PAKISTAN SUGAR JOURNAL

The first and only research journal regularly published since 1985

International Standard Serial Number -1028-1193

PAKISTAN SUGAR JOURNAL

Open Access Link www.srdb.gop.pk

Annual Subscription Rate (4 Quarterly issues)

Pakistan PKR 1,000/-

Overseas US$ 100/-

Published at Shakarganj Sugar Research Institute (SSRI) with the Patronage of Sugarcane Research & Development Board (SRDB)

Cited by
Asia Net Pakistan (Factiva International Australia)

Commonwealth Agriculture & Biology International (CABI-UK)

Subscription & Advertisement

M. Ehsan Khan, SRDB, ehsan@srdb.gop.pk

Waqas Raza Arshad, SRDB waqas@srdb.gop.pk

PAKISTAN SUGAR JOURNAL

CONTENTS **Page**

FEEDING DETERRENCE OF FIVE INDIGENOUS PLANT OILS AGAINST SUGARCANE RAT, BANDICOTA BENGALENSIS

Syeda Azra Tariq
Vertebrate Pest Control Institute, Southern-zone Agricultural Research Centre,
PakistanAgricultural Research Council, Old Blocks 9-10, University of Karachi, Pakistan
sazratariq@yahoo.com

ABSTRACT

For sugarcane rodents control usually rely on the use of synthetic rodenticides which may be risky for human health and hazardous for environment. Some naturally occurring bioactive plants products may be used to deter rodents. In this regard feeding deterrence of five plants oils; *Azadirachta indica* (neem); *Valeriana officinalis* (valarian), *Acorus calamus* (sweat flag), *Curcuma longa* (turmeric) and *Saussurea lappa* (costus) was evaluated against the sugarcane rat *Bandicota bangalensis* at 0.50%, 1.00% & 2.0% in comparison with untreated (control) in the laboratory. The results of paired choice tests revealed that consumption of neem oil showed maximum feeding deterrence as 65.94% for 2.00%, 56.79% for 1.00% and 45.41% at 0.50% treatments. Turmeric revealed maximum feeding deterrence as (64.58%) for 2.00%. Costus oil treated bait showed maximum feeding deterrence (59.35%) at 2.00%. The results may be highly significant for feeding deterrence activity against the ratsin lodged sugar canes at sugar mills.

Key-words: Feeding deterrence, *Bandicota bangalensis*, bait, Plant oil, *Azadirachta indica*,*Valeriana officinalis*, *Acorus calamus*, *Curcuma longa* and *Saussurea lappa*.

INTRODUCTION

Sugarcane (*Saccharum officinarum*) is a major, widespread and high value crop for making sugar, sugar-related products, chipboard, paper etc. Its production accounts for 3.4 percent in agriculture's value addition and 0.7 percent in GDP. During 2020-21, the crop was cultivated on 1,165 thousand hectares, an increase of 12.0 percent compared to last year's sown area of 1,040 thousand hectares. During the current year increased in production (81.009 million tonnes against 66.380 million for last year) was attained (*Pakistan Economic Survey 2020-21*). Sugarcane crop may be effected by diseases and pests; insects and rodents. Rodents inflict significant damage to sugarcane standing crop as well as lodged canes (Beg *et.al*., 1979; Roberts, T. J.,1997). They eat inner sweet core by gnawing, resulting in cane damage, sugar loss and increased susceptibility to insect and disease attack. They usually gnaw the internodes of sugarcane and inflict direct damage to thecrop. The sugarcane fields offer suitable habitation for rodents feeding and breeding. Major rodent species destructing sugarcane are *Bandicotabeng-alensis*, *Nesokia indica*, *Millardia meltada* and *Mus badooga*. Numerous scientists of Vertebrate Pest Control Institute carried out laboratory and field studies in regard of rodent management in standing as well as lodged sugar canes. They developed models, packages for the growers (Brooks *et al*., 1979; Smeit & Khan, 1980; Khokhar & Rizvi, 1999; Pervez *et al*., 1998, 1999, 2005 & 2019; Khan & Munir, 2006; Ahmed *et al*., 1915; Tariq *et al*., 2009, 2020 & 2021). Management of rodent infestation usually rely on the use of toxic rodenticides which may be risky for human health and hazardous for environment (Gray *et al*. 1994). The anticoagulants are being used for rodent control since last three decades. Increasing concern about health and environment has led to the need for searching safe plants bioactive products against rodents (Tariq, S. A, 2021). Keeping these facts in mind a study was designed to increase the knowledge regarding the practical applications of natural plant products that can minimize the use of synthetic toxic chemicals. In the study, four plants *Azadirachta indica* (neem); *Valeriana officinalis* (Valerian), *Acorus calamus* (sweat flag), *Curcuma longa* (turmeric) and *Saussurea lappa* (costus) were tested to estimate their feeding deterrence against rodents. The study may be helpful in development of IntegratedPest Management models by the addition of these bio active indigenous plants beside the well documented neem plant.

MATERIALS AND METHODS

Seeds of *Azadirachta indica* (neem) were collected, shade dried and preserved. Roots of *Valeriana officinalis* (Valerian), rhizomes of *Acorus calamus* (sweat flag), *Curcuma longa* (turmeric) and *Saussurea lappa* (costus) were provided from the Hamdard Research Institute of Unani Medicine (HRIUM), Faculty of Eastern Medicine, Hamdard University. All the plant material was preserved in wax quoted paper bags for biological assays. The plant oils were obtained by extracting plant powders withn-hexane on Soxhlet's extraction apparatus. The rats *Bandicota bangalensis* (Gray and Hardwicke) were live- trapped from sugar cane fields, Thatta district, lower Sindh (24o 45, N; 67o 55, E) Pakistan. The rats of approximate same size were sexed, weighed and caged individually in laboratory for 15 days. The rats were fed on mixed grain diet, containing rice, millet, wheat and maize during acclimation period and between the trials. Water was provided *ad libitum*. Plants oils were mixed individually in a ratio accordingly in three (2.00%, 1.00% and 0.50%) doses and were tested in comparison with control (bait without plant oil).Ten rats (five male and five female) were used in all trials beside control (one male and one female). The rats were weighed and caged singly, starved for four hours (before the start of each test). The rats were offered 20g bait for each concentration as well as 20g plain bait (without plant oils) for five days. Bait eaten (g) in paired choice was recorded after 24 hours. All trials were replicated five times, under the same temperature and humidity, results are depicted in Tables 1&2. The percentage of feeding deterrence activity in the choice test condition was calculated using the Isman *et al*. (2000) formula after some modification. Data on feeding

deterrence by the rats were also subjected to factorial analysis of variance (ANOVA). The two factors were: 1. Type of plant oil and 2. Concentration of the plant oil. The follow-up of ANOVA included Fisher's least significant test (LSD).

RESULTS AND DISCUSSION

The bait consumption of the plants oil treated bait and control bait are depicted in Table-1 whereas the percent feeding deterrence is shown in Table-2.

Feeding Deterrence by *Azadirachta indica* (Neem) Oil: Consumption of neem oil treated bait was 1.56±0.32g in comparison to 4.58±0.26g in control at 2.00% concentration, showing 65.94% feeding deterrence. Whereas at 1.00% concentration 56.79% and at 0.50% concentration 45.41% feeding deterrence was observed. Neem oil was found highly significant ($p<0.001$, Table-3) for antifeedant activity of the rats.

Feeding Deterrence by *Valeriana officinalis*(Valerian) Oil: For valerian oil the results were reversed. The maximum bait consumption 2.05± 0.07 g (42.42% feeding deterrence) was noted at 2.00% whereas minimum bait consumption 1.60± 0.15 g (55.06% feeding deterrence) was shown at 0.50% concentration. The results were highly significant ($p<0.001$) Table-3) in comparison to control.

Feeding Deterrence by *Acorus calamus* (Sweet Flag) Oil: Sweet flag showed 69.17% deterrence by consuming 1.48±0.15g bait in 0.50% treatment whereas at 1.00% the consumption was 1.55±0.16g (67.71% deterrence). The oil proved it highly significant as feeding deterrent ($p<0.001$, Table-3).

Feeding Deterrence by *Curcuma longa* (Turmeric) Oil:
Maximum feeding deterrence (64.58%) was noted at 2.00% by revealing 1.70±0.27g bait consumption The findings are highly significant ($p<0.001$, Table-3).

Feeding Deterrence by *Saussurea lappa* (Costus) Oil: Costus oil treated bait showed maximum feeding deterrence (59.35%) at 2.00% (1.13±0.39g consumption); whereas minimum feeding deterrence (51.80%) was noted at 0.50% (1.34±0.31g consumption). This deterrence was calculated in comparison to 2.78±0.36g consumption in control. The results were highly significant ($p<0.001$, Table-3). Many scientists workedon feeding deterrence of plant oils against insect pests; however this is the first hand study on feeding deterrence of plant oils against rodents. In this study consumption of neem oil treated bait showed feeding deterrence as 65.94% for 2.00%, 56.79% for 1.00% and 45.41% at 0.50% treatments. Neem is documented a useful feeding deterrent for insects. For valerian oil the results were reversed. The maximum bait consumption (42.42% feeding deterrence) was noted at 2.00% whereas minimum bait consumption (55.06% feeding deterrence) was shown at 0.50% concentration. It seemed that valerian has some attraction for the rats at higher doses. In sweet flag oil treated flour deterrence was reciprocal to the concentration and gradually decreased with the increase of concentration. The oil showed deterrence as 69.17% in 0.50%, 67.71% in 1.00% and 59.38% in 2.00% concentrations (Tariq *et al*. 2007) reported that at only 0.01% concentration *Acorus calamus* oil reduced the feeding activity of American bollworms (*Heliothis armigera*), spotted bollworm (*Earias fabia*) and pink bollworm (*Pectinophora gossypiella*) significantly as compared to control. These results are similar with the present results. Turmeric revealed maximum feeding deterrence for consumption of treated bait as (64.58%) in 2.00% concentration and minimum deterrence (60.21%) in 0.50% concentration. The findings of this study may be highly significant for Integrated Pest Management for rodents. By spraying of neem and turmeric oil mixed water on standing or lodged sugar canes rodents may be repelled. The results may be highly significant for antifeedant activity against the rats in lodged sugar canes at sugar mills.

Table-1 Consumption of plant oils treated bait by the rat *Bandicota bangalensis* in 24 hours

Concentration (%)	Consumption of flour				
	***Azadirachta Indica* (Neem)**	***Valeriana officinalis* (Valerian)**	***Acorous calamus* (Sweet flag)**	***Curcuma longa* (Turmeric)**	***Saussurea lappa* (Costus)**
0.000	4.58±0.26	3.56±0.21	4.80±0.30	4.80±0.30	2.78±0.36
0.50	2.50±0.32	1.60±0.15	1.48±0.15	1.91±0.17	1.34±0.31
1.00	1.98±0.12	1.83±0.12	1.55±0.16	1.79±0.18	1.27±0.30
2.00	1.56±0.32	2.05±0.07	1.95±0.33	1.70±0.27	1.13±0.39
LSD*0.05	0.7	0.44	0.74	0.71	0.49

All values are mean of five replicates ± Standard Error, *Fisher's Least Significant Difference

Table-2 Feeding deterrence of plant oils treated bait by the rat *Bandicota bangalensis*

Plants oil	Concentration (%)	Deterrence (%)
Azadirachta indica	0.50	45.41
(Neem)	1.00	56.79
	2.00	65.94
Valeriana officinalis	0.50	55.06
(Valerian)	1.00	48.60
	2.00	42.42
Acorous calamus	0.50	69.17
(Sweet flag)	1.00	67.71
	2.00	59.38
	0.50	60.71
Curcuma longa	1.00	60.21
(Turmeric)	2.00	64.58
Saussurea lappa	0.50	51.80
(Costus)	1.00	54.32
	2.00	59.35

FD (%)= FC-FT/ FC x 100 Where, FD= Feeding Deterrence, FC=Feeding in Control (0.00%) bait FT=Feeding in Treated bait

Table-3 Two-way ANOVA for the consumption of plant oils treated bait by the rat *Bandicota bangalensis*

Dependent Variable	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	26.875	3	8.958	25.3711	2.54E-06
Neem within Groups	5.650	16	0.353		
Total	32.525	21			
Valerian Between Groups	11.773	3	3.924	36.674	2.12E-07`
Within Groups	1.712	16	0.107		
Total	13.484	19			
Sweet flag Between Groups	37.586	3	12.529	41.097	9.59E-08
Within Groups	4.878	16	0.3049		
Total	42.464	19			
Between Groups	33.908	3	11.303	40.692	1.03E-07
Turmeric Within Groups	4.444	16	0.278		
Total	38.353	19			
Between Groups	9.004	3	3.001	22.328	5.8E-06
Costus Within Groups	2.151	16	0.134		
Total	11.155	19			

RECOMMENDATIONS

Sugarcane provides an ideal condition for rodent populations to exist and cause widespread damage to the sugarcane, standing or lodged. The rodent control often performed is inadequate for high rodent populations. The growers should have good knowledge of the rodent damage as per the sugarcane variety, rodent species and its biology in relation to crop timings. For economical rodent control services the sugarcane growers and personnel involved in sugar industry may contact Vertebrate Pest Control Institute (VPCI), Southern-zone Agricultural Research Centre (SARC), (Karachi), Pakistan Agricultural Research Council (PARC). An Integrated Pest Management approach is being practiced to manage rodent's damage in sugarcane crop by VPCI, PARC-SARC.

The package is successfully developed for rodent management after a series of trails and is ready for adoption by the end-users; advisory services are alsoavailable.

ACKNOWLEDGMENTS

I acknowledge Dr. Attaullah Khan, Director General, Southern zone Agricultural Research Centre for his help and support. I am very thankful to Ms. Rahila Minhas for her help in these studies. Field and laboratory assistants of VPCI, PARC-SARC are acknowledged for their help in these studies.

REFERENCES

Ahmed, S. M., Pervez, A., Tariq, S. A., Hasan, Z. U. & Khadija, E. (2015). Laboratory evaluation of some bait bases to formulate palatable bait for the control of short-tailed mole rat, *Nesokia indica*. Pakistan J. Zool., 47 (5): 1387-1391.

Beg, M. A., Khan, A. A. & Begum, F. (1979). Rodent problem in sugarcane fields of central Punjab. Pak. J. Agri. Sci., (16): 123-129.

Brooks, J. E., Htun, P. T., Naing, H. D., Walton, W. & Tun, M. M. (1979). Laboratory evaluation of rodenticides for use against South-East Asian commensal small mammals. WHO, Geneva (WHO/VBC/79.720).

Gray, A. C., Eadsforth, V., Dutton, A. J. & Vaughan, J. A. (1994). The toxicity of three second-generation rodenticides to Barn Owls. Pestic. Sci., (42): 179-184.

Isman, M. B. (2000). Plant essential oils for pest and disease management. Crop Protect. 19: 603-608.
Khan, A. A. & Munir, S. (2006). Effect of zinc phosphide bait on rodent abundance in sugarcane field. Proc. of 41st Annual Convention, Pak. Soc. of Sugar Technol.: 206-213.

Khohar, A. R. & Rizivi, S. W. A. (1999). Vertebrate pest problems in Sindh and their impact on agricultural economy. Pakistan Food and Agriculture Review: 7-8.

Pakistan Economic Survey (2020-21). Economic Advisor's Wing, Finance Division. Ministry of Finance. Government of Pakistan, Islamabad.

Pervez, A., Ahmed, S. M., & Rizvi, S. W. A. (1998). Comparative efficacy of Bromadiolone, Cholecalciferol and Zinc Phosphide against short-tailed mole rat, *Nesokia indica* in captivity. Turkish. J.of Zoology, (22): 137-140.

Pervez, A., Ahmed, S. M., Ahmed, S. & Rizvi, S. W. A. (1999). The significance of additive to enhance poison bait acceptance against rodents damaging paddy in lower Sindh, Pakistan. Pak. J. Zool., 37 (3): 207-210.

Pervez, A., Ahmed, S. M., Ahmed, S. & Rizvi, S. W. A. (2005). Comparative field efficiency of some additive formulated baits against rodent pests of wheat crop in Sindh, Pakistan. Pak. J. Zool., 37 (4): 269-274.

Pervez, A., Ahmed, S. M. & Tariq, S. A. (2019). Assessment of sugarcane varietal damage from field rats and their management strategy in Sindh. Pak. Sug. J., XXXIV (01): 11-14.

Roberts, T. J. (1997). The Mammals of Pakistan, Oxford Uni. Press, pp: 361.

Smiet, A. C., Fulk, G. W. & Lathiya, S. B. (1980). Rodent ecology in sugarcane in lower Sindh, Pakistan. Acta Theiol., 25 (8): 81-97.

Tariq, R. M., Naqvi, S. N. H., Zafar, S. M. N. and Burrero, A. S. (2007). Toxic effects of botanical pesticide from *Acorus calamus* (AC) and *Annona squamosa* (AS) against Bollworms at ARI Tandojam, Sindh, Pakistan. *Pak. J. entomol.,* 22 (1-2): 31-36

Tariq, S. A., Pervez, A. & Ahmad, S. M. (2009). Development of rodent and wild boar pest management models for sugarcane crop. Proceedings of 44th Annual Convention Pak. Soc. of Sugar Technol: 149-154.

Tariq, S. A., Ahmed, S. M., Zafar, H. & Keerio, Q. A. (2020). Laboratory studies on feeding preference of short-tailed mole rat, Nesokia indica, for some commercial varieties of sugarcane. Pak. Sug. J., XXXV (01): 4-9.

Tariq, S. A (2021).Rodenticidal Activity of Datura metel against Sugarcane Rat, Nesokia indica, Pak. Sug. J., XXXV1 (02): 16-22.

EXISTING AND FUTURE STRATEGIES ON DETECTING AND MANAGING *COLLETOTRICHUM FALCATUM* CAUSING RED ROT OF SUGARCANE

Waqas Raza Arshad* and Abdul Khaliq**
*Sugarcane Research and Development Board Punjab Pakistan **Sugarcane Research Institute Faisalabad Pakistan waqasr.arshad@gmail.com

ABSTRACT
Sugarcane is an important industrial crop because it is the major source of white sugar. It is also one of the crops for the alcohol and biofuel industries. Disease-causing organisms can significantly decrease the productivity of sugarcane plants and sugar quality. Among the disease-causing organisms, *Colletotrichum falcatum* Went causes the most significant economic loss (5–50%) in the sugarcane production due to red rot disease. This loss results in only 31% sugar recovery. It is reported that *C. falcatum* can kill sugarcane plants. Currently, there is no sustainable way of preventing red rot disease from spreading in sugarcane plantations. Many popular sugarcane varieties are no longer used in sugarcane cultivation because of their susceptibility to *C. falcatum*. The objectives of this manuscript were to: (i) summarize existing approaches for the early detection of red rot disease and controlling techniques of red rot disease in the field and laboratory and (ii) assess red rot disease control effectiveness so as to propose better methods for mitigating the spread *C. falcatum*. If our proposition is adopted or practiced, it could significantly contribute to the mitigation of *C. falcatum* infection in the sugarcane industry. This could enable achieving sustainable cultivation of sugarcanes to guarantee the sustainability of the sugar industry in the tropics and the subtropics.

Keywords: *Colletotrichum falcatum*; sugarcane; red rot disease; detection and management

INTRODUCTION

Sugarcane (*Saccharum officinarum* L.) is regarded as one of the essential cash crops because it improves the socio-economic livelihood of many sugarcane growers. Although sugar is widely used in our daily lives, there is a growing interest in sugarcane as one of the potential economic crops for bio-energy (ethanol) production. The worldwide occurrence of sugarcane is approximately 26.3 million hectares and the gross production is approximately 1.9 billion tons. The major sugarcane producing countries are Brazil, India, Thailand, Pakistan, China, Mexico,United States of America and Australia. In spite of the public concern about the excessive sugar consumption in the world, The daily consumption of sugar is on the increase trend especially in the developing countries where the per capita consumption is relatively low. For example, the global sugar demand is projected to increase to 203 Mt by 2028 and this will add 32 Mt to the existing tonnage. Increasing the demand for sugar will be driven by Asian, Middle Eastern and North African countries. Currently, the small-scale sugarcane planters are facing many challenges including biotic and abiotic factors. These factors have been implicated in the decreasing sugarcane production. The abiotic factors include extreme heat, drought, typhoons, flooding, frost and poor soil fertility. It is believed that, productivity of the sugarcane plants is reduced because of water stress. The life cycle of sugarcane plants is affected by approximately 240 sugarcane diseases. Approximately 100 fungi, 10 bacteria, 50 nematodes, and 10 viruses have been identified as pathogens of sugarcane worldwide. Out of the many biotic stresses of the sugarcane, the *Colletotrichum falcatum* Went causes significant reduction in the quality and yield of susceptible sugarcane cultivars. The red rot occurs in 68 sugarcane producing countries. This disease decreases sugarcane yield by 5–50%. The loss results in only 31% sugar recovery.

Besides reducing yield attributes, the red rot reduces the sugarcane juice quality (as sucrose content, purity, Brix) and commercial cane sugar. Red rot disease is the major disease due to the destructive effects of the disease as the main cause for the withdrawal of the many sugarcane varieties in the sugarcane cultivation worldwide. The variations in the morphologic and pathologic characteristics of the genus of *Colletotrichum* are related to their geographical origins. However, because of many overlapping characteristics within the species' complex, identification using morphologic techniques is not enough. Moreover, managing red rot disease in the field is difficult as the genetic makeup of this fungus keeps changing. The objectives of this manuscript were to: (i) summarize the existing approaches for early detection of red rot disease and controlling techniques of red rot disease in the field and laboratory and (ii) assess red rot disease control effectiveness so as to propose better methods for mitigating the spread *C. falcatum*.

Pathogen, Infection and Transmission

Colletotrichum falcatum belongs to the Ascomycota phylum. This pathogen is a facultative parasite. It occurs inanamorphic and teleomorphic forms but the amorphic stage which infects standing canes is the most important stage. The distinctive morphological and cultural characteristics of *C. falcatum* include the development of acervuli with setae, presence or absence of teleomorph, pinkish appearance of colony, sporulation and growth rate. These characteristics are well described by Sharma and Tamta, 2015. Many fungal isolates are significantly different. Diversity in virulence within pathotypes had revealed that a red rot pathogen undergoes adaptive changes in host cultivars. Viswanathan *et al.* 2019 reported that isolates are virulent in susceptible varieties, but not in resistant and moderately susceptible varieties. Virulence frequencies of isolates range from 21.3–40% on mildly susceptible varieties compared with 62.9–97.9% on susceptible varieties. This suggests that *C. falcatum* isolates differ in their host infectivity. Thus, it is very important to identify the extent of pathogen diversity and the way infection occurs to develop effective disease control and planting management. Many sources of inocula are involved in the transmission of the disease. The pathogen is primarily disseminated through soil and diseased setts, whereas secondary distribution is through irrigation water, rainfall splashing, midrib lesion dew brushing, wind dispersal and other field vectors. The relative importance on how inoculum is spread depends on the time of the year and the conditions under which the cane is growing. The pathogen infects stalks through nodes leaf scar, growth ring, root primordial and buds. The pathogen enters the nodes of a sugarcane plant through the inner epidermis of the lower part of the leaf sheath of this plant. In an unfavorable condition, the fungus produces appressoria on rind and leaves. During soil borne transmission, latent fungal structures, namely appressoria, dense-walled hyphae chlamydospores, and setae play important roles in the dispersal of disease. *Colletotrichum falcatum* thrives on unhealthy stalks or stubble fragments. Although *C. falcatum* is not a definite soil-inhabitant pathogen, there is enough evidence to suggest that fungal propagules are perpetuated by debris borne inocula. The red rot appearance depends on type of the infection and environmental conditions. Usually disease occurs at early growth stages and symptoms are often difficult observe. The red rot disease is divided into four types, namely tiller, lamina, mid rib and stem red rot. The infection also causes alterations in the color of the lamina leading to straw color in the middle and dark reddish-brown on the edges as the black acervuli progresses. Eventually, the infected leaves split and hang at the lesions, whereas, in the rib red rot, red color is observed through the whole mid rib. Reddening of the internal tissues with alternating red and white patches (with an alcoholic scent) are the main indicators to suggest occurrence of the disease in the stalk at the later stages. Generally, the disease symptoms appear when leaves of the spindle (3rd and 4th leaves) show drying that wipe

the top along the leaf margins. This discoloration persists from the tip to the base until all the crown leaves wane and red rot infected canes are separated from the nodes easily.

Identification of *Colletotrichum falcatum*

The most important requirement in any good disease management practice is the accurate identification of the pathogen. Characterization of *C. falcatum* isolates by cultural, pathologic and molecular methods is commonly used to confirm the presence and to study the genetic and phenotypic diversity within a population. The different methods that have used to identify *C. falcatum* are subsequently discussed.

Traditional Methods

The traditional approaches that are used to detect andidentify diseases include isolation and characterization pathogens using inoculation testing. *Colletotrichum* species are described primarily based on morphologic features such as mycelia development, production of mycelia dry matter, mycelia color, texture, topography, shape and size of conidia. The conidia of *Colletotrichum* species are easily seen using a compound microscope, but the accuracy of the conventional identification method and its reliability depend on depth of experience. In addition, phenotypic detection is time-consuming and requires skilled or skillful personnel. However, because of the many overlapping characteristics within the species complex, identification using morphologic techniques is not enough. Pathogenicity and virulence tests are also another part of the conventional techniques for the identification of plant pathogens. Generally, the pathogenicity test requires longer time to confirm the pathogen. Moreover, the morphologic and pathologic identification techniques are time-consuming. In addition, these techniques are significantly affected by environmental factors.

Serologic Methods

Serological methods are used to identify red rot fungus. A body of knowledge has been developed on the serological variability among *C. falcatum* isolates using for example, enzyme- linked immune sorbent assay (ELISA) technique. The findings of Viswanathan *et al.* 2019suggest the possibility of using the serological technique to quantify the pathogen colonization and how they correlate with host resistance.Based on *C. falcatum*colonization in cane stalk, Viswanathan *et al.* 2019 classified the host reaction to the pathogen as resistant,moderately resistant andsusceptible. The authors concluded that the pathogen colonization was higher in nodal regions compared with the intermodal tissues. Hiremath and Naik tried to detect *C. falcatum* in sugarcane tissue using multiple serological analyses such as ELISA, dot immune binding assay (DIBA) and western blotting. Viswanathan *et al.* 2019 demonstrated that the ELISA technique could detect *C. falcatum* infection of sugarcane tissue in stalks using polyclonal antiserum raised against the pathogen. In addition, they found isolated polyclonal antibodies were specific to *C. falcatum*.

Khalid *et al.* 2012 isolated two protein molecules from the mycelium of *C. falcatum* race cf. 05 at 27 kDa and 45 kDa molecular weights—after which it was used to develop polyclonal antibodies. The antibodies produced were species specific and they had high affinity for *C. falcatum* (1:50,000 and 1:500 dilution). Another simple, fast and targeted assay for the laboratory analysis of sugarcane (C 671) red rot (at the early growth stage of sugarcane plants) using DIBA with dilution ratios of 1:1000 and 1:100 antigen and secondary antibody, respectively. Although these techniques are promising, the disadvantage of the serologic tests is the possibilities of false positives. The false positives are caused by cross-reaction of antibodies with plant debris or unrelated organisms. Using ELISA, Viswanathan *et al.* 2019 showed that seed cane indexing is possible for red rot infection. This index identifies red rot resistance in a shorter time, and can also be used to screen large populations. This technique is appropriate for rapid screening because it enables early detection of pathogen colonization before symptoms are evident. In addition, the technique enables pathogen assessment load of different nodal sites of sugarcane plants with plant

growth promoting rhizobacteria. However, the nonspecific reaction caused bycertain cane tissues we must be fixed.

Molecular Method

Colletotrichum species are characterized using different molecular approaches. Unlike the traditional methods, molecular techniques not affected by environmental factors. The presumed existence of intermediate forms between species, morphologic plasticity and overlapping of the phenotype make the use ofthe traditional method less effective. These barriers hinder the use of classical criteria to identify these pathogens. As a result, the molecular biology technique encompasses alternative and supplementary approaches because they are important techniques for overcoming the difficulties in identifying up to species level. For the good detection of *Colletotrichum* species, molecular phylogeny combined with morphologic and cultural traits,pathogenicity and physiological tests are recommended. Molecular approaches such as sequence analysis of the internal transcribed spacer(ITS) region between large and small subunits of ribosomal DNA (rDNA) are commonly used to detect fungi such as *Colletotrichum* spp.. Combinatin of multiple genes characterization, such as ITS, actin, glyceraldehydes-3 phosphate dehydrogenase (GPHD) and beta-tubulin could offer more accurate for identification of fungal taxa. Inter simple sequence repeat (ISSR) markers have been effective multilocus markers for genetic diversity analysis, finger printing and mapping of genomes. This approach enables us to understand pathogen population dynamics. The advantages of ISSR markers are semi-arbitrary markers, highly polymorphic, highly informative, low cost and only low quantities of template DNA are needed. To successfully improved crop productivity, genetic characterization of pathogenic variants of crop pathogens is essential. Molecular biology is a good tool for fungal taxonomists.

Viswanathan *et al.* 2019 documented that *C. falcatum* draft genome size is approximately 48.16 Mb. This genome has 12,270 genes with 90% and 84% identical genes for *C. graminicola* and *C. sublineola*, respectively. In addition, Viswanathan *et al*. 2019 reported that *C. falcatum* genome has plant cell wall degrading enzymes (CWDE), transposable components, primary secondary metabolites, candidate secretory effectors (CSEPs), membrane carriers, signaling molecules, carbohydrate-active enzymes (CAZymes), matting proteins, sclerotic development proteins and a special member of the *Colletotrichum* family. This report improves our understanding on species that are close to *C. falcatum*.

Scindiya *et al.* 2017 showed that RNA-mediated silencing of PKS1 gene in *C. falcatum* causes the red rot in sugarcanes. The authors believe that the gene homologs are responsible for *C. falcatum* virulence and its pathogenesis. Scindiya *et al.* 2017 showed that two isolates —viz., Cf 671 and Cf92020 differed phylogenetically with multiple gene homologs differing in their virulence. Intra and interspecific variation as well as genomic sequenced origins between two *C. falcatum* isolates (Cf671 and Cf92020) have been identified. During interaction with the host–pathogens, expression of pathogenic gene homologs with both isolates occurs. Scindiya *et al.* 2017 showed that molecular approaches can be used to differentiate between closely related species with few morphologic differences and stains or even distinct isolates within the species.

According to Nandakumar *et al.* 2020 green fluorescent protein (GFP) can be used to explore the interactions between *C. falcatum* and sugarcane to establish pathogenesis, colonization and dissemination of this fungus in host tissues. The authors demonstrated that the GFP transformed *C. falcatum* strain was firmly incorporated in the mitotic stability. Moreover, the *C. falcatum* transformants retained morphologic features and growth parameters because the wild type and virulence type were not altered relative to wild *C. falcatum.* The *C. falcatum* pathotypes tagged with GFP specifically showed differences in *C. falcatum* colonization through cooperative and incompatible sugarcane encounters. However, these molecular methods are expensive and need specific primers to amplify DNA for identification of pathogens.

Image Processing Method
Padhy *et al.* 2016 reported that image processing techniques are innovations in agriculture and one of such innovations is automatic disease detection. Computer vision-based image processing techniques and detection algorithms had been used detect midrib red rot, leaf scald and mosaic diseases in sugarcane. The steps involved in this disease detection method are image acquisition, image preprocessing, image segmentation and feature extraction and classification. The image processing method is not commonly used in the field because the length and width of the sugarcane leaf blade vary upto 60 inches and three inches, respectively. Proportional adjustments are required to cover the entire leaf region. Disproportional changes can reduce image resolution, resulting in poor segmentation of the diseased section of leaves. To increase precision, it is essential that leaves are cut into pieces. Another important factor in image acquisition is that the rate of evaporation of sugarcane is 150 to 200 times greater than in other plants. As a result, the sugarcane leaf wrinkles after it is removed from the stem. Therefore, rapid capturing of the images is recommended. This method gives rapid results. It must be stressed that because this method is a new innovation more experimental results are required to validate the method.

Fluorescence Imaging
Noninvasive strategies to photograph multispectral fluorescence patterns or leaf temperatures across contaminated plants have significantly improved our knowledge on plant responses to biotic stress. With this technique, chlorophyll fluorescence is measured as an incident light factor on plant leaves, and variations of the fluorescence parameters are used to examine the response of pathogenic pathogens to changes in photosynthetic system and pathways of photosynthetic proton transport. Temporal and spatial differences in chlorophyll fluorescence had been used to successfully detect causative pathogens with powdery mildew and leaf rust in wheat leaves. Although this technology can be used to detect diseases and photosynthetic anomalies in sugarcane leaves, the practical use of the technique in the field is limited.

Thermography in Disease Detection
Thermography provides information on the variations in plant leaves' surface temperature and plant canopies. Thermographic cameras can track emitted infrared radiation as well as analyzing color variance.Earlier studies had suggested that phytopathogens can inhibit lack of water in stomata-regulated plants. Thermographic imaging can monitor the resulting infection, and the volume of water culminated can be calculated without specific temperature considerations. Several research scientific groups have related plant pathogen infection to temperature changes. For example, thermographic image of healthy oil palm tree is compared to palm infected with basal stem rot disease (BSR). The images captured at the same scale can suggest that temperature of the leaves of BSR-infected trees is higher than the leaves of healthy trees. Thermography is also a good means of measuring soil borne pathogen infection heterogeneity. Nevertheless, because of their high susceptibility to changes in environmental conditions, the practical application of thermography in disease control is limited. Thermographic identification is usually disease-specific and because of this limitation it is not capable of differentiating

Hyperspectral Imagery.
Hyperspectral imagery can be used to gather valuable information on the health of plants over a wide range of wavelengths (350 to 2500 nm).With hyperspectral imagery, valuable information on plant canopies such as chlorophyll pigment status, plant cell structure condition and plant structural water content can be obtained. In production agriculture, hyperspectral imagery is widely used for the detection of crop diseases. This technology is versatile and offers rapid interpretation of image data. For sugarcane diseases, Apan *et al.* 2004 analyzed multiple narrow band indices from EO-1 Hyperion imagery. Forty spectral foliage indices were produced with emphasis on leaf pigment-based lines, internal leaf

composition and water content of leaf. Discriminatory function analysis was used to pick an optimal range of indices dependent on their similarities to the discriminatory method. The outcome showed that Hyperion imagery can be used to identify orange rust disease in sugarcane crops. The findings suggested that the spectral reflections (signatures) in the areas with sugarcane orange rust disease were significantly different. Although sugarcane plants are vulnerable to multiple diseases and pests, only Apan *et al.* 2 004 conducted a research to identify and delineate infested cane areas using hyperspectral remote sensing. Research on orange rust disease diagnosis showed positive outcomes using hyperspectral remote sensing. Nevertheless, further research is required to identify the pests and diseases that are caused by other phytopathogens. Although the hyperspectral technology has worked well in association with different methods of band analysis and pattern recognition algorithms, more research projects are needed to improve information on ease of use, large scale coverage, plant variability and the economic viability for using this technology.

DNA/RNA-Based Affinity Biosensor

Anew affinity biosensor had been developed using nucleic acid fragments as pathogen identification components. The DNA-based biosensor enables early identification of diseases before occurrence of visible symptoms. This is based on probability identification at molecular level. The specific DNA sequences had been widely used to classify genetically engineered organisms, viruses and fungi. Depending on the precise nucleic acid hybridization on the sensor and DNA analytes sequence of immobilized DNA probe DNA-based biosensor, it is possible to identify genetic and infectious diseases rapidly, reliably and accurately. The most commonly used DNA assay is the single stranded DNA (ssDNA) on electrodes with electro active markers to test hybridization between the DNA source and the supplementary DNA analysis. Identification of DNA analytes is achieved based on the differences in physio-chemical characteristics such as mass, temperature, optical and electrical characteristics resulting from the two-stranded DNA hybridization (dsDNA) that occurs during the analysis. Although the use of DNA-based biosensors for the detection of plant diseases is promising, it requires small amount of nucleic acid and PCR is often required before continuing to downstream analysis. The drawbacks of biosensors based on DNA include a single DNA detector synthesis criterion, target DNA amplification, high cost (DNA-based molecular beacons) and insufficiency for real-time detection.

Management of Red Rot Disease

Red rot disease management is conventionally based on cultural practices, use of resistant varieties, disease free planting materials, physical, biologic and chemical control, among others. These methods are intended to restrict incidence of red rot after replanting to increase the productivity of sugarcane plants. However, the management strategies for minimizing red rot incidence had not yielded acceptable results. To date, no single method is able to mitigate the disease incidence. Integrated disease management (IDM) is one of the excellent practices for disease control approaches. Integrated disease management practices decrease red rot occurrence, increase growth parameters and increase sugarcane performance attributes compared to non-IDM practices. Integrated disease management involves all the methods of disease control. The subsequent discussion focuses on this aspect.

Agronomic and Cultural Practices

Opting for the good agricultural practices and integrating cultural and biologic control methods as a preventive measure should be of utmost priority. The use of healthy planting materials, certified seeds, field sanitation, crop rotation and proper drainage facility could significantly minimize red rot disease. These cultural practices have been suggested not only to reduce the inoculum from the field, but to also reduce crop losses. Mono cultivation of the same crop with the same cultivar increases the inoculum level resulting in the development of the disease. The crop must be rotated after two to three years/cycles in the

heavily infested field and the ratooning should be discouraged. Authorized enforcement of nursery programs is very important. Disease and pest free seeds/setts and mixtures with other varieties must be guaranteed. The most useful method for control of the pathogen is the use of disease-free setts. Jain reported that the geographical origins of pathogen isolates had not been related to molecular and pathologic heterogeneity. This suggests that the most effective ways of avoiding this destructive disease is through the use of disease-free planting inputs in commercial cultivation. Adopting field sanitation practices such as removing and burying of crop debris, withered leaves, stubble, among others before planting is essential. Sugarcane fields should be well leveled, and hygienic farming should be adhered to. Regular field inspection and roughing of diseased plants could minimize the occurrence of red rot disease. Moreover, because the disease is associated with soil nutrient imbalance, fertilizer management is very important. The above mentioned practices have been reported to minimize the disease incidence and severity. However, these practices are unable to eradicate the disease.

Physical Treatment

Infected planting materials are the primary source of pathogen inocula for the occurrence of red rot disease in sugarcane fields. Many researchers had documented that sett borne red rot infection can be suppressed using heat therapy. Arade *et al.* 2014 reported that moist hot air therapies (54 C for 3 h and RH 95%) can completely eradicate sett borne infection. In combination with heat and chemotherapy, mixing synthetic chemicals in a hot water controlled the red rot. Arade *et al.* 2014 stated that using moist hot air at 54 C for 2 h was more effective in reducing the incidence of the red rot than using hot water at 50 C for 2 h. Singh and Singh, 1989 reported that aerated stream at 52 C or sett, soaking in cold running water for 48 h followed by 150–180 min of hot water treatment at 50C can eliminate the pathogen from infected setts. Other practices which had been recommended for the red rot management include burning waste, preserving enough soil moisture and timely harvesting of contaminated or susceptible crops. The advantages of the physical treatment are as follows: eco-friendly, easy to adopt, cheaper, and it kills setts borne pathogens. However, this intervention is time-consuming.

Chemical Control

In-vitro studies suggest that the chemical control method completely inhibits C. falcatum growth. For example, Benomyl® 50 WP, Folicar® and Radomil® 75WP (100%) at a level of 5–50 µgmL−1 completely inhibited fungal growth [72]. Similarly, Bharadwaj and Sahu [73] reported complete inhibition of C. falcatum mycelia growth using Bavistin®. However, their effectiveness in the field remains unproven. In the field, the role of sett treatment controls the primary source of red rot from setts [30], and the use of fungicides to combat red rot in the field is usually restricted to setts treatment. It is possible to reduce red rot incidence by treating the infected setts with carbendazim and benomyl for 30–60 min [74]. In some studies, dip treatment of sugarcane setts (handling debris borne infection 24 h before planting) with 0.25% suspension of thiophanate methyl and carbendazim metabolite effectively controlled red rot disease. Rahman *et al.* 2016 reported that Topsin® M treatment protected canes against red rot disease and the effectiveness increased cane yield. Using thiophanate methyl at 0.25% as sett treatments considerably suppressed red rot disease incidence. The defense had been attributed to chemical antifungal effects on the pathogen. Fungicide thiophanate methyl also increased germination rate, tillers count, number of millable cane, weight of single cane, length of the cane, diameter of cane and yield of cane. In spite of these positive results, the literature is replete with many findings that the chemical treatment method has miniature effect on red rot disease because of rinds impermeability, presence of abundant nutrient in the area, the existence of fibrous nodes at the cutting ends, poor fungicide solubility and water in setts. The benefit of the chemical treatment method is its efficacy because the effectiveness is better than other methods, but it is not eco-friendly.

Use of Resistant Varieties against Red Rot

The recurrent outbreak of red rot in epiphytotic condition had compelled breeders to develop red rot resistant varieties. The evolution of new races of the pathogen is a major factor for the breakdown of new varieties. Among the species of sugarcane, *S. spontaneum* is the most resistant species whereas *S. officinarum* is the least resistant species. Although the inherited genetics of the red rot resistant genes are not well established, there is significant progress in the development of resistant varieties against the red rot. The red rot resistance is transferred in sugarcane species through interspecific, intraspecific or intergeneric crosses. The focus of the breeding work in the Indian sub-continent is the development of red rot resistant varieties through interspecific crosses. However, because the pathogen varies, after a disease-resistant variety is released for commercial cultivation, within 8 to 10 years, it becomes vulnerable to red rot disease because the pathogen evolves into a new and more virulent strain. There are significant attempts to detect genes and markers which are related to the red rot resistance. Because of exceedingly heterozygous polyploid seed genome along with a constricted genetic base (based on the conventional and genetic mapping methods), it has been difficult to breed for red rot resistance in sugarcane. Although tags of differentially articulated sequences had been identified in response to the infection of *C. falcatum* they do not contribute to the discovery of the functional target gene(s) for resistance of the red rot because the study was conducted on a particular sugarcane genotype without exploring the history of segregation and epistatic interactions. Singh *et al.* 2014 developed recognized target genes for the red rot resistance following linkage imbalance-based interaction mapping. However, their role in imparting resistance to disease is yet to be confirmed thus, restricting their use as molecular markers for the detection of resistant genotypes and markerassisted collection for sugarcane. Recently, Nayyar *et al.* 2017 discovered β-1, 3 glucanasegene expressions from *Trichoderma* sp. The β-1, 3 glucanasegene is Responsible for the improvement of transgenic sugarcane that is resistant to the red rot. The integration of transgenic genes and their expression was confirmed in the first generation of T_0 plants by quantitative reverse transcription PCR up to 4.4 times higher expression than with non-transgenic sugarcane. Two virulent pathotypes of *C. falcatum* (Cf08 and Cf09) which cause the red rot have been shown in bioassays of transgenic plants where some plants had resistance to Cf08 and mild resistance to Cf.09. Nayyar *et al.* 2017 clarified that the resistant transgenic plants cells did not lose sucrose because of inhibition of fungal hyphae-to-hyphae or hyphae swelling. Hyphallysis occurs because of the action of β-1, 3-glucanase on the β-1,3-glucosyl enzyme linkages of the fungal cell wall. This transgenic resistant and moderately tolerant sugarcane can also be used to develop resistant varieties against *C. falcatum*.

Biologic Control and Natural Products

Eco-friendly and sustainable alternative approach to manage diseases is biological control. Different bio-control agents have been used either alone or in combination with other management methods to control *C. falcatum* in sugarcane. Among the biocontrol agents, plant growth-promoting rhizobacteria (PGPR) that are allied with root of sugarcane would be useful in sustaining plant growth through developing many plant growth-supporting metabolites. Plant growth promoting rhizobacteria at the rhizosphere of sugarcane plants improves the growth of sugarcane plants by colonizing their rooting zones. Plant growth-promoting rhizobacteria can also inhabit *C. falcatum*. In recent times, different genera of bacterial such as *Enterobacter*, *Pseudomonas*, *Burkholderia*, *Bacillus*, *Gluconaceto bacter* and *Ochrobactrum* are known (in-vivo and in-vitro trials) to effectively inhibit *C. falcatum* in the sugarcane rhizosphere. Patel *et al.* 2019 conducted an in-vivo study against three strains of *C. falcatum*. The findings showed that *Ochrobactrum intermedium* (TRD 14) effectively regulated the pathogenicity of *C. falcatum* (cfNAV) and it also enhanced the growth of sugarcane plants by 8.2%. Furthermore, the sugarcane plants with *O.*

intermedium (TRD 14) increased stem diameter. In the case of *Acinetobacter* sp. (PK9) and *Bacillus* sp. (RSC 29) protection against *C. falcatum* strains, it was observed that the height and diameter of the stem of the sugarcane plants were not significantly improved. The sugarcane plants started drying after 45 days, but in the absence of red rot disease, the two strains increased height of the sugarcane stem. The most promising results were noticed using *Escherichia* sp. (VRE34) because it effectively suppressed disease apart from improving the growth of plants. *Trichoderma harzianum* is another bio-agent which is being used to management red rot disease. The effectiveness of *T. harzianum* is related its direct parasitic effect on *C. falcatum*. *Trichoderma harzianum* application is reduces the economic losses in Susceptible varieties. In addition, the use of *T. harzianum* increases cane yield because of the increased germination and shooting of biomass. *Trichoderma* bio-pesticide application is eco-friendly, economical, besides improving soil quality. *Trichoderma harzianum* can directly control *C. falcatum* by producing systemic resistance in treated sugarcane plants. The application of *T. harzianum* strain Th37 on stubbles at 20 kg/ha increased nitrogen (N), phosphorus (P) and potassium (K) availability by 27.65% and 44%, respectively. The level of red rot defense increased to78%when in combination with TMC/salicylic acid (SA) and 86% with metabolites/SA, where defense was 60% and 71%, respectively. Plant based extracts had also been reported to suppress *C. falcatum*. It has been reported that ginger, onion, and garlic extracts can inhibit mycelia development of *C. falcatum*. Applications of essential oils such as menthe oil, patchouli oil, peppermint oil and palm oil can also mitigate *C. falcatum* infection. According to Imtiaj *et al.* 2007 *Datura metal* and *Curcuma domestica* leaf extracts can inhibit both mycelia and conidial growth of the red rot pathogens. Similarly, tobacco and dhup smoke (incense) are thought to inhibit the red rot conidial germination. These findings were not obtained from field experiments. Thus, stages and detailed studies on their effectiveness in field evaluations are required. The use of biocontrol agents and natural products are eco-friendly, economically efficient for improving soil health and good for pathogens suppression for a long period. However, their field effectiveness is very low and currently, there is dearth of information on this aspect. To this end, emphasis on the formulation of durable bio products whose potencies can withstand different environmental challenges is essential.

Legislation (Quarantine)

Plant quarantine laws enable government agencies to protect the entry of alien insects and pathogens into countries. Uncontrolled setts cane movement is primarily responsible for spreading red rot disease. Therefore, it is important to limit cane transport from an infected area to disease free zones/areas. Only research stations with valid phytosanitary certificates should import seeds. Stringent implementation of these regulations is urgently needed. The lack of skilled personnel to certify the setts, extension services, and laboratory facilities in sugarcane growing countries may limit the implementation of the quarantine laws.

Conclusions and Future Perspectives

The red rot pathogen, *C. falcatum*, is a major threat to the sugar industry. It is believed that the inocula resides in crop debris, infected soils and infected setts, resting appressorial cell, conidia and mycelia. The genetic make-up of this fungus varies, making the management of red rot disease in sugarcane plantations difficult. Extensive disease testing using ELISA has been conducted and verified using PCR assay. Specific antibodies are required to avoid false positive and negative. The use of DNA-based nanosensors and DNA microarrays is also promising because these technologies are easier to adopt, they are more reliable and more cost-effective compared with the traditional PCR-based techniques. Non-molecular approaches such as screening for hyperspectral reflective data are being studied with some degree of success, but there is a long way from achieving accurate identification. Although biocontrol methods are promising, they require

extensive field evaluation to develop bio-formulated products. It may be argued that both DNA fingerprinting and genome sequencing are ideally placed to include the evidence that is crucial for promoting phylogenetic and systematic research (until natural remedy to disease resistance becomes a reality). The currently the sugarcane planters are focused on the management of red rot disease through eliminating diseased materials that blur cultural traditions. Until *C. falcatum* resistant varieties are fully development, sanitation is the most practical red rot disease management method. In addition, an intense genome project on *C. falcatum* is urgently needed. Nonetheless, the occurrence of disease destruction has been considerably reduced as the field disease management has been well established. To sustain the sugarcane industry, intensive breeding work on coming out with red rot disease resistant sugarcane variety or developing biologic control technologies are essential.

Conflicts of Interest

The authors declare no conflict ofinterest.

REFERENCES

Apan, A.; Held, A.; Phinn, S.; Markley, J. Detecting sugarcane 'orange rust' disease using EO-1 Hyperion hyperspectral imagery. *Int. J. Remote Sens.* **2004**, *25*, 489–498.

Arade, P.C.; Singh, P.; Mahatma, M. Characterization of *Colletotrichum falcatum* Went. Causing red rotin sugarcane complex. *Bioscan* **2014**, *9*, 375–379.

Imtiaj, A.; Alam, S.M.; Islam, A.K.M.R.; Alam, S.; Lee, T.S. In vitro studies on *Colletotrichum falcatum* the causal of red rot disease of sugarcane. *Eurasian J. Agric. Environ. Sci.* **2007**, *2*, 511–517.

Khalid, A.I.; Bukhari, K.; Nithya, V.; Valluvaparidasan, V.; Paranidharan, R.; Velazhahan, M. Detection of *Colletotrichum falcatum* causing red rot of sugarcane by enzyme linked immune sorbent assay. *Arch. Phytopathol. Plant Prot.* **2012**, *45*, 823–830.

Nandakumar, M.; Malathi, P.; Sundar, A.R.; Viswanathan, R. Use of Green Fluorescent Protein Expressing *Colletotrichum falcatum* the Red Rot Pathogen for Precise Host–Pathogen Interaction Studies in Sugarcane. *Sugar Tech* **2020**, *22*, 112–121.

Nayyar, S.; Sharma, B.K.; Kaur, A.; Kalia, A.; Sanghera, G.S.; Thind, K.S.; Sandhu, J.S. Red rot resistant transgenic sugarcane developed through expression of β-1, 3-glucanase gene. *PLoS ONE* **2017**, *12*, 1–16.

Padhy, J.B.; Kumar, D.D.; Manish, L.; Lavanya, C. Leaf Disease Detection Using K-Means Clustering and Fuzzy Logic Classifier. *IJESTA* **2016**, *2*, 5.

Patel, P.; Shah, R.; Joshi, B.; Ramar, K.; Natarajan, A. Molecular identification and biocontrol activity of sugarcane rhizosphere bacteria against red rot pathogen *Colletotrichum falcatum*. *Biotechnol. Rep.* **2019**, *21*, e00317.

Rahman, M.S.; Khatun, K.; Rahman, K. Sugarcane and sugar industry in Bangladesh: An Overview. *Sugar Tech* **2016**, *18*, 627–635.

Scindiya, M.; Malathi, P.; Kaverinathan, K.; Viswanathan, R.; Sundar, A.R. Molecular characterization of pathogenicity gene homologs in *Colletotrichum falcatum* causing red rot in sugarcane. *Sugar Tech* **2017**, *19*, 563–572.

Sharma, R.; Tamta, S. A Review on red rot: The "cancer" of sugarcane. *J. Plant Pathol. Microbiol.* **2015**, *1*, 1–8.

Singh, R.K.; Kumar, P.; Tiwari, N.N.; Singh, S.P.; Tiwari, A.K.; Kumar, A. Role of endochitinase gene and efficacy of *Trichoderma* against *Colletotrichum falcatum* Went. Causing red rot disease in sugarcane. *Sugar Tech* **2014**, *16*, 180–188.

Singh, K. and Singh, R.P. Red rot. In *Sugarcane Diseases—Major Diseases*; Red, R., Ricaud, C., Egan, B.T., Gillaspie, A.G., Hughes, C.G., Eds.; Elsevier: Amsterdam, The Netherlands, 1989; pp. 169–188.

Viswanathan, R.; Padmanaban, P.; Selvakumar, R. Emergence of New Pathogenic Variants in *Colletotrichum falcatum*, Stalk Infecting Ascomycete in Sugarcane: Role of Host Varieties. *Sugar Tech* 2019, *22*, 473–484.

PRODUCTS AND BY-PRODUCTS OF SUGARCANE INPAKISTAN

Muhammad Ehsan Khan
Sugarcane Research and Development Board Pakistanehsankhanuaf@gmail.com

ABSTRACT

Sugarcane is one of the leading commercial crops of Pakistan and plays a significant role in national economy by sustaining as largest organized agroindustry. Sugar sector contributes significantly in revenue generation and sustainability to our GDP. Handling and management of these byproducts are huge task because those require lot of space and storage. As the sugarcane plant growth advances toward maturity, sugar is gradually stored in cane stalks. During harvesting mature cane stalks are possibly cleaned of tops and trash and brought to the sugar factory. For sustainable growth in income of sugarcane farmers, it is essential that sugar and by-products witness higher growth as compared to the growth in revenue from sugarcane. The current study highlights the demands of sugarcane by products and their effective utilization for profitable and sustained income to sugar industry.
Key words: Sugarcane, Byproducts, Pakistan

INTRODUCTION

Sugarcane is one of the leading commercial crops of Pakistan and thereby the largest sugar market of the world in terms of volume. Since from 1947, when sugarcane production was traced in Pakistan (12.8 million tonnes), and today, where Pakistan is the sixth largest producer in the world, sugarcane production was (86.96 million tonnes) has come a long way. Owing to the agro-climatic suitability of cane cultivation and subsequent development of sugar industry, sugarcane cane cultivation in Pakistan has seen rapid stride. Widely accepted as the original home of sugarcane (Saccharam species) and world's largest consumer (8^{th}), area under sugarcane cultivation was 1.164 million ha, production of 80.96 million tonnes with productivity of 69.55 tonnes/ha (Annual Report of PSMA, 2021).

sugarcane producing country in the world. With decreasing amount of sugarcane production the next five major countries were India, China, Thailand, Pakistan and Mexico (Sarwar *et al.,* 2010). In Pakistan, after textile industry, sugar industry is the second largest industry. Its importance in day to day life adds its value. In this respect, it has the lot of importance in Pakistan's Agriculture.

Sugarcane is considered as the crop for the future and contributes significantly to the GDP. Out of this, nearly 60% is paid to the sugarcane farmers by the sugar mill as prices of cane. Sugar mills process the harvested sugarcane and has the benefits of obtaining multiple products and by-products which are the potential raw materials for several (the extractive,

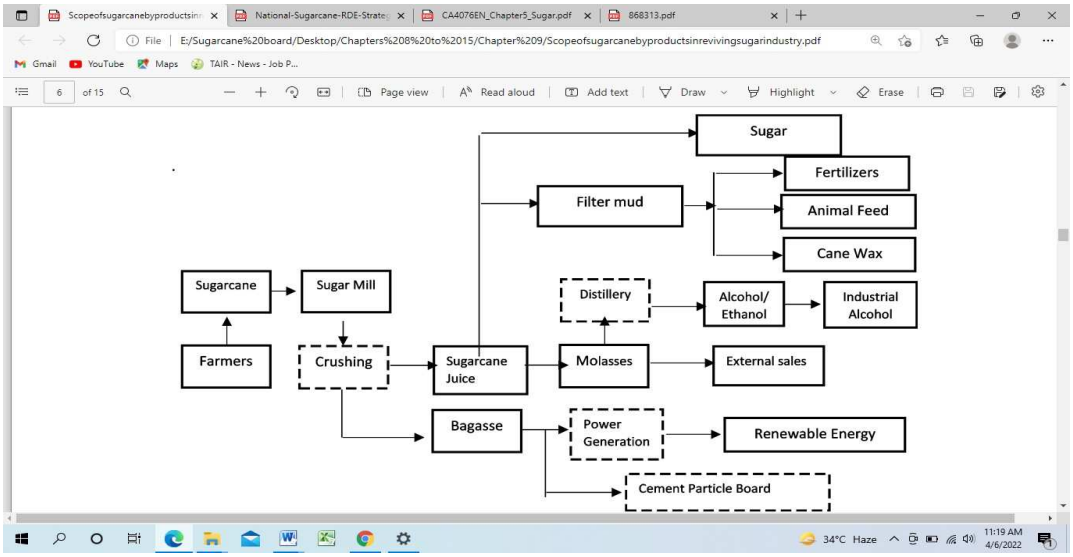
Brazilicalis and bio-chemical) industr

and power. Despite the growing importance of sugarcane, there are some inherent challenges in this sector.

Commercial Uses

Sugarcane once harvested from field goes to crushing, and the main product obtained is refined sugar, by processing its sucrose content. During the processing of sugarcane in a sugar mill, a set of by products are produced. These include, bagasse, molasses, ethanol and filter mud/Press mud. It is estimated that 100 tonnes of sugarcane produce 14.3 tonnes of raw sugar, 30 tonnes bagasse, 5.2 tonnes filter cake, 2.6 tonnes molasses and 50.7 tonne wastewater (Allen *et al.,* 1997, Partha, N. *et al,* 2016). The process of extraction is outlined below;

Figure-1 By Products of Sugar Industry



Source: Chakraborthy, M. and P. S Priya (2020).

Bagasse

It is the dry fibrous residue left after the juice extraction from cane stalks. During the milling process in mill tandem it is separated aside and stocked for further use to release steam and generate electric power for sugar mill's operation in a number of countries like Australia, Brazil and Mauritius (Deepchand, 2016). Sugarcane produces two types of bio-mass, sugarcane bagasse and cane trash. Fresh cane bagasse is 30–32 % of the weight of cane crushed and is composed of 48 % fiber (having 50 % moisture), and 2 % soluble matters. Complete analysis of fiber shows the following ingredients (Patorau, 1986).

Cellulose: 45 - 50 %,
Hemicelluloses: 20 - 25 %
Lignin: 18 - 24 %,
Sugars: 2 %
Wax: 1 %,
Ash: 1 — 4 %

Bagasse use as energy:

sugar mills; when burnt it produces sufficient heat for production of steam to use as energy and generate electricity for various mechanical and processes operations. According to Patorau, 1986, a typical sugar factory requires 35 kWh and 450 kg of steam per ton of cane. A modern factory would require 30 kWh with 300 kg steam per ton of cane and save 50 % of its bagasse. According to Isabirye *et al.,* (2013) the energy output of bagasse is shown as under:
Bagasse production: 30 % of cane crushed
One ton of bagasse produces 2 tons of steam
5 ton of steam produces 1.0 Mwt of electric power
Thus, 2.5 ton of bagasse would produce 1.0 Mwt of electric power.
On crushing cane of 13 % fiber usually 20 % of bagasse is saved as surplus
Introduction of new technologies has made it possible to have maximum efficiency in steam production energy production. By inducing some improvement in steam generating system bagasse saving has now been increased to 30 % and by using high temperature high pressure boilers with steam turbines, bagasse savings are reported to be more than 40%. The new technologies help generate extra electricity with lesser bagasse use. Thus, a sugar mill of 10,000 TDC capacities would produce steam and power of 10–12 MWht for its own variable need and at the same time, it saves huge stock of bagasse that can extend the operating duration of boilers for co-generation of equivalent load of electricity beyond the cane-crushing season of sugar mills. It has now been made possible to save tremendous quantum of bagasse for co-generation of electricity and its delivery to national Grid.
During the year 2020-21, Pakistan sugar industry crushed 58.60 million tons' cane, while the sugar Industry has the installed capacity to

crush 82.88 million tons' cane in a working season of 135 days (Annual Report of PMSA, 2020-21). On the total installed capacity, the energy production potential is stipulated as under;
Cane crushing capacity: 82.88 million tons' cane
Bagasse production: 30 % of cane crushed = 24.864 million tons
Bagasse saving after meeting its own energy requirements; 40 % =9.9456 million tons

Energy cane

Coal, fossil fuel and wood have been the only source of heat energy in the past. Scientists are looking for new renewable energy resources, as 80 % of total world energy is being supplied from fossils. At constant production and consumption, the present known resources of oil are reported to exhaust in 35 years, natural gas 60 years and coal 150 years (Zafar, 2018). Besides the depletion of fossils fuel, its use creates serious environmentalproblems associated with global warming. With growingneed for alternative energy, other than the fossils fuel, there has been resurgence in interest in biomass of field crops as a renewable energy source. Sugarcane is the most efficient convertor of solar energy into biomass, the bagasse, tops and trash that again become the source of heat release and generation of electricity. Cane breeders are planning for producing multipurpose cane for meeting the requirements of both sugar and energy.
The objective is to develop more vigorous and stout cane to produce more fiber than sugar. Sugarcane varieties typically have 12 – 14 percent sucrose and 13 — 15 percent fiber. Bagasse is obtained to the tune of 30 - 32 % the weight of cane crushed, having50 % moisture. To make best use of sugarcane biomass for energy production, objectives are focused to develop varieties having 10 - 12 % sucrose with 22-24 % fiber. This will help save significant quantum of fiber (bagasse) in the process. The energy contained in cane bagasse is important alternative to address the expected shortage of fuel resources.

Cane trash

The cane trash including leaves and tops represent 15 % of the weight of cane stalk at harvest. Nevertheless, most of this is disposed of through burning and creating environmental pollution problem. In mechanized harvesting, cane crop in some countries is yet burnt ablaze and the next day, cane stalks are mechanically harvested for supply to sugar mills. In case of manual harvesting cane after harvesting is manually cleaned of its tops and trash. Some of the tops are taken away for animal feed and a little trash stays in the field. These tops and trash if collected from cane field can be utilized as a viable fuel supplementary to bagasse for combustion and conversion as co-generation of energy into heat or electricity. Some sugar mills in India manage to collect trash from cane fields and prepare 20 kg weight compact blocks and mechanically thrust these into the steam boiler hole for combustion (Malik, 2005).
In case of green cane mechanized harvesting almost 68% of cane trash is blown out of the cane and stays in cane field as trash blanket, while 32 % is taken to sugar mill together with cane as extraneous matter (Zafar, 2015). Researchers are planning to collect this field trash as bails and utilize it for energy purposes. About 7–12 tons of cane trash can be obtained from one hectare and every ton of sugarcane trash contains 5.4 kg N, 1.3 kg P2O5 and 3.1 kg of K_2O and small quantity of micronutrients (Sing and Suleman, 1995). To enhance microbe's activities a layer ofpress mud compost can also be spread on cane trash with one bag of Urea per hectare,as a starter. In addition to thermal and electrical energy, which is obtained from sugarcane bagasse, dozens of by-products are being developed from this raw material. Some of the by-products produced from bagasse are mentioned hereunder:

Paper, Board, Pulp:

Bagasse isutilizedto manufacture different types of hard and soft boards, which are commercially used as a substitute of wood. Its use is very common for preparation of wide range of particleboards including win boards hard boards, and MDF boards. For the manufacture of furniture, doors and window and Amirah's, these boards have greatly replaced wood. This is getting cheap and more efficient than wood. Bagasse is further biodegraded for the production of pulp. Pulp is also utilized for production of writingpaper and tissue paper, newspaper and preparation of

boxes and molds.

Xylose sugar– xylitol, Erythritol

With some digestive techniques, it is getting common to produce energy free sweeteners from bagasse. Through microbial process, using yeast, hemicellulose from bagasse is hydrolyzed to xylose, which is then hydrogenated to produce xylitol. Xylitol is low caloric organic sweetener and is specifically used by diabetic patients. It has 40% low calories than sugar, prevents weight gain, and is a good alternative to sugar. Another low-calorie product erythritol has 0.25 calorie per gram compared to 4 calories per gram of sugar, it tastes very close to sugar. Glycemic index of erythritol is '0' compared to '6' in sugar.

Bio-plastic

Through some chemical degradation, sugarcane-bagasse is transformed into a commercial bio plastic product named as 'Bio cycle'. It is used to produce auto parts, packaging material, toys, credit card, tetra packs and bottle packing of some beverages (Coca Cola), Kitchenware, cutlery ware and disposable shopping bags. Unlike petroleum-based plastics, this product is completely biodegradable and compostable. Sugarcane ethanol has also emerged as an important ingredient to substitute for petroleum in the production of plastic. It has same physical and chemical properties similar to regular plastic. Tiles, prepared from bio-plastic have gained household importance and are being used in kitchens and toilets.

Furfural

Bagasse can be transformed into furfural, which is a starting point for a large number of resins. It can be used to produce furfural alcohol, pharmaceuticals, mono chloro-acetic acid, propionic acid, maleic anhydride and some herbicides. Furfural is a colorless, inflammable, volatile aromatic liquid. It has many industrial uses, such as solvent for refining of lubricating oil. Also used in nylon production, as well as molding powders. Also used to produce furfural alcohol, which is utilized in pharmaceuticals, fungicides, pesticides and solvents.

Tops and trash

During harvesting, tops and leaves of cane stalks are left in the field. These are 15 to 25 percent of cane plant, including 5 to 7 % dry leaves and 13 to 20 % green tops. In early harvesting when cane is not fully mature, tops and trash constitute 20 - 25 %; with advance in maturity, this section is reduced to almost 15%. The cane juice is processed for its boiling, clarification, evaporation, condensation, crystallization to sugar and formation. The main products obtained during these process operations are sugar, bagasse, molasses and press mud. The quantitative output of these products from cane crop are displayed in the following figure. General output of a typical cane plant is as under; Cane: 100 tons (including 15-20 tons' tops and trash Sugar: 10 tons Bagasse: 30 tons (20 tons used for generating energy for sugar mill operations, 10 tons used for other uses or cogeneration). Molasses: 4 - 4.5 tons Filter press cake: 3 - 3.5 tons Water (in cane juice): 63 tons. Exhausted as steam during boiling and evaporation of juice; recycled for use during various processes. To be brief, leaving trash in the ield, Sugar, bagasse, molasses and press mud are the main products of sugarcane. Sugar is marketed as such and is consumed in various ways. As for other products, through advancement in innovations dozens of commercial by- products have been formulated. Biotechnologies have been made available for the production of several by-products like ethanol, acetic acid, yeast, wax, xylose sugar, pulp, paper, boards, bio-plastic, furfural etc. Thus, besides direct use of the products several Co-product industries have been established. Now, by-products have more economic importance than sugar itself. Economic importance of Products and By-products of commercial importance are briefly mentioned as under;

Molasses

Molasses is dark viscous effluent obtained during preparation of sugar in its final crystallization and centrifugal stage. It is the residual syrup, from which crystalline sugar cannot be obtained by simple means. Normally, molasses yields 4–4.5 % of the weight of cane crushed in the factory. Simple composition of molasses is as under (Patorau, 1986);

Water: 20 % other carbohydrates: 4 Sucrose: 35 % Nitrogen compounds: 4.5 % Fructose: 9 % Ash: 12 % Glucose: 7 % Besides converting it to a number of by-products, it is directly utilized for producing ethanol, as an ingredient in cattle and poultry feed and as molasses-based fertilizer in field and garden crops.

Ethanol (Ethylalcohol)

The molasses produced is just 4-4.5 % of the weight of cane crushed, appears to be a small fraction but is of great economic significance. Molasses containing a large fraction of fermentable sugar, is diluted three times with water and allowed to ferment in the presence of yeast culture (Saccharomyces cerevisiae), either by batch or continuous process of fermentation. The process completes in three continuous phases at around 27^0 C temperatures. On completion of the process in the 'still', alcohol vapors are removed as rectified spirit or ethanol, through fractional distillation in a specified column under reduced pressure. The solid and slurry remains at the bottom is spent wash/slops or vinasse and is composed of un-fermentable sugars, water-soluble amino acids, lignin and other organic fractions. In present day economics, major role of molasses is the production of ethanol. One ton of fair quality molasses produce 240 liter of ethanol. It can also be directly produced from cane juice. Ethanol produced is 72 liters per ton of cane or 100 liters per ton of juice. This ethanol is at present controlling the world trade as power alcohol. Brazil, which is the main supplier of sugar in the world trade, is at the same time a large producer of Alcohol. This alcohol is mixed with petrol at 20 % ratio to make the gasoline, which is more environmentally friendly. In case, over-production of sugar creates glut in the world trade, Brazil supports its economy by converting its market to ethanol, so much so that cane juice is directly fermented to produce ethanol without producing sugar. It may be emphasized that Government of Brazil has made it mandatory to blend 20 to 25 percent anhydrous ethanol with gasoline (Fabio and Matoso, 2015) and sugarcane ethanol represent 17.6 % of the country's total energy consumption (Anon,2009).

During 2020-21, Pakistan Sugar Industry produced 2.69 million tons of molasses (Annual Report of PSMA, 2021). The country has made a considerable breakthrough in producing ethanol from molasses. At present 21 distilleries are known to be in operation, which consume a large quantum of molasses produced from sugarcane and sugar beet in the country. These distilleries have the daily ethanol production capacity of 2.65 million liters. (Table- 1). Subject to prevailing market rates, ethanol or molasses have proved a big source of foreign exchange earnings in the country.

Yeast:

Yeast are complex, protein rich living unicellular organisms. Two types have been isolated, Saccharomyces cerevisiae to produce baker's yeast and Torub utilis to produce food yeast. 4 Kg. of molasses is required to produce 1 Kg. of dry baker's yeast.

Table-1 Names of Sugar Mills having ethanol production unit in Pakistan

Sr. No.	Name of Sugar Mills having ethanol production unit	Installed capacity, liters per day
1.	Al Abbas sugar mills and distillery Ltd., Mirwah, Mirpur Khas	165,000
2.	Ansari sugar mills and distillery Ltd., Maatli	100,000
3.	Chashma sugar mills and distillery Ltd., Dera Ismail Khan.	100,000
4.	Colony sugar mills and distillery Ltd., Phalia.	125,000
5.	Crystaline Chemical Industries, Sargodha.	100,000
6.	Dewan sugar mills and distillery Ltd., Dewan city, Sujawal.	125,000
7.	Frontier sugar mills and distillery Ltd., Takht Bhai	25,000
8.	Habib sugar mills and distillery Ltd., Nawabshah.	150,000
9.	Haseeb Waqas sugar mills and distillery Ltd., Nankana.	125,000
10.	Hunza sugar mills and distillery Ltd., Shahkot, Faisalabad.	125,000

11.	Khazana sugar mills and distillery Ltd., Peshawar.	25,000
12.	Matyari sugar mills and distillery Ltd., Matyari.	100,000
13.	Noon sugar mills and distillery Ltd., Bhalwal.	80,000
14.	Premier sugar mills and distillery Ltd., Mardan	46,000
15.	Premier Chemical Idustries, Sheikhupura.	425,000
16.	Shakarganj mills and distillery Ltd., Jhang.	325,000
17.	Shah Murad sugar mills and distillery Ltd., T.M. Khan.	125,000
18.	Saleem sugar mills and distillery Ltd., Charsada.	40,000
19.	Tandlianwala sugar mills and distillery Ltd., Kanjwani	125,000
20.	Unicol Pvt. Ltd. Mirpur Khas.	100,000
21.	United distillery Ltd., Sadiqabad.	120,000
	Total	2,651,000

Source: K. B Malik (2020).

Spent wash

It is also known as Stillage, spent wash, vinasse or effluent. It is a waste product of distillery industry producing ethanol and is produced at the rate of 13 liters per liter of ethanol. It is caramelized and cumbersome effluent, very difficult to handle due to very high BOD (40,000 ppm) and COD (80,000 - 100,000 ppm). It requires higher oxygen concentration for oxidation of the organic matter contained in it, therefore when it is discharged to a drain or river it ex-haust the dissolved oxygen affecting the flora and fauna present in the ecosystem (Pande and Sinha 1997). The organic constituents present in higher concentration undergo reduction, generating unpleasant odor. This is very noxious fluid with pungent smell and pollutes the environments. In its storage in open tanks, it even contaminates the ground water through its seepage and if disposed in open water drains it kills all the aquatic creatures. Spent wash is quite rich in micronutrients; contain large amounts of organic matter, Nitrogen, Phosphorus, Potassium, Sulphur and Calcium, besides high salt load of sulphates and chlorides of Potassium, sodium and calcium. Due to high acidic nature can be used as an amendment in alkaline soils. The economical solution to minimize the pungent smell of spent wash and increase the pH to a desired level is to treat the effluent water in lagoons. Lagoon treated spent wash becomes considerably safe to use as spray on fallow land before land preparation and also to apply in standing crop mixed with irrigation water. The distillery management had installed a project to dehydrate the effluent at high temperature; resultantly spent wash residue is dried as powder. The nutrients contained in the pack are shown in Table-2. During this process of dehydration, the high temperature steam produced is recycled, utilizing it in the sugar mill process house. It exclusively solved the pollution problem of distillery making the environment pollution free for the living being. The powder is rich source of nutrients and is sold in one, two and five kilo packs for orchards and vegetable farming.

Table-2 Composition of the spent wash powder

Nutrient	Percent	Nutrient	Percent
Moisture	9.42	Ferrous	0.02
Nitrogen	2.93	Manganese	0.03
Phosphorus	0.39	Boron	0.02

Source: K. B Malik (2020).

In Pakistan, some of the sugar mills like Habib (Be Nazeerabad) and Shakarganj (Jhang) have adopted sprinkling system of spent-wash on limited scale. The effluent is sprayed on sugarcane press mud and is manually stirred to mix the contents. Repeated application and stirring help to prepare a limited quantum of Bio-fertilizer. However, this technique needs to be improved.

Use of molasses asfertilizer

All the organic and inorganic fertilizers applied to sugarcane during its course of growth phases, are partly absorbed as macro and micro nutrients through plants roots. By termination of growth, a subsequent amount of the nutrients is available in sugarcane biomass and plant solute the cane juice. In case these products are used as fertilizer, all the nutrients contained in, are recycled back into soil. Detailed chemical composition of molasses is reproduced in Table-3.

Table-3 Composition of final cane molasses

Contents	Percent	Contents	Content %	Nitrogen %
Water	18.85	Aspartic acid	0.3740	0.0397
Total solids	81.15	Serine(asparagine)	0.5415	0.0722
Total sugars	48.87	Glutamic acid	0.0332	0.0032
Sucrose	31.76	Proline	0.0086	0.0011
Invert sugars	15.44	Glycine	0.0068	0.0014
Apparent purity	30.08	Alanine	0.0769	0.0124
True purity	39.14	Valine	0.0263	0.0033
Brix	89.76	Isoleucine	0.0118	0.0013
Pol	27.0	Leucine	0.0059	0.0006
Organic non-sugars	24.89	Tyrosine	0.0380	0.0029
Nitrogen	0.90	Phenylaline	0.0322	0.0027
Inorganic constituents (ash)	13.82	Cysteine	+	

Source: K. B. Malik (2020).

CONCLUSION

Sugarcane is one of the most highly remunerative crops which has encouraged farmers to expand acreage and increase production over the years. There is a dire need to expend the use of sugar industry waste. It is suggested that there is significant scope of expanding the growth of sugarcane by-products In Pakistan, use of sugarcane by-products were limited as compared to other countries like Brazil and India. A larger focus on full potential use ofby-products will have a major bearing on the future potential of sugarcane crop.

REFERENCES

Allen, C.J. et al. (1997), "New technologies for sugar milling and by-product modification", in: Keating, B.A. and J.R. Wilson (eds.) in Intensive Sugarcane Production: Meeting the Challenges Beyond 2000, Proceedings of the Sugar 2000 Symposium, CABI, Wallingford, United Kingdom., pp. 267-

285.Mackintosh, 2000.

Annual Report of Pakistan Sugar Mills Association 2020-21.

Chakraborty, M. and P. S. Priya (2020). Scope of Sugarcane By-Products in Reviving Sugar Industry. Indian J. of Res. Volume (10): 1 January-June 2020.

Deepchand, K. (2016), "Sugar cane bagasse energy cogeneration - Lessons from Mauritius", Parliamentarian FArvind Kumar Swarnakar, IRJET Vol 3 Issue 7, July 2016.

Fabio, A. and F. Matoso (2015). Ethanol content in gasoline rises today.Global.com 22.3. 2015.

Isabirye, M.; Raju, D.V.N.; Kitutu, M.; Yemeline, V.; Deckers, J.; Poesen, J. (2013). Sugarcane biomass production and renewable energy. Published in Intech Open.

Malik, K. B. (2020). Cane and Sugar Production. Published by Punjab Agricultural Research Board. Technologies for the prosperity of agricultural stakeholders. Pp. 52.

Malik, K.B. (2005). To standardize a uniform formula for sugar recovery assessment in individual cane samples. Proceedings of Workshop on Agriculture and Process. Pakistan Society of Sugar Technologists.pp 95-117.

Manohar, Rao, P.J (1997). Industrial utilization of Sugarcane and its Co-products. ISCPK publishers and distributors. Delhi, India.

Pande, H.P and B.K. Sinha (1997). Using the distillery waste as fertilizer. Sugarce, Agribusiness Alternatives. Oxford and IBH Publishing New Delhi.

Partha N., and V. Subramanian (2016), "Recovery of chemicals from Press mud-A Sugar Industry waste". Indian Chemical Engineering. Section A, Vol 48, No 3.

Patorau, J.M (1986). Alternative use Sugarcane and its By-products in Agro-industries (1986). Experimental consultation on sugarcane as feed, AGA, Santo Domingo, Dominican Republic.

Sarwar G, Schmeisky H, Hussain N, Muhammad S, Ibrahim M, Safdar E (2008) Improvement of soil physical and chemical properties with compost application in rice-wheat cropping system. Pak J Bot 40:275–282.

Sarwar MA, Ibrahim M, Tahir M, Ahmad K, Khan ZI, Valeem EE (2010) Appraisal of press mud and inorganic fertilizers on soil properties, yield and sugarcane quality. Pak J Bot 42(2):1361–1367.

Sing, J.B. and S. Suleman (1995). Sugarcane Agro Industrial Alternatives. Oxford and IBMm Publishing Company, New Delhi.

Zafar, S (2015). Sugarcane trash as biomass resource. BioEnergy Consult, 29-6-2015.

Zafar, S. (2018). Sugarcane Trash Vs Biomass Resources. BioEnergy Council, Powering clean penergy future -17 April, 2018(From Net).

USE OF CELL AND TISSUE CULTURE IN SUGARCANE PLANT IMPROVEMENT

Muhammad Ehsan Khan* and Kanza Khan**
*Sugarcane Research and Development Board (Punjab), Faisalabad.
**University of Agriculture, Faisalabad.
Corresponding Author email: ehsankhsrdb@gmail.com

ABSTRACT

Sugarcane is an industrial crop and plays a considerable role in the world economy. Almost, 80% of the world sugar is produced from sugarcane and the rest is from sugar beet. In 1960's biotechnological research work start on sugarcane crop with in vitro culture worldwide. Efforts to improve sugarcane crop by molecular applications have commenced in last five years in Pakistan. As sugarcane crop have limitations such as complex genome size (2n = 120-180), narrow genetic base, poor fertility, susceptible to biotic and abiotic stresses and long duration to breed elite genotypes. It is a suitable candidate for application of biotechnology and genetic engineering tools. Biotechnological applications for sugarcane plant improvement have been applied in the areas of: (1). Cell and tissue culture for rapid propagation genetic transformation and molecular breeding (transgenic and marker assisted breeding) (2). Molecular diagnostics of sugarcane pathogens (3). Use of molecular markers for development of genetic maps (4). Variety identification and testing and (5). Molecular characterization of various traits. The purpose of this review is to highlight the recent research work done in sugarcane biotechnology in Pakistan with special focus on cell and tissue culture for rapid propagation genetic transformation for sugarcane plant improvement.
Key words: Biological approaches, Sugarcane, genetic improvement, Pakistan

INTRODUCTION

Sugarcane, belonging to the Saccharum spp., is a significant industrial crop and is among the top ten most cultivated crops globally. It contributes more than 70% of the world's sugar and is a vital raw material for sugar-producing and allied industries. Pakistan is the 5th largest leading producer of sugar, including traditional cane sugar sweeteners, khandsari, and Gur, production. The Saccharum complex comprises crucial sugarcane genotypes derived from S. officinarum, S. spontaneum, and S. robustum crosses. Even though conventional breeding has led to agronomically improved cultivars, challenges such as a narrow gene pool, complex genome, poor fertility, and a long breeding/selection cycle make further improvements challenging. Conventional breeding for incremental improvements in economic traits and increased production of sugarcane in Pakistan is hindered due to the unavailability of specific climate for flowering and lack of certain economic traits (Patade *et al.,* 2009).

Furthermore, contemporary sugarcane cultivars have a fluctuating chromosome count (2n=100-120) and infrequently blossom. As a typical glycophyte, sugarcane displays stunted growth or no growth when exposed to salinity, resulting in a yield that is 50% or less than its actual potential. To maintain sugarcane production and enhance productivity, addressing concerns such as tolerance to biotic and abiotic stresses, nutrient management, and improved sugar recovery is crucial. Both conventional and biotechnological techniques have contributed significantly to overcoming some of these challenges. Genetic transformation is necessary, but slow multiplication procedures and declining varietal vigor pose economic and biological problems. Therefore, a rapid, efficient, and callus-free in vitro method for clonal propagation is crucial. This article outlines the development of in vitro culture systems and biotechnological approaches for sugarcane improvement.

Somatic Embryogenesis an in vitro culture systems

Sugarcane has two primary methods of plant regeneration: direct and indirect morphogenesis. With direct morphogenesis, plants are regenerated directly from tissues such as immature leaf roll discs and shoot tip culture, which is the primary method for commercial propagation of sugarcane (Suprasanna et al., 2006). Indirect morphogenesis involves the initial culturing of leaf roll sections or inflorescences on an auxin-containing medium to produce an undifferentiated mass of cells, or callus. Somatic embryogenesis techniques have two primary objectives:

(i). the development of a highly efficient method for propagating a large number of uniform plants in less time and possibly at a lower cost than conventional propagation methods; and (ii). a cell culture-based regeneration system useful for genetic transformation.

Embryo genic cultures have also been applied in various areas, such as obtaining virus-resistant plants through somaclonal variation, mutagenesis and in vitro selection, and developing transgenic plants.

Efforts have been continuously made to refine in vitro morphogenesis protocols for improved efficiency. Literature studies showed that two successfully developed protocols were available for that;

a). direct somatic embryogenesis (DSEM) and

b). indirect somatic embryogenesis (ISEM)

using young leaf rolls and immature inflorescence segments from sugarcane cultivars. In different lab experiments various media combinations of coconut water (CW), kinetin, zeatin, and TDZ were compared to optimize callus growth and regeneration. CW and zeatin were found to be more effective than other growth regulators for callus induction, while CW alone was effective for plant regeneration (Ali and Iqbal, 2012)

Somatic embryo differentiation through partial desiccation

Mutation breeding has made significant contributions, resulting in the development of several mutant varieties. In our work, we aim to improve sugarcane using in vitro culture in combination with radiation-induced mutagenesis (Suprasanna *et al.,* 2007). The combination of soma-clonal variation and in vitro mutagenesis can be advantageous in the rapid isolation of salinity and drought tolerant lines through in vitro selection. Previous studies have used radiation-induced mutagenesis and in vitro techniques to develop salt-tolerant mutants in sugarcane (Ali *et al.,* 2010).

Various steps of a mutation-breeding program can utilize in vitro techniques. Meristematic cells or tissues and mitotically active cells can be propagated under tissue culture conditions to obtain a sufficient amount of material for mutagenic treatments. Intrasomatic competition, which can discriminate against mutagen-affected cells and result in a loss of their cell progenies, can be controlled by modifying in vitro conditions to enhance the competitiveness of mutant cells (Desai *et al.,* 2007). In sugarcane, we have successfully demonstrated the use of partial desiccation for 4-6 hours to stimulate and improve the somatic embryo differentiation and regeneration response of gamma-irradiated embryogenic callus cultures. This method has also been successfully extended to other sugarcane cultivars. Partial desiccation induces water deficit, which stimulates ethylene evolution and may influence morphogenetic response in vitro. Therefore, partial desiccation treatment can be a simple and innovative approach to enhance the regeneration response of higher-dose gamma-irradiated cultures.

Induction of Stress tolerance through priming

Priming techniques have been utilized to hasten the synchronized germination of seeds, improve seedling establishment, stimulate vegetative growth, and enhance crop yield in several field crops, particularly under sub-optimal conditions such as salinity stress (Bruce etal., 2007). Primed plants are believed to exhibit enhanced stress tolerance owing to the activation of cellular defense responses, improved osmotic adjustment, and a better antioxidant system upon exposure to stress(Suprasanna et al., 2008). The molecular mechanisms responsible for priming effects are thought to involve the accumulation of signaling proteins or transcription factors, as well as

chromatin remodeling that potentially facilitates faster and stronger responses to subsequent stress exposure. We examined the impact of halopriming on germination and subsequent growth in four sugarcane cultivars with varying salt tolerance. Priming during germination led to an improvement in both the percentage and rate of germination. Two-month-old sugarcane plants subjected to 15 days of isosmotic (-0.7 MPa) NaCl (150mM) or polyethylene glycol (PEG 8000; 20% w/v) stress showed improved growth performance in terms of shoot length, shoot and root fresh weight (Patade *et al.,* 2009). The primed plants also exhibited less salt- and dehydration-induced leaf senescence. Improved osmotic adjustment was found to be more crucial than antioxidant capacity in facilitating growth under stress conditions. Expression analysis of stress-responsive genes revealed up-regulation of NHX and down-regulation of SUT1, P5CS, and PDH. Our findings through review suggest that halo-priming can be an effective approach for enhancing abiotic stress tolerance in sugarcane (Patade *et al.,* 2010).

Transgenic Sugarcane

The potential applications of gene transfer, leading to the creation of transgenics, are rapidly expanding in sugarcane (Suprasanna *et al.,* 2007). These applications include insect and herbicide resistance, alteration of sucrose content via down-regulation of pyrophosphate-dependent phosphofructokinase and soluble acid invertase gene, and the production of high-value compounds such as pharmaceutically important proteins, functional foods and nutraceuticals, biopolymers, precursors, enzymes, and bio-pigments. Sugarcane can serve as a bio-factory for these products in the near future. The availability of efficient transformation systems provides the opportunity to improve commercially important traits in elite germplasm, ultimately leading to the development of an ideal plant type of sugarcane.

CONCLUSION

The cellular and molecular toolbox available for sugarcane research has created numerous opportunities. Ongoing studies focus on creating novel in vitro culture techniques for quick propagation and developing germplasm with desirable traits. In the near future, the progress in sugarcane biotechnology has the potential to revolutionize plant productivity and commercial outcomes.

REFERENCE

Ali S., J. Iqbal and M. S. Khan (2010). Genotype independent in vitro regeneration system in elite varieties of sugarcane. Pakistan J. Bot., 42(6): 3783-3790.

Ali, S. and J. Iqbal (2012). Influence of physical factors on callogenesis in sugarcane (Saccharum officinarum L.). Sci Int. (Lahore), 24(2): (in press)

Lakshmanan, P., Geijskes, R.J., Aitken, K.S., Christopher L.P. Grof, Bonnett, G.D. and Smith, G.R. Sugarcane Biotechnology: the challenges and opportunities. In Vitro Cell. Dev. Biol.—Plant 41(2005):345–363.

Desai, N.S, Suprasanna, P. and Bapat, V.A. A simple and reproducible method for direct somatic embryogenesis from immature inflorescence segments of sugarcane. Current Science 87(6) (2004a): 764-768.

Suprasanna, P, Patade YV and Bapat, VA. Sugarcane Biotechnology - A Perspective On Recent Developments And Emerging Opportunities. In: Advances in Plant Biotechnology. Rao, G.P., Yipeng, Zhao., Volodymyr, V.R., Bhatnagar, S. (Ed.) Science Publishers, USA. (2007) Pp 313-342.

Suprasanna, P., Desai, N.S., Sapna, G. and Bapat V.A. Monitoring genetic fidelity in plants derived through direct somatic embryogenesis in sugarcane by RAPD analysis. Journal of New Seeds 8(3) (2006):

Desai, N.S., Joseph, D., Suprasanna, P. and Bapat, V.A. Study of elemental variations during somatic embryogenesis in sugarcane using photon induced X-ray probe. Nuclear Instruments and Methods - B (NIMB) 252(2007): 299-302.

Patade, V.Y., Suprasanna, P. and Bapat, V.A. Gamma irradiation of embryogenic callus cultures and in vitro selection for salt tolerance in sugarcane (Saccharum offcinarum L.). Agricultural Sciences in China (2008) 7(9): 101-105.

Suprasanna, P, Rupali, C, Desai, N,S, and Bapat, V.A. Partial desiccation improves plant regeneration response of gamma-irradiated embryogenic callus in sugarcane (Saccharum Spp.). Plant Cell Tiss Org Cult. 92(2008):101–105.

Bruce, T.J.A., Matthes, M.C., Napier, J.A. and Pickett, J.A. Stressful ‘‘memories’’ of plants: Evidence and possible mechanisms. Plant Science, 173(2007): 603–608.

Patade, V.Y., Sujata, B. and Suprasanna, P. Halopriming imparts tolerance in sensitive sugarcane cultivar to salt and PEG induced drought stress. Agri Ecosys. Environ. 134(2009): 24–28.

Patade, V.Y., Rai, Archana N. and Suprasanna, P. Transcript expression analysis of sugarcane shaggy-like kinase (SuSK) gene identified through cDNA subtractive hybridization in sugarcane (Saccharum officinarum L.). Protoplasma 248 (2010), In Press

A REVIEW ON THE IMPACT OF CLIMATE CHANGE ON SUGARCANE CROP

Kanza Khan* and Muhammad Ehsan Khan**
*University of Agriculture, Faisalabad **Sugarcane Research and Development Board, Punjab
Corresponding Author email: kanzakhan3346@gmail.com

ABSTRACT

Sugarcane, scientifically known as *Saccharum officinarum* L., is a large perennial grass belonging to the Gramineae (Poaceae) family and is a monocotyledon. This C_4 agro-industrial crop is cultivated widely in tropical and sub-tropical regions around the world. The crop requires a long duration of 10-15 months, and in some cases up to 18 months, to mature, which depends on the geographical location. There are various factors that affect the growth and development of sugarcane, with climate change being a major factor that can either positively or negatively impact the crop. Among the many climatic factors, rainfall and temperature are the most crucial weather parameters for sugarcane productivity. However, with the increase in temperature and CO_2, global warming is leading to a deficit in rainfall, resulting in a decline in crop production. Therefore, it is crucial to assess the effects of climate change on crop growth to maximize the potential yield of all sugarcane varieties in the future.
Keywords: Climatic transformation, Control, Sugarcane, Pakistan

INTRODUCTION

Sugarcane, also known as *Saccharum officinarum* L., is a C_4 crop that holds a crucial position in the Pakistan economy. It is a significant cash crop of the country, cultivated over an area of approximately 1260 thousand hectares, including both tropical and sub-tropical regions. Sugarcane is a tall perennial plant that grows upright up to 5-6 meters and produces multiple stems, also known as canes. The crop undergoes four distinct growth phases: Germination phase (up to 45 days after planting-DAP), Tillering phase (45-120 DAP), Grand growth phase (120-270 DAP), Maturity and Ripening phase (270-360 DAP), with each phase having specific climatic requirements (Moore and Frederik, 2014). Sugarcane thrives in regions with a tropical or sub-tropical climate, with a mean temperature range of 28-32°C being best suited for its growth. Temperatures exceeding 45°C reduce tillering and growth, while temperatures below 20°C may slow down growth. Areas with a minimum temperature below 5°C are not suitable for sugarcane cultivation. A relative humidity of 70-85% during growth and 55-75% during the ripening phase is ideal. Relative humidity below 50% during the growing season is unsuitable for sugarcane cultivation.

Brazil is the largest cane growing country, followed by India, China, Australia and Pakistan with nations accounting for more than 70 % of the global cane acreage in 2021-22. Among Punjab states, Muzafargarh, contributes around 44% of the total sugarcane production (133.40 million tonnes), making it the largest sugarcane growing state in the country. Climatic factors play a crucial role in the productivity of a variety and the crop. Climate change could impact agriculture in various ways, such as affecting productivity, growth rates, photosynthesis, transpiration rates, and ultimately, the quality and quantity of the product. Climate change is expected to directly affect food production worldwide. An increase in the mean seasonal temperature could reduce the duration of many crops and result in a decline in their yield.

The warming temperatures, which are already nearing the upper limit for crops, will have an immediate impact on crop yields. Sugarcane is particularly vulnerable to changes in temperature and rainfall, with even slight deviations from normal weather patterns during different growth stages leading to significant losses in yield and sugar production (Glasziou et al. 1965; Mali et al. 2014;

Zhao and Li, 2015). Climate change, caused by various factors such as population growth, industrialization, and deforestation, has been leading to a steady increase in mean minimum temperature across all climatic regions in Pakistan, as reported by the Pakistan Meteorological Department (IMD) (Rathore et al. 2013). Such changes in temperature and rainfall patterns may have a negative impact on dry matter and sugar accumulation in the future, necessitating a shift in planting, harvesting, and crushing schedules. Water availability and temperature regimes are crucial in determining the yield and quality of sugarcane crops worldwide (Zhao and Li, 2015). The concentration of greenhouse gases has also surpassed the highest levels recorded over the past 800,000 years, leading to increased rainfall, irregular distribution of rainfall, hot extremes, floods, droughts, cyclones, and glacial retreats. For any agricultural commodity, variation in yield is closely tied to growing-season weather, which can impact pests, diseases, and weeds, ultimately affecting production and productivity.

Climatic requirement of the growth of Sugarcane

The growth of sugarcane crop is influenced by various climatic requirements. The crop requires a yearly rainfall of 1500-2000 mm to produce 100 ton millable cane. For plant crop, the average water requirement is 88kg water/kg of cane and 884kg water/kg of sugar. In Punjab districts, the average water requirement per hectare for the entire sugar season is 57 lakh liters. During different growth phases, the water requirement for subtropical zones is 17% at the germination phase, 24% at the tillering phase, 37% at the grand growth phase, and 22% at the maturity phase. Whereas, in tropical weather, the water requirement is 12% at the germination phase, 22% at the tillering phase, 40% at the grand growth phase, and 26% at the maturity phase (Bhardwaj, 2013). The crop needs adequate rainfall during the vegetative growth phase to form thick and tall cane, whereas less rain is required during the ripening time to produce good quality juice. The optimal rainfall range for higher cane yield is between 1100-1500mm/yr, but it can also be grown successfully at lower levels, even down to 600mm of annual rainfall.

Temperature:

Different stages of crop require different optimum temperature ranges. The ideal temperature for the germination of cane sett is between 28º-32 ºC, and a temperature above 38 ºC impedes germination and reduces photosynthetic rates, whereas below 32 ºC, it affects germination, resulting in a poor plant population. During the tillering phase between March and June, atmospheric temperatures ranging from 30º to 32 ºC are preferred. The ripening period requires low temperatures in the range of 12º-14 ºC, which reduces the vegetative growth rate and enriches sucrose in the cane. Temperatures below 5ºC and above 35ºC are unfavorable for young leaves and buds. High temperatures can increase abiotic diseases and convert sucrose content into fructose and glucose, reducing sugar accumulation. Sucrose accumulation is facilitated by temperatures below 19 ºC, and the optimum temperature lies between 12º and 14 ºC, while sucrose accumulation has been reported to decline above 26.6 ºC (Clements, 1980; Binbol et al. 2006; Gawander, 2007; Fageria et al. 2010).

Sunlight:

The growth stage of sugarcane, especially the formative stage, is heavily reliant on the intensity of sunlight, which promotes photosynthesis and stabilization ranges. During the cloudy and short days season, tillering is hindered, while 7-9 hours of bright sunshine on average is optimal for tillering, stalk formation, and good crop growth; Fageria et al. 2010). To ensure proper sunlight exposure and maximize yield, adequate spacing between rows and plants is necessary. In sugarcane, the upper six leaves' canopy intercepts 70% of the sunlight radiation, which leads to reduced photosynthesis rates in lower leaves due to mutual shading. In areas with short growing periods, closer spacing is beneficial to intercept more solar radiation and achieve higher yields, whereas wider spacing is recommended for long growing seasons to avoid mutual shading and tiller shoot mortality (Srivastava and Rai, 2012).

Photosynthesis:
The growth stage of sugarcane, especially the formative stage, is heavily reliant on the intensity of sunlight, which promotes photosynthesis and stabilization ranges. During the cloudy and short days season, tillering is hindered, while 7-9 hours of bright sunshine on average is optimal for tillering, stalk formation, and good crop growth; Fageria et al. 2010). To ensure proper sunlight exposure and maximize yield, adequate spacing between rows and plants is necessary. In sugarcane, the upper six leaves' canopy intercepts 70% of the sunlight radiation, which leads to reduced photosynthesis rates in lower leaves due to mutual shading. In areas with short growing periods, closer spacing is beneficial to intercept more solar radiation and achieve higher yields, whereas wider spacing is recommended for long growing seasons to avoid mutual shading and tiller shoot mortality (Srivastava and Rai, 2012).

Relative Humidity and Wind:
Humidity and wind may have a comparatively smaller impact on sugarcane cultivation, but they can still affect the crop significantly under extreme conditions. Warm weather with 80-85% humidity is favorable for rapid cane growth, while a moderate humidity level with limited water supply is ideal for the ripening phase (SC, 2012). Wind does not harm the plant until it reaches a velocity that can cause cane breakage or leaf damage. However, high velocity wind can be harmful in the initial growth stage and can cause moisture loss if it persists for a long duration.
Generally, two sets of climatic parameters are required in the sugarcane plant's life cycle. The grand growth phase, spanning from July to September and coinciding with the monsoon season, requires long durations of bright sunshine, warm temperatures (28-32 ºC), optimum rainfall, and high humidity for rapid growth of both the plant and cane length, leading to a good yield. However, during the ripening season, which is a phase of sugar storage, clear skies without precipitation, warm days, and dry weather conditions with a relative humidity of about 51% are needed to achieve 12% cane weight and 15% cane height (Srivastava and Rai, 2012). The evapotranspiration demand risk is very high during the grand growth phase because of the active growth and high water demand, necessitating frequent irrigation using surface water and groundwater resources.
Cane growth slows down after October, and ripening begins when the temperature drops below 19.4 ºC, and relative humidity remains moderate (60-65%). The rapid build-up of sucrose and its accumulation begins in October and continues until December. The final sugar output is influenced by the climate during the maturity phase, which is favored under cold and dry weather conditions with a large diurnal temperature variation and adequate soil moisture (Moore and Frederik, 2014).

Global change in temperature
According to the latest scientific assessment on the earth's climate system, changes have been observed on both global and regional scales since the pre-industrial era. The evidence suggests that human activities are the primary cause of most of the warming (0.1 ºC per decade) observed over the last 50 years (Gautam et al. 2013). The Intergovernmental Panel on Climate Change has projected that the global mean temperature may increase between 1.4 and 5.8 ºC by 2100 (Gautam et al. 2013), which could have severe impacts on the global hydrological system, ecosystems, sea level, crop production, animal husbandry, and related processes. The impacts are expected to be particularly severe in tropical areas, which are mainly comprised of developing countries such as Pakistan. Climate change has been observed at both global and local scales.

Effect of Green Houses:
Greenhouse gases, such as carbon dioxide, methane, nitrous oxide, and halocarbons, have increased significantly since the pre-industrial era, along with a decrease in stratospheric ozone and an increase in tropospheric ozone, leading to direct effects on weather conditions. Additionally, various factors such as sulfate and nitrate aerosols, black carbon and organic matter from fossil fuel burning, biomass burning, mineral dust, land use changes, clouds, solar variability, and stratospheric and tropospheric water vapor

contribute to the increase of greenhouse gases. The impact of extreme weather events on Pakistan agriculture raises questions about the role of human activities in climate change. In recent years, many such events have been linked to the rising levels of greenhouse gases, including the prolonged drought in Australia, the scorching European summer of 2003, the intense hurricane seasons in the North Atlantic in 2004 and 2005, the heavy rainfall in different districts of Pakistan, in July 2005, and others. The concentration of greenhouse gases, particularly carbon dioxide, methane, and nitrous oxide, has increased due to fossil fuel combustion and land use changes. Agriculture contributes significantly to the emission of methane and nitrous oxide (Cerri et al. 2007). The impact of global warming, caused by the greenhouse effect, will lead to changes in temperature, rainfall, solar radiation patterns, and will have both positive and negative effects on sugarcane production (Srivastava and Rai, 2012). Gradual recession of glaciers, floods, droughts, cyclones, frequent hot extremes, and increased rainfall are some of the effects of global warming due to the greenhouse effect.

Effect of Increase in CO_2

It is anticipated that climate change will cause weeds to migrate northward. While most cash crops are negatively affected by increasing CO_2, C3 "invasive" weeds tend to respond positively. Recent research suggests that glyphosate, the primary herbicide used in the Pakistan and other countries, loses its effectiveness on weeds grown in elevated CO_2 environments expected in the future. This migration and proliferation of weeds will adversely affect crop productivity as many weeds, pests, and fungi thrive in warmer temperatures, wetter climates, and higher CO_2 levels. Climate change is likely to increase the range and distribution of weeds and pests. Although rising CO_2 can stimulate plant growth, it also reduces the nutritional value of most food crops by decreasing the concentration of protein and essential minerals in many plant species (Ziska et al. 2014).

Extreme weather effects on Farmers

From 1900 to 2020, Pakistan experienced numerous severe droughts which affected a large part of its population, making it the most significant natural disaster. Since agriculture in Pakistan heavily relies on the monsoon season from June to September for about 75% of its annual precipitation, the variability of monsoon rainfall has a direct impact on food grain production. Extreme weather events like droughts, floods, and hailstorms have a significant impact on agriculture and food security, which is the primary source of income for many rural populations. Such events also damage agricultural infrastructure, soil conditions, water resources, and natural ecosystems, causing significant losses to the economy, with Pakistan annually losing around 2% of its GDP and 12% of central government revenues to natural disasters. The form, frequency, and increasing intensity of extreme events are largely attributed to changes in the earth's climate. Venkateswarlu and Shanker (2012) reported that rainfed agriculture would be more negatively impacted by climate change due to rainfall variability and reduced rainy days. This would have greater implications for farmers' choice of crops, varieties, and cropping patterns/systems than irrigated agriculture. Additionally, studies have shown that for every 1 ºC increase in temperature, water requirements will increase by 10%, which will severely affect productivity and water use efficiency in several crops.

Effect of climate on disease development

Concerns have been raised over the quality degradation and yield reduction of sugarcane. One of the major threats to sugarcane in this region is Pokkah Boeng disease caused by Fusarium moniliformae, which has shown an increasing trend of disease incidence and made most commercial cultivars susceptible. According to Vishwakarma et al. (2013), the severity of airborne disease Pokkah Boeng increases manifold under cloudy weather and high humidity up to 70-80% with favorable temperatures during the rainy season (June, July, August, and September). Unseasonal rain, changes in relative humidity, and heavy dew influence the crop's microclimate and can lead to unpredicted insect and disease

incidences (Sharma et al. 2013). This is the most active growth period where about 80% of cane weight is attained. A shift to a higher thermal regime due to lack of rain during the elongation phase also affects the dynamics of disease and pest attacks, which ultimately influences the cane and sugar yield (Bhardwaj et al. 198).

CONCLUSION

Assessing the impact of climate change on sugarcane growth is crucial for maximizing future yields, as different phases of sugarcane have varying temperature, rainfall, evaporation, sunshine, and humidity requirements. Rainfall and temperature are particularly crucial for sugarcane productivity, and recent observations have shown a decline in rainfall and an increase in temperature during certain growth phases, as well as a rainfall deficit during critical growth stages. Breeding stress-tolerant and low-input varieties, improving soil fertility, and addressing disease and pest scenarios are important future research priorities under changing climate conditions. Climate change is expected to affect sugarcane production directly or indirectly, including through changes in extreme weather events such as droughts. Greenhouse gas emissions may also negatively impact sugarcane production. Although sugarcane is a hardy crop, even slight temperature increases may have negative effects when combined with irregular rainfall patterns. The formative and elongation phases of sugarcane growth are particularly vulnerable to water scarcity, which can greatly reduce yields and productivity. For these reasons, addressing the impacts of climate change on sugarcane growth is of utmost importance for maximizing yields and maintaining productivity in the future.

REFERENCES

Berding N, Hurney AP. Suckering: a faced of ideotype selection and declining CCS in the wet tropics. Proceeding of the Australian Society of Sugar Cane Technology. 2000; 22:153-162.
Binbol NL, Adebayo AA, Kwon-Ndung. Influence of climate factors on the growth and yield of sugar cane at Numan, Nigeria. Climate Research. 2006; 32:247-252. 3. Bhardwaj SC, Gupta JN, Jain BK, Yadav SR. Comparative incidence of stalk borer, Chiloauricilius Ddgn. in autumn and spring planted and ratoon crops of sugarcane. Indian Journal of Agriculture Research. 1981; 15:135-140.
Bhardwaj A. Benefits of micro irrigation system sugar recovery and productivity. Indian Sugar Mills Association (ISMA) 8th oct, 2013, 1-20.
Cerri CEP, Sparovek G, Bernoux M, Easterling WE, Melillo JM, Cerri CC. Tropical agriculture and global warming: impacts and mitigations options. Scientia Agricola. 2007; 64:83-99.
Clements HF. Sugarcane crop logging and crop control. University Press, Hawaii. 1980, 520.
Costantino L, Breon FM. Aerosol Indirect Effect on Warm Clouds over South-east Atlantic, from co-located MODIS and CALIPSO Observations. Atmospheric Chemistry and Physics. 2013; 13:69-88.
Eastman R, Warren SG. A 39-Yr. survey of Cloud Changes from Land Stations Worldwide, 1971-2009: Long-term Trends, Relations to Aerosols, and Expansion of Tropical Belt. Journal of Climate. 2013; 26:1286-1303.
Fageria NK, Virupax C, Baligar, Jones CA. Growth and mineral nutrition of field crop, 3rd Ed. CRC Press. 2010, 437-456.
Gautam HR, Bhardwaj ML, Kumar R, Climate change and its impact on plant diseases. Current Science. 2013; 105(12):1685-1691.
Gawander J. Impact of climate change on sugarcane production in Fiji. WMO Bull. 2007; 56(1):34-39.
Glasziou KT, Bull TA, Hatch MD, Whiteman PC. Effects of temperature, photoperiod duration, and diurnal and seasonal Temperature changes on growth and ripening. Australian Journal of Biological Science. 1965; 18:53-66.
Gryspeerdt E, Stier P, Partridge DG. Satellite observation of cloud regime development: the role of aerosol processes. Atmospheric Chemistry and Physics. 2014; 14:1141-1158.
Hatfield J, Takle G, Grotjahn R, Holden P, Izaurralde RC, Mader T et al. Ch. 6: Agriculture. Climate Change Impacts in the United States: The Third National Climate Assessment, J. M. Melillo, Terese (T.C.) Richmond, and G. W. Yohe, Eds., U.S. Global Change Research Program USGCRP. 2014, 150-174.
Jika M. Improved sugarcane production practices at Savannah Sugar Company Limited SSCL. Presented at Monthly Technical Report Meeting (MTRM), Adamawa Agricultural Development Project AADP, Yola, 1997.
Kumar R. Yield response of sugarcane to weather variations in North-East Andhra Pradesh, India. Arch Met Geoph Biocl Ser B. 1984; 35:265-276.
Kumar A, Tripathi P, Gupta A, Singh KK, Singh PK, Singh R, Singh RS, Tripathi A, Rainfall variability analysis of Uttar Pradesh for crop planning and management. Mausam. 2018; 69(1):141-146.
Lal DM, Patil SD, Singh HN, Ghude SD, Tiwari S, Srivastava MK. Influence of aerosol on clouds over the Indo-Gangtic plain India. Climate Dynamics. 2013; 41:601-612.
Lester R, Gurenko E. Financing Rapid Onset Natural Disaster Losses in India: A Risk Management Approach, impacts on rainfall and temperature in sugarcane growing Upper Gangetic Plains of India. Theoretical and Applied Climatology, 2018. https://doi.org/10.1007/s00704-018- 2378-8.
Vishwakarma SK, Kumar P, Nigam A, Singh A, Kumar A. Pokkah Boeng: An Emerging Disease of Sugarcane. Journal of Plant Pathology amd Microbiology. 2013; 4(3):170-174. 41.
Zhao D, Li YR. Climate Change and sugarcane production: potential impact and mitigation strategies. International Journal of Agronomy. 2015 https://doi.org/ 10.1155/2015/547386
Ziska L, Crimmins A, Auclair A, DeGrasse S, Garofalo JF, Khan AS et al. Ch. 7: Food Safety, Nutrition, and Distribution. The Impacts of Climate Change on Human Health in the United States: A Scientific Assessment. U.S. Global Change Research Program USGCRP (2014), Washington, DC. 2014, 189-216

SUGAR INDUSTRY ABSTRACTS

Dispersion of Sicyos polyacanthus seeds by mechanical sugarcane harvesters in Tucumán, Argentina

Ignacio L Olea

Proceedings of the International Society of Sugar Cane Technologists, volume 30, 1144–1148, 2019

Sicyos polyacanthus (tupulo) is an annual vine, capable of growing over sugarcane's closed canopy and producing 1000-1800 viable seeds per plant that can be disseminated through mechanical harvesters. The aim of this study was to evaluate the spatial distribution of S. polyacanthus seeds in fields as a consequence of mechanical harvesting with two different harvesters. Six experiments were carried out in pre-harvest burned sugarcane fields. Seeds were placed into 12 paper bags with 100 seeds per bag and placed inside the upper third of different stalks of a single plant, located at the center of each plot. In 2017 seeds were tinted with fluorescein sodium salt for visualization with an ultraviolet light source, while in 2018 red paint was added to facilitate daylight seed counting. A CASE 8000 harvester was used in five experiments and a John Deere 3520 was used in one. The sample-area was established in the direction of harvesting in a plot of 30 m by 60 m. The variograms and cross-validation provided the best fit when using a spherical model. Based on that, contour maps were developed using Kriging interpolation in Surfer 8 software. For each of the 6 experiments, dispersion was spatially clumped up to 4.44 m (sill: 0.424, range: 4.44), 4.26m (sill: 0.494, range: 4.26),8.25 m (sill: 1.09, range: 8.25),7.22 m (sill: 0.98, range: 7.22),6.70 m (sill: 1.11, range: 6.70),and 10.18 m (sill: 0.725, range:
10.18), respectively. Beyond these distances the dispersion was random. These results indicate that the initial dispersion was similar for both types of harvesters and models across all experiments, but with some differences in the maximum distance that the seed was spread.

Sugarcane genotypic variation in physiological and yield traits and their relationships

D Zhao, M Irey, C LaBorde and C-J Hu

Proceedings of the International Society of Sugar Cane Technologists, volume 30, 1144–1148, 2019

Sugarcane (a complex hybrid of Saccharum spp.) physiological, growth, and yield traits are important for growers to evaluate cultivars and for breeders to select best genotypes in variety development programs. Little is known about the relationships between physiological traits measured during early growth and final yield components. A field experiment was conducted on a sand soil to investigate physiological and yield characteristics of 18 sugarcane genotypes and to determine relationships between the physiological and yield traits in crops of plant cane, first ratoon, and second ratoon. The physiological traits included leaf SPAD reading, leaf net photosynthetic rate (Pn), and canopy normalized difference vegetation index (NDVI). The yield traits included stalk population, mean stalk weight, stalk diameter and stalk length, cane yield (t/ha, TCH), commercial recoverable sucrose (CRS), and sucrose yield (t sucrose/ha, TSH). Among the three physiological traits, Pn had the greatest variation (CV = 12–16%). Among the yield traits, TCH had the greatest and CRS had the smallest variation across genotypes. Crops also affected the genotypic variation in these traits. TCH, TSH, and mean stalk weight positively correlated to leaf Pn. No correlations were detected between leaf SPAD reading and most yield traits, except for CRS that negatively correlated with leaf SPAD reading across genotypes. Stalk population and TCH most highly correlated with NDVI. Therefore, measurements of leaf Pn or canopy NDVI during sugarcane growth may be useful for predicting yield potential across genotypes.

2G lactic acid production from sugarcane bagasse using Bacillus coagulans NCIM 5648

SV Patil, KS Konde, SD Patil and RV Burase

Proceedings of the International Society of Sugar Cane Technologists, volume 30, 1144–1148, 2019

Lactic acid is one of the most important organic acids and is used extensively around the globe in a range of industrial and biotechnological applications. Lactic acid has great potential for producing biodegradable and biocompatible polylactic acid. Eco-friendly processing using advanced enzymes and the fermentable capability of many of the agro-residues makes them attractive candidates in fermentation biotechnology to develop value-added by-products.

Lignocellulosic raw materials are the most abundant renewable feedstock that has attracted considerable attention as a substrate for biofuels and biochemicals production. Bagasse and sugarcane trash are by- products of the sugar industry with well-established supply chains. We screened three lactic acid producing Bacillus coagulans strains (NCIM 2323, NCIM 2030 and NCIM 5648) on synthetic media at the 500 mL shake-flask scale. Maximum lactic acid production of 65.2 g/L with a productivity of 0.9 g/L/h and a yield of 0.86 g/g of glucose was obtained for Bacillus coagulans NCIM 5648. This was validated on synthetic media for Bacillus coagulans NCIM 5648 at the 4-L fermenter scale with lactic acid titer, productivity and yield of 94.1 g/L, 1.3 g/L/h and 0.94 g/g of glucose, respectively. Bagasse samples of different cultivars of sugarcane were analysed to determine their composition. Of the different pre-treatment methods, acid hydrolysis was used for fractionation of the bagasse components. The pretreated bagasse was subjected to hydrolysis using the Cellic® CTec-2 and Cellic® HTec-2 (from Novozymes) enzyme complex and further fermented using the selected strain Bacillus coagulans NCIM 5648. The lactic acid titre, productivity and yield from HNO3 pre-treated and Cellic® CTec-2 and Cellic® HTec-2 hydrolysed 15% (w/v) bagasse was 37.8 g/L, 0.5 g/L/h and 0.81 g/g of sugar, respectively. This demonstrates the possible use of bagasse for second-generation (2G) lactic acid production using Bacillus coagulans NCIM 5648.

Reduction in bagasse moisture and improvement in mill extraction by controlling reabsorption of juice in mills

MS Sundaram, DS Nikam and PR Ghundre

Proceedings of the International Society of Sugar Cane Technologists, volume30, 1144–1148, 2019

Reduction in moisture % bagasse is always a major task for mill engineers since it leads to significant improvement in boiler efficiency and steam-to-bagasse ratio. Bagasse driers were developed to reduce bagasse moisture, but they require huge capital investment and also consume additional energy. Hence, a new concept was developed to reduce bagasse moisture by controlling the reabsorption of juice in mills. This control of reabsorption has been achieved by providing a modified arrangement of top and bottom roller scrapers with a discharge nip pressure chute. This innovative arrangement is termed a "Moisture Control Unit" (MCU). In this system, bagasse flow after the discharge nip is passed through a controlled passage which reduces excess speed coefficient and forward slip of the bagasse. These actions help in controlling the reabsorption of extracted juice and increase juice drainage at the discharge nip, which directly reduces bagasse moisture. It permits further increase in imbibition, thus improves extraction and reduces bagasse pol, with reduced bagasse moisture. About 60 units are installed in sugar factories in India. Performance of these Moisture Control Units during the last three cane-crushing seasons was studied. The working results after the installation of the MCU on all mills in four or five mill tandems indicate that 2.5-3.0 units reduction in bagasse moisture, 3-4 units increase in primary extraction (pol basis) and 0.4-0.5 units increase in RME are achievable. Reduction in moisture % bagasse also increases GCV of bagasse and ultimately boiler efficiency. The installation of a "Moisture Control Unit" results in reduction of bagasse moisture

coming out from the discharge nip of each mill, which results in reduction of final bagasse moisture with additional drainage of juice and increase in overall mill extraction. These results were accomplished with little investment and without any significant increase in power consumption in the milling tandem.

A decision-support system for determining sugarcane pest reservoirs

Pierre Martin Pierre Silvie, Pascal Marnotte, and François-Régis Goebel

Proceedings of the International Society of Sugar Cane Technologists, volume 30, 1144–1148, 2019

Predicting the establishment of pest reservoirs, and therefore pest infestation in sugarcane agroecosystems, is a challenge for the implementation of integrated pest management (IPM) programs. The objective of this work was to develop a Decision Support System that suggests plant species, located in a production area, that host a pest. A Knowledge Based system on Biological Interaction (KBBI) was then developed and coupled to DECIPESTS, a Decision support system for PEST management in sugarcane. KBBI compiles biological interrelations listed in the scientific literature, at three trophic levels (plant, pest, and natural enemy). DECIPESTS is based on IPM expertise and field observations and identifies the potential causes of insect pest infestations. KBBI and DECIPESTS both use Cogui software to handle and combine pieces of knowledge. According to observed damage, DECIPESTS informs on the potential pest. In return, KBBI suggests the wild and cultivated plants that are able to host the pest. The comparison with a local floristic inventory enables the determination of pest reservoirs. Applied to a case study in Senegal, KBBI and DECIPESTS showed for instance that Eldana saccharina can be hosted by nine wild plant species located in the irrigation canals: Typha domingensis, Sorghum verticilliflorum, Phragmites australis, Paspalum scrobiculatum, Echinochloa pyramidalis, and four species of the Cyperus genus. KBBI also showed that this species can be hosted by Zea mays and Oryza sativa, two neighbouring cultivated crops of socio-economic importance in the area. This latter result indicates that the management of Eldana saccharina has to be tackled jointly by local farmers to be successful. While DECIPESTS uses a tactical approach to identify possible causes of pest infestation, its combination with KBBI makes it a strategic tool to enhance IPM strategy at a local scale.

Treatment of spray-pondoverflow wastewater of sugar mills using microalgae

Amruta Urdukhe, Sunil Dalvi and Deepali Nimbalkar

Proceedings of the International Society of Sugar Cane Technologists, volume30, 1144–1148, 2019

Spray pond overflow is one of the effluents generated from sugar manufacturing processes and has high chemical oxygen demand (COD), total dissolved solids (TDS), total suspended solids (TSS), turbidity and hardness. Until recently it was discharged untreated, resulting in disturbance to the aquatic flora and fauna. It can also damage crops and aquatic vegetation due to the presence ofexcessive nutrients and is totally unfit for livestock to drink. Treatment of spray-pond overflow prior to discharge is now mandatory in India. We focused on algal treatment of spray-pond overflow. Microalgae are used for the bioremediation to reduce contaminants in the waste water due to their ability to assimilate nutrients in the cell. Here, 24- and 48-hour algal treatments were given to spray-pond overflow and physicochemical analysis was carried out by using standard methods. Measured outflow parameters and average reductions were: COD (57.9%), TDS (20.7%), total hardness (20.2%), calcium hardness (32.4%), sulphate (33.5 %), potassium (35.3%), turbidity (77.6%) and TSS (97%). Algal treatment can be an effective pre-treatment method forspray-pond overflow and help to reduce the energy costs of conventional treatment to achieve disposal limits under relevant standards.

Analysis of disease-screening trials for sugarcane mosaic using the Sites Regression model

Y Puchades, M La O Hechavarría, E Rodríguez and M Rodríguez

Proceedings of theInternational Society of Sugar Cane Technologists, volume 30, 1144–1148, 2019

This study was carried out to evaluate the reliability of the trials to estimate resistance to sugarcane mosaic disease in the Sugarcane Breeding Program of Cuba. Experiments were planted in the Jovellanos, Florida, and Guaro localities in two consecutive years. They comprised 10 clones and the controls used in the SCMV resistance studies: B42231 (R), 39MQ832 (Int) and C236-51 (S). Data for the percentage of infection was recorded. Factorial ANOVA (clones, localities and years) and the GGE model showed a strong influence of the environment on the manifestation of the mosaic symptoms. Trials were reproducible between localities and these are divided into two sub-regions. Resistant clones and the B42231 control gave the least stable response, which reinforces the importance of the environment in disease development. These results showed the use of the GGE model as a tool to analyse the reliability of resistance tests to sugarcane diseases developed in multiple environments.

QTL mapping for earlyripening and high-sucroserelated traits in sugarcane

Proceedings of the International Society of Sugar Cane Technologists, volume30, 1144–1148, 2019

In Mauritius, sugarcane growers incur major losses early during the harvest season due to the low sucrose content of sugarcane stalks. Breeding for early ripening cultivars is, however, highly labour and resource intensive. Marker-assisted selection is thus being sought to complement field selection of early high-sucrose sugarcane clones. A population of 300 individuals, derived from a cross between CP 67412 (early ripening/ high sucrose) and M 245/76 (late low sucrose) was established in replicated trials with three harvest dates H1, H2 and H3 for harvesting in May, August and November, respectively. Progress in sucrose content was measured among the trials in two approaches; between the harvest dates H1, H2 and H3 and within H1 among 10- and 12-months old canes. Several other traits were surveyed that may contribute to early ripening and Pol % cane (PPC) including; percentages of flowering stalks, pithiness and millable stalks, elongation rate, internode size and differentiation. Restriction-site-associated DNA sequencing was used to genotype the mapping population and the Sorghum bicolor genome was used as reference for SNP marker calling. More than 13,000 SNP markers were scored between the mapping parents. Based on marker segregation ratio and percentage missing markers, 147 genotypes were selected for linkage map construction using 4000 single-dose markers. The marker data produced a linkage map of CP 67412 consisting of 3380 markers distributed among 239 linkage groups. The phenotypic variation (R2) of the three most prominent QTLs with a LOD score > 3 was in the range of 8.8-20.8% for the following traits: PPC, internode length and differentiation, percentage flowering and millable stalks, elongation rate, pithiness and sucrose yield. Only two QTLs were identified for earliness of ripening (harvest date and age) with LOD > 3. The lack of major QTLs (R2 > 35%) among the traits under study may be attributed to the complexity of the traits controlled by several genes and their high dependence on environmental conditions.

INTERNATIONAL EVENTS CALENDAR

2021 CONFERENCES & MEETINGS

March 1-4	**American Society of Sugar Beet Technologists (ASSBT) 2021 Meeting,Denver USA**
March 10	**APS / ISO 4th Morocco Conference – Virtual Event, London England**
April 20-23	**42nd Australian Society of Sugar Cane Technologists Conference, Bundaberg Australia**
June 14-16	**32nd ICUMSA Session, Vienna Austria**
June 14-16	**50th Annual Joint Meeting of American Society of Sugar Cane Technologists,New Orleans USA**
June 19-21	**International Conference on Sugarcane Research, Coimbatore India**
November 23-24	**30th International Sugar Organization Seminar, London England**

GUIDELINES FOR AUTHORS

Dear Fellow Author(s),

Pakistan Sugar Journal (PSJ) offers research, analysis, and reviews to keep its local and international readership up to date with latest developments in the sugar industry. PSJ takes into account the application of research and focuses on areas in agriculture related to sugar, milling and processing.

In order to have your articles published in the PSJ, you are requested to adhere to the below instructions and prerequisites to enable timely review of your submissions by the editorial board:

I. Write the title of your article in CAPITAL LETTERS in the center of the page.
II. Write the complete name of all authors with their addresses — it is compulsory in the text. References should be cited by author and years as, for one, two or more authors (Hammer, 1994, Hammer and Rouf, 1995; Hammer *et al.*, 1993), respectively.
III. Write HEADINGS in bold letters and in the center of the page.
IV. Type your article only in ARIAL format.
V. Send TABLES and FIGURES on separate page with bold title and mark its numbers correctly.
VI. Observe the following rule for REFERENCE, for one author: Hussain, K. 1991 for two authors; Khan, M. and A. Habib 1995, for more than two; Ali, K., A. Hussain and S. Nasir, 1990.
VII. Always send two soft copies and one hard copy of CD. Please do not use FLOPPYDISK for this purpose.
VIII. Send copies on an A-4 size page, preferable LASER PRINT in word document
IX. Papers published in the PSJ are free of charges (for authors).
X. Send your papers to following address by mail or email:

Dr. Shahid Afghan
Editor-in-Chief, Pakistan Sugar Journal
Shakarganj Sugar Research Institute, Jhang (Pakistan)
Phone: +92 47 111-111-765 | Ext. 602, 603
Email: shahid.afghan@shakarganj.com.pk.