCRD		SECD		SHID	
	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM	Untreated Cu PPM	Treated Cu PPM
SW	1.39±0.10	0.97±0.11	1.43±0.00	0.002±0.06	0.13±0.12	1.10±0.03	2.49±0.03	1.21±0.13	10.0±0.00	6.20±0.00	4.84±0.05	2.34±0.00
H1	1.41±0.01	1.21±0.10	1.22±0.00	0.09±0.15	1.01±0.10	0.07±0.00	2.62±0.03	1.09±0.00	9.21±0.07	7.00±0.00	5.61±0.16	4.23±0.00
H2	1.52±0.01	1.32±0.09	1.01±0.08	0.09±0.05	1.17±0.12	1.00±0.14	3.10±0.00	2.94±0.00	9.02±0.05	7.91±0.03	5.72±0.02	4.62±0.09
H3	1.33±0.00	0.11±0.07	0.94±0.09	0.81±0.02	1.19±0.00	1.00±0.00	4.22±0.06	3.76±0.00	8.04±0.03	7.41±0.00	5.51±0.04	4.99±0.07
H4	1.79±0.00	1.00±0.04	0.87±0.09	0.01±0.00	2.00±0.03	1.00±0.02	3.80±0.05	1.11±0.00	6.63±0.02	5.91±0.10	6.60±0.06	5.90±0.05

Abbreviations: * SW (sewerage water), H1: (House-1), H2: (House-2), H3: (House-3), H4: (House-4)

** All housed were selected within 500m distances of Sewerage Drains

The 48hrs treatment of MOSP proved very useful for removal of heavy metals from water samples. The maximum concentration of Cu 2.21 ± 0.05 was noted at SKD that was 75% reduced after MOSP treatment (Table 3). These findings are in agreement with Sharpudin et al. (2018) who used CAMOL and CAMOB (Citric Acid modified *Moringa oleifera* leaf and bark) for removal of Cu and Cr. Beltran-Heredia and Sanchez-Martin (2008) had also removed Cu from water sample by use of *Moringa oleifera*.

Cadmium is among toxic heavy metals and serious environmental pollutant. It enters water systems by various natural and human activities. Even a small increase in its concentration may be carcinogenic. In Table: 4 it is obvious that MOSP reduced the Cd concentration after treatment but comparing of data with permissible limit of WHO (2011) indicates that Cd concentration is still higher in all treated or untreated test samples and same were the findings of Idrees et al. (2018). Table-4 is also indicating that underground water table is effected by cadmium toxicity as all 24 samples collected from houses within 500m radius of primary drains are contaminated by cadmium pollution. The maximum limit 6.64 ± 0.10 ppm of Cd was reported in SW of UCRD that had been reduced to 4.41 ± 0.10 ppm after MOSP treatment but still this value is very high. The published data supports bioremediation property of *Moringa oleifera* but in current study the result was not highly effective for

removal Cd. Sharma et al. (2006) also reported better removal of Cd by SMOS (shelled *Moringa oleifera* seeds). As noticed in current study removal of Cd by MOSP is 20-40% while Sajidu et al. (2005) has reported 48% reduction in Cd concentration but contrarily Meneghel et al. (2013) reported 98% removal of Cd by *Moringa* seed cake.

Lead is another very toxic heavy metal. The highest concentration of Lead (Pb) 10.0 ± 0.00 ppm is reported in current study at SECD that was reduced to 6.20 ± 0.00 ppm. Although this decreased vale is still very higher than the permissible limits defined by WHO (2011). The 40% reduction in lead (Pb) concentration is not in agreement with findings of Subramanium et al. (2011) and Chushan et al. (2014) as they reported up to 78.1 and 77.8% removal of Pb respectively and Ali, 2020 has also reported 82.17% reduction in lead (Pb) concentration. In this situation the increase in quantity and contact time of MOSP is recommended.

CONCLUSION

Moriga oleifera is a plant with best phytoremediation properties. Its different parts like leaf, bark, root, flower, seed pod and seeds are used to purify the water of many kinds like groundwater, surface water, polluted water, wastewater and municipal water. In this paper the potential application of *Moringa oleifera* seed as bioremediation to purify the sewage water and water has been studied.

According to current research findings it is found that *Moriga oleifera* seed powder is useful natural tool to improve certain Physico-chemical parameters but not all. However it had very good remedial properties for removal of heavy metal like Cr, Cu, Cd and Pb. On the basis of above it is suggested that *Moringa oleifera* seed may be used as cost effective water purifier and natural adsorbent to remove heavy metals and neutralizing the Physico-chemical parameters.

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Comparison of Menstrual Pain among Dysmenorrhea and Non-Dysmenorrhea University Students

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ABSTRACT: *Dysmenorrhea is a gynecological state of cramps with painful menstruation. In this condition, pain initiates for a short period before or at the beginning of the menstrual cycle and lasts for 1-3 days. A comparative cross-sectional study was carried out at The University of Lahore, to assess the menstrual pain among dysmenorrhea and non-dysmenorrhea university students. A total of 150 menstruating females were divided into two groups: the dysmenorrhea group: 75 females aged between 21 to 30 years complaining of dysmenorrhea and non-dysmenorrhea group: 75 females aged between 21 to 30 years having normal menstruation were enrolled. Data were collected through interview form and participating females were pre-informed regarding the risks and benefits of the study and written informed consent was taken from the participants. Participants were assessed through a pre-tested questionnaire using a non-probability convenient sampling technique and data were analyzed using SPSS version 21.0. Among the non-dysmenorrhea group 55 females were having pain in 1 hour, while in the dysmenorrhea group only 9 females were having pain for 1 hour and 39 females were having pain >3 hours. According to the non-dysmenorrhea group, 9 females were using hot applications whereas in the dysmenorrhea group 34 were used hot applications. Among the non-dysmenorrhea group, 26 females were effected due to menstrual pain and were unable to perform daily activities whereas in the dysmenorrhea group 60 females were affected and unable to perform their daily activities. It was concluded that in dysmenorrhea groups more females were taking painkillers, using hot applications, doing massages and were unable to perform the daily activity as compared to the non-dysmenorrhea group.*

Keywords: *Comparison, menstrual pain, females, non-dysmenorrhea, dysmenorrhea, university students*

INTRODUCTION

Dysmenorrhea is a gynecological state with painful cramps in the uterine origin (Kural et al., 2015). Dysmenorrhea is described by pelvic cramp with pain initiating for a short period before or at the beginning of the menstrual cycle and lasting 1 to 3 days (Unsal et al., 2010). Menstruation has been familiar among the females from the 19th era to now and has been viewed as the new problem (Kordi et al., 2011). In the age of fertility, each female experiences a menstrual cycle for four hundred times (Do Amaral et al., 2005). The 1/7th of a female's life is to be associated with menstruation (Lee et al., 2009). The pain in the menstrual cycle may spread to the lower back or the thigh. Dysmenorrhea may be categorized into minor, modest and severe (Iliyasu et al., 2012). The individual becomes moody and depressed in pain and this disturbs social communication (Iliyasu et al., 2012). In this period, some females and adolescents lose their hunger and they may absent from work or school (Gumanga and Kwame-Aryee, 2012). Dysmenorrhea is categorized: primary or secondary (Iacovides et al., 2015). Primary dysmenorrhea is described by painful cramps in uterine origin that experienced before the start of menstruation or during the menstrual cycle in the lower area of the abdomen with no evidence of the pathology of pelvic (Rapkin, 2012; Hudson, 2007). The beginning of primary dysmenorrhea (PD)

is usually at or just after six to twelve months of the menarche when ovulatory phases are beginning (Shah et al., 2013). Some females are released from this condition after their first baby and other females suffering from this problem until their menopause (Iliyasu et al., 2012). The duration of the distress in the menstrual cycle is mostly eight to seventy-two hours and is mostly related to menstruation (Shah et al., 2013). For example, body mass index (<20 or >30) (Haidari et al., 2011; Roberts et al., 2012), age, low socioeconomic status, alcohol consumption, smoking (Nohara et al., 2010), initial age at menarche, long period of the menstrual cycle, heavy menstrual bleeding, obesity (Gagua et al., 2012), marriage history (Bajalan et al., 2018), history of dysmenorrhea (Tavallaee et al., 2011). Abdul-Razzak et al. (2010) reported a correlation between low consumption of dairy items and the possibility of dysmenorrhea. Rodrigues et al. (2011) revealed that dysmenorrhea is extremely common among young adults and adolescents and is interrelated to school/ academic absenteeism (Rodrigues et al., 2011). Another study showed that almost 35% of the undergraduates in university had acute dysmenorrhea and around 21% had medium to acute PMS. The results also showed that set the intervention programs, such as educational courses, to decrease the harmful impacts of the problem on the undergraduates' productivity and value of life (Al-Dabal et al., 2014). Negi et al. (2018) noticed that PMS, dysmenorrhea

and irregular menstrual cycle were associated with daily activities and eating habits among young females. Dysmenorrhea was correlated with the consumption of junk food (66.10%), and PMS was correlated with lacking daily activities (78.94%). A greater prevalence of medium and severe primary dysmenorrhea in underweight as compared to other obese (OB) groups (Rafique and Al-Sheikh, 2018).

The researcher was aimed to find out the comparison of menstrual pain among dysmenorrhea and non-dysmenorrhea university students. To find out the lifestyle factors associated with dysmenorrhea.

MATERIAL AND METHODS

A comparative cross-sectional study was carried out among 150 menstruating females studying at the University Institute of Diet and Nutritional Sciences, Faculty of Allied Health Sciences, The University of Lahore, Lahore. The sample size was calculated using the WHO formula. Menstruating females, selected through non-probability convenient sampling technique were divided into two groups: non-dysmenorrhea and dysmenorrhea. Among the dysmenorrhea group: 75 females aged between 21 to 30 years complaining of dysmenorrhea and in the non-dysmenorrhea group: 75 females aged between 21 to 30 years having normal menstruation were enrolled. In exclusion criteria: Females with

amenorrhea and females with other institution was not included in this study. Data were collected through interview form and Participating females were informed regarding the risks and benefits of the study and written informed consent was taken from the participants. Data were collected through a pre-tested questionnaire during 4 Months (January to April 2019). SPSS version 23.0 was used for data analysis. Females with amenorrhea, aged below 21 years and above 30 years, non-cooperative ones and females from other institutions were excluded. Ethical approval was taken from the Institutional Review Board (IRB), of The University of Lahore.

RESULTS

The present study was carried out in females with dysmenorrhea and non-dysmenorrhea. A questionnaire was used and information regarding pain one day before the onset of menstruation, pain on the first day and pain on the second day, changes in daily activities, etc. It was noticed during the study in dysmenorrhea group 21 were experiencing pain one day before the onset of menstruation whereas only 3 were having pain one day before the onset of menstruation in non-dysmenorrhea group 3. It was also recorded that the total duration of pain in hours per day in the non-dysmenorrhea group was 12 females having no pain while 55 females were having pain in 1 hour (Table 1).

Table 1: Prevalence of pattern of pain in dysmenorrhea and Non-dysmenorrhea group

Sr No.	Groups	Pattern of pain		
		Pain one day before the onset of menstruation	Pain on the first day	Pain on the second day
1.	Non-Dysmenorrhea	03	20	11
2.	Dysmenorrhea	21	53	13
3.	Total	24	73	22

9 females were having pain in 1 hour in the dysmenorrhea group, 15 females were having pain in 2 hours, 12 were having pain in 3 hours and 39 females were having pain in >3 hours. It was also noticed that 12 females didn't

complain of pain at the onset of menstruation in the non-dysmenorrhea group. Whereas 55 reported pain for 1 hour and no one complained of pain for 2, 3, or >3 hours of pain (Table 2).

Table 2: Distribution according to pain duration in dysmenorrhea and non-dysmenorrhea group

Sr No.	Group	The total duration of pain in hours per day					Total
		0	1	2	3	>3	
1.	Non-dysmenorrhea	12	55	0	0	0	75
2.	Dysmenorrhea	0	9	15	12	39	75
3.	Total	12	64	15	12	39	150

It was also reported in the non-dysmenorrhea group 26 were having disturbed daily activities during menstruation whereas in the dysmenorrhea group 60 were having disturbed daily activities (Table 3)

Table 3: Effect of menstrual pain on daily activities in dysmenorrhea and non-dysmenorrhea group

Sr No.	Group	Effect of menstrual pain on daily activities		Total
		No	Yes	
1.	Non-dysmenorrhea	49	26	75
2.	Dysmenorrhea	15	60	75
3.	Total	64	86	150

According to Fig. 1, in dysmenorrhea group 11 were experiencing pain in the lower abdomen only while in non-dysmenorrhea group 25 were experiencing pain in the lower abdomen only. Among the dysmenorrhea group, 21 were experiencing pain in the lower abdomen and back only whereas in the non-dysmenorrhea group 10 were having pain in the lower abdomen and

back only. In the dysmenorrhea group, 44 were having pain in the lower abdomen, back and legs whereas in the non-dysmenorrhea group 16 were having pain in the lower abdomen, back and legs. And in dysmenorrhea group 2 were experiencing pain in other body parts whereas in non-dysmenorrhea group 14 were experiencing pain in other body parts.

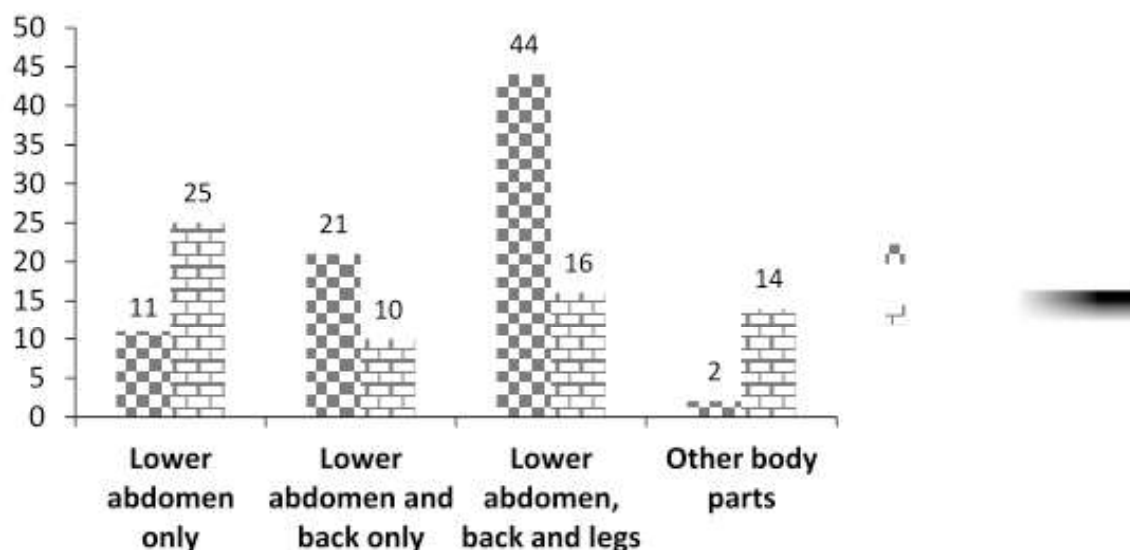


Fig. 1: Pattern of pain among participants in different body parts of Dysmenorrhea and Non-dysmenorrhea group

Fig. 2 showed that in the dysmenorrhea group 34 were using hot applications whereas in the non-dysmenorrhea group only 9 were using hot applications. Among dysmenorrhea group 6 was doing massage whereas among the non-dysmenorrhea group no one was doing massage. In the dysmenorrhea group, 45 were doing bed

rest while in the non-dysmenorrhea group 27 were doing bed rest, during menstruation. Among the dysmenorrhea group, 6 were taking any other precautions whereas in the non-dysmenorrhea group only 2 were taking other precautions. In non-dysmenorrhea groups, no one was taking medicines while in the dysmenorrhea group 18 were taking medicines.

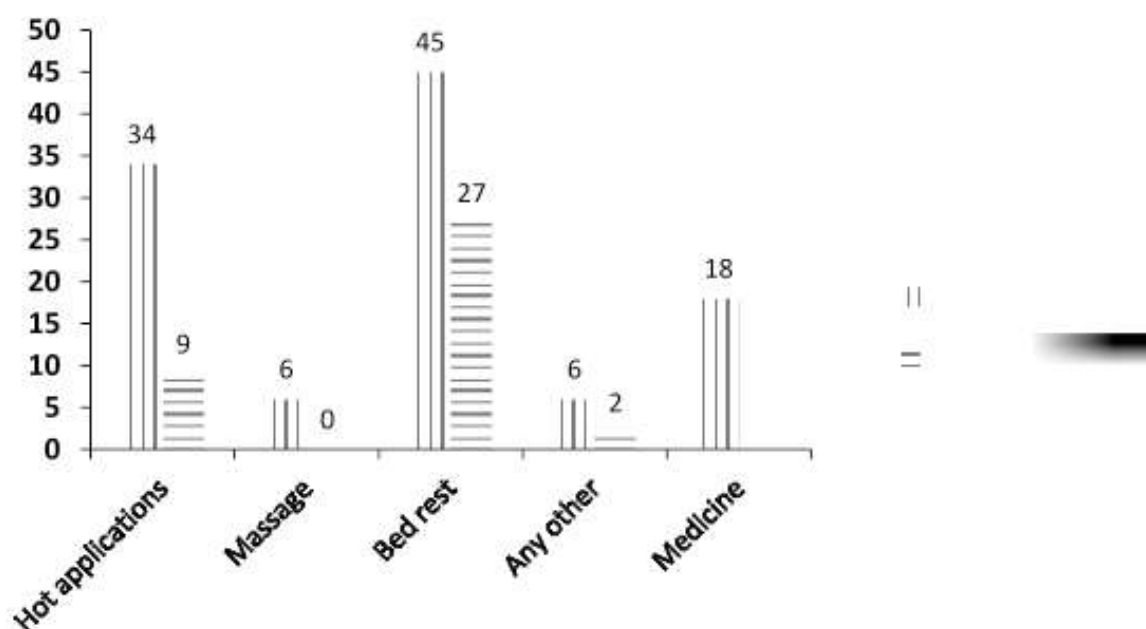


Fig. 2: Distribution of participants regarding treatments during menstruation in dysmenorrhea and non-dysmenorrhea group

DISCUSSION

Results of the current study revealed that in non-dysmenorrhea group 3 patients were experienced early pain before the onset of menstruation whereas in dysmenorrhea group 21 were experienced pain early pain before the onset of menstruation. A similar study by Abdul-Razzak et al. (2010) showed that 87.4% of females with symptoms of pain

starting a few days before menstruation. In the current study, in the non-dysmenorrhea group, 12 females were having no pain and 55 females were having pain in 1 hour, while in the dysmenorrhea group 9 females were having pain in 1 hour, 15 females were having pain in 2 hours, 12 were having pain in 3 hours and 39 females were having pain in >3 hours. Similar results

were revealed in a study and 34.1% females frequently experienced the pain 48.7% were sometimes and 17.2% rarely (Hirata et al., 2002). In another study 69.60% were having minor pain, as compared to 30.40% were having medium and severe pain in the menstrual cycle (Zurawiecka and Wronka, 2018). The results of the current study showed that among non-dysmenorrhea groups, no one was taken medicines while among the dysmenorrhea group 18 were taken medicines. In the study of Rodrigues et al., out of 48 females, 135 were using therapeutic methods to get rid of the pain of dysmenorrhea. Most commonly medications were used to get relief from pain to contain oral tablets (37.0%) and NSAID's (38.5%) (Rodrigues et al., 2011). In 2014, the study was performed which showed 57% of females were using analgesics to get rid of pain (Al-Dabal et al., 2014). Similar findings were observed by Rafique and Al-Sheikh during 2018, 55.8% of the undergraduates were relieving relief from pain by using non-steroidal anti-inflammatory drugs (NSAIDs) (Rafique and Al-Sheikh, 2018). Findings of the current study revealed that in non-dysmenorrhea group 26 were the effect of menstrual pain on daily activities whereas in the dysmenorrhea group 60 were the effect of menstrual pain on daily activities. In the study of Rodrigues et al., during 2011 results showed that 65.7% were showed limits in their daily living due to pain (Rodrigues et al., 2011). Similar results were also found by Rafique and Al-Sheikh (2018) 54.5% of

the undergraduates were an effect on their physical activities. In another study 78.94% of students with PMS were lacking daily activities (Al-Dabal et al., 2014; Negi et al., 2018).

CONCLUSIONS

It was concluded that females with dysmenorrhea were having more pain duration and intensity in the menstrual cycle and were taking pain killers medicines, using more hot applications, doing massage and were unable to perform daily activities in menstruation as compared to the non-dysmenorrhea group, among which female was not usually doing such activities in menstruation.

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CONFLICTS OF INTEREST

There is no conflict of interest to declare.

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Thalassemia: Current Situation in Pakistan

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ABSTRACT: *Thalassemia, inherited genetic disorder is among the main medical issues facing Pakistan like many other countries of the world. The main factor of thalassemia prevalence with frequent rate in Pakistan is consanguineous marriages causes misery not only to the patients but for their families also. The current treatment strategies such as blood transfusion, chelation therapy (Fe), bone marrow and stem cell transplantation are not compatible due to high iron load, development of alloantibodies, arthritis and severe agranulocytosis like side effects. In developed countries regular screening of pregnant women is done to control the prevalence of Beta-thalassemia but in Pakistan genetic counselling and parental diagnosis resources are limited to control the birth rate. Additionally, in the absence of coherent policy at national level creates more pressure on the health care system of the country. Thalassemia in Pakistan is managed by traditional diagnostic methods, such as chromatography, Hb electrophoresis and ARMS-PCR. The ARMS-PCR can only detect the commonly known mutations while the other ethnic specific, novel mutations remain obscure. Next-generation sequencing (NGS) offers advantages over standard PCR, including higher sensitivity and the ability to identify novel mutations. Health system and cultural issues creates hurdles to implement the policies of public health genomics in Pakistan. So, the government of Pakistan must take some measure to ensure the health quality of human and also make short and long term policies to overcome this genetic disorder.*

Keywords: *Thalassemia, Inherited genetic disorder, Pakistan, Consanguineous marriages, Health system and Policies*

INTRODUCTION

Thalassemia, a Greek word meaning “sea blood” is autosomal recessive disorder caused by less hemoglobin (Hb) production due to the presence of defective gene for the synthesis of alpha and beta globulin chain (Shakeel et al., 2016). Globally, it is the second most common hemoglobinopathy after sickle cell disease (Khalid et al., 2019). Pakistan unfortunately lies among the most thalassemia burdened countries of world and have high prevalence rate of alpha and beta type among the population (Zaheer et al., 2020). Alpha- thalassemia is caused by reduction of alpha-globulin chain of hemoglobin that leads to excess of beta-chains which form Hb Bart’s (Khan et al., 2015). Cooley and Lee in 1925 first characterized the inherited disorder of Beta-thalassemia is due to reduced synthesis of globin chains leading to synthesis of hemoglobin with an impaired oxygen binding capacity (Adil et al., 2012). In Pakistan; this inherited hemoglobinopathy has 5-7% of gene carrier frequency so, the general population has roughly 9.8 million carriers pool (Ehsan et al., 2020).

Beta-thalassemia

This type is further divided into Beta-thalassemia major, intermedia and minor as well. Beta-thalassemia major is also termed as Cooley’s anemia / Mediterranean anemia causing patient to become transfusion dependent and occur if two persons carrying a single copy of

defected Beta-globulin gene give birth to an offspring. However, those with Beta-thalassemia intermedia and minor show less severity in symptoms because of having one copy of gene and act as carrier. Moreover, 200 different types of mutations have been reported causing diverse level of Beta-globulin gene expression (Muhammad et al., 2017).

Prevalence rate of Thalassemia

World Health Organization (WHO) reported that now thalassemia has most prevalence rate as inherited genetic disorder in the world due to the migration. Globally, it is estimated that around 30, 000 out of 56, 000 conceptions have been affected by the Beta-thalassemia (Khalid et al., 2019).

Thalassemia gene in Pakistani population is found in affected families only rather than distributed randomly (Ahmed et al., 2016). Pakistan has the highest prevalence of children infected with Beta- thalassemia in the world due to consanguineous marriages, disease gene frequency, high birth rate, large pedigree size and other social culture factors like low income and lack of awareness (Muhammad et al., 2017).

Public and Private Sector Organization

In Pakistan management program of thalassemia poses great challenge as the ideal management program should accomplish the multidisciplinary team to address the all aspects. However, In Pakistan more than 40 public and private/Non-Government organizations

(NGO) including Ganaga Ram hospital Lahore, Fatimid Foundation Multan Center, Pakistan Thalassemia Center, Islamabad, Kashif Iqbal Thalassemia Care Center, Karachi and Sundas Foundation Lahore etc. are working for cure and eradication of thalassemia (Asif and Hassan, 2016).

Globally, many countries such as Italy, Iran and Greece have successfully managed and eradicate the thalassemia from their population via establishment of excellent healthcare program. Developing countries like Pakistan need more platforms at national level and more efforts are required for future management and prevention of thalassemia in population. The government should also establish the Technical Advisory Group (TAG) at district level of Pakistan for the complete guidance of the effected families (Zaheer et al., 2020). The other responsibilities of these TAG are to conduct awareness program about the thalassemia at local level even in school and colleges for future eradication objective.

Treatment Measures and their Complications

Currently, this heterozygous disorder in Pakistan is coped by repeating the blood transfusion, chelation therapy and transplantation (bone marrow or stem cell) (Muhammad et al., 2017). Thalassemia major patients in Pakistan suffering a severe myocardial iron load due to repeated blood transfusion cycles (Alvi et al., 2016). The multiple blood

transfusions in thalassemia patient cause the production of alloantibodies against the foreign red blood cells as a most common side effect (Zaheer et al., 2020). The immunity status and antigenic variation between the recipient and donor's blood originated the phenomena of alloimmunization in thalassemia patient. This complication further creates serious difficulty in matching the compatible blood for recipient and leads to life-threatening condition (Qidwai et al., 2018).

The developing countries including Pakistan are at high risk of getting transfusion transmitted viral infections such as human immunodeficiency syndrome (HIV), hepatitis C virus (HCV) and hepatitis B virus (HBV) due to repeated blood transfusion (Ahmed et al., 2016). The other factors which caused high prevalence of viral infection in thalassemia patient in Pakistan are inadequacy of blood banks, poorly managed transfusion safety standards, limited and costly medical facilities etc. The frequency of HCV spread in thalassemia patients (32.1%) is 6 times greater as compared to non-thalassemia ones (6.2%). Research studies also revealed that in Pakistan, the prevalence of HCV infection in β -thalassemia patients is higher in Punjab (45.98%) as compared to Sindh (31.81%) and Khyber Pakhtunkhwa (28.04%) (Sabiha et al., 2020).

Hemochromatosis (Iron overload) in β -thalassemia patients is another worst