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Research Article

ECO-PHYSIOLOGICAL EVALUATION OF THE BETEL VINE VARIETIES CULTIVATED IN BHOGARAI AREA OF BALASORE DISTRICT, ODISHA, INDIA FOR DISEASE MANAGEMENT AND INCREASING CROP YIELD

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ABSTRACT

Two varieties betel vine (*Piper betle* L.) locally called as *Chandrakana* Pan and *Bali* Pan have been traditionally cultivated as a cash crop in the Bhogarai area of Balasore district of the state of Odisha, India. In the present study, leaf extracts obtained from the two varieties, have been evaluated for various eco-physiological parameters such as total chlorophyll content, protein content and peroxidase activity of healthy as well as diseased leaves (foot rot, leaf spot and powdery mildew) and the results have been discussed. Total chlorophyll content, protein content and peroxidase activity of healthy leaf of *Chandrakana* Pan is 2.065 mg g^{-1}, 1.7 mg g^{-1} and 0.004 µkat g^{-1} respectively whereas the corresponding values for *Bali* Pan are 2.513 mg g^{-1}, 3.9 mg g^{-1} and 0.003 µkat g^{-1} respectively. Powdery mildew diseased leaves of *Chandrakana* Pan, showed reduction of chlorophyll content and protein to 39 % and 59 % respectively whereas *Bali* Pan showed reduction of the same parameters to 35 % and 5 % respectively. Moreover, the peroxidase activity of the diseased leaves of *Chandrakana* Pan was found to be higher than that of *Bali* Pan. Thus, under diseased condition, *Chandrakana* Pan showed relatively better results for all the three parameters, and all the three studied diseases, indicating that *Chandrakana* variety is more disease resistant in the studied agro-climatic region.

INTRODUCTION

Agro-ecosystem, defined as ecological systems modified by human beings to produce food, fibre or other agricultural products [6] is basically very valuable for the humans and their domesticated animals. The food material needed for human consumption is directly derived from the crops and waste products are mostly composted whereby the nutrients are recycled. Production of crop is the result of interaction of the genetic characters and the environmental factors that govern the crop growth. Varieties differ mostly in their genetic constitution besides physiological adaptability. Rainfall, temperature and soil constitute the most important environmental factors. Crop land or agro-ecosystem, one of the man-made ecosystems is very much dependent on external energy supply (irrigation, fertilizers etc.) on one hand and other inputs such as land preparation, weed and pest control, quality of seed or seedlings on the other [18]. Domestication of cultivated plants is an evolutionary process operating under the influence of human activities. With the origin of modern agriculture, new techniques of cultivation have been used bringing more plants into cultivation. In agricultural studies, more emphasis is given on production of economic return but in the study of plant ecology, production includes other non-economic parts such as straw, wastes and underground parts. The roots play an important role in supplying nutrients, after decomposing, to the subsequent crops in the soil. The activity of soil micro-organisms mostly influenced by the quality of underground parts left for decomposition. Productivity in crops is maintained by virtue of large energy inputs involving cultivation, irrigation, fertilizer application and genetic selection. The proper functioning of the crop fields depends mostly on the using of human labour as an important energy input [16]. Loss of salts and soil nutrients such as sodium, potassium, phosphorous etc. can be resulted due to the continuous harvest of the same crops from the same lands over the years. In order to maintain the high rate of production, the loss of nutrients should be compensated with additional supply of fertilizer.

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The green heart shaped leaves of betel vine are known as *pan* in India. Nature has a source of medicinal agents for thousands of year and modern drugs are isolated from natural sources. Leaves of *Piper betle* possess several bioactive components and are used in traditional medicinal systems. *Pan* leaves are used as a post meal mouth freshener and the crop is grown in India, Sri Lanka, Malaysia, Philippines, Thailand, Taiwan and other southeast Asian countries [21]. Medicinal plants are a source of economic value in Indian sub-continent [14]. Medicinal plants serve as the main source of medicine for the majority of the rural population in the developing countries and thus contribute significantly to the health care system of these countries [17]. Local medicinal plants are still used for primary health care needs by as much as 80% of the people in the developing countries [23]. Betel vine (*Piper betle* L.) belongs to Genus *Piper* of the family Piperaceae. It is a perennial dioecious plant. Bhogarai area of Balasore district this plant is cultivated as a cash crop. The leaves of pan are the economically important part. There are about 100 varieties of betel vine in the world, of which about 40 are found in India, 30 in West Bengal and two varieties are cultivated in Bhogarai. The most probable place of origin of betel vine is Malaysia [4]. Betel vine is an important, traditional and an ancient crop of India. Currently more than 200 cultivars are cultivated in several states of India for its leaves, which are used mainly for chewing purposes. The farmers and consumers name the cultivars after their localities, village or towns. However, the cultivers with prefix *Desi* in their names invariably refer to the cultivar *Bangla* in West Bengal, cultivar *Kapoori* in Maharashtra, cultivar *Desavari* in Madhya Pradesh [3], cultivar *Bali* and *Chandrakana* in Bhogarai. The traditional nomenclature is thus confusing.

Importance of the study

Betel leaves are the most important and valued plant part, used as a chewing agent and are of religious, medicinal and ceremonial value in India. There are three important crops that the farmers of the Bhogarai area cultivate. These are *Dhana* (rice), *Pana* (betel vine) and *Mina* (fish). Among these three, the betel vine cultivation has been widely adopted by the farmers of this locality due to the following reasons. i.e. good economic return to sustain the family. Easy availability of all materials required for preparation of make shift farm house for cultivation of betel vine, locally known as *"Pana Baraja"*. Availability of manual labourers required for different field practices, harvesting, transporting and packing of pan leaves throughout the year. Being engaged in different types of works related to betel vine cultivation, the land less and marginal farmers get avenues to sustain their livelihood. The soil and other agro-climatic conditions of the locality are very conclusive for betel vine cultivation. Increasing demand of the market for betel vine leaf, both at national and international levels has sustained the cultivation in this region. Because of all these reasons, the betel vine cultivation has got popularity in the area. But now a days, the farmers of this locality are facing many problems due to the decrease in the market demand of the betel leaf due to popularity of packaged mouth fresheners like " *Gutkha*" both in the local and national markets. The consequence of this is the sharp decline in the commercial value of pan leaf as a result of which the betel vine cultivation is not now economically sustainable. Increase in the price of different raw materials required for the preparation of "*Pana Baraja*". Many times, the farmers are facing problems to manage the diseases due to lack of proper training to save their crop. The unemployed youths of the rural area are now mostly attracted towards different cities; as a result for different practices of the field, there is a case shortage of manual labourers. During summer period, most of the feeding ponds of the "*Pana Baraja*" dry up as a result there is acute shortage of water to meet the crop demand. The "*Pana Baraja*" of the area have been heavily damaged during the recent tropical cyclones like *Philine* and *Hud Hud*. Because of all these reasons, this traditional cultivation which serves as the economic back-bone of Bhogarai area is now at stake. For effective and sustainable improvement of this age-old cultivation, through socio-economic and eco-physiological studies related to betel vine cultivation of Bhogarai area are highly essential. Since these aspects of analysis of this area have not been done till date, initiatives have been taken in this study for documenting eco-physiological aspects of betel vine cultivation of Bhogarai area of Balasore district, Odisha, India. Thus the objective is to study physiological parameters like total chlorophyll content, protein content and Peroxidase activity of healthy leaf tissues and three types of diseased leaf tissues of two different varieties of *Pan* cultivated in this area for a perspective insight into the disease management and increasing crop yield.

Study area and climate

Balasore is one of the coastal districts of the state of Odisha, India. It lies on the northern most part of the state. The district is located at 20°.48' to 21°.59' North latitude and 86°.16' to 87°.29' East longitude. The average altitude of this district is 19.08 metre. The district covers an area of 3634 sq. km. Balasore district can be divided in to three geographical regions, namely coastal belt, the inner alluvial plain and North-Western hills. The coastal belt is about 81 km. This region is mostly flooded with brackish water of estuarine rivers which is unsuitable for cultivation. Presently this area is utilized for coconut and betel cultivation. Prawn culture and salt manufacturing units are also developing in this area recently. The local economy of Balasore district largely depends on the cultivation of paddy, fish and betel vine. Two important rivers of Odisha, namely Subarnarekha and Budhabalanga pass through this district from west to east before surging in to the Bay of Bengal. The soil of central region is mostly clay, clay loam and sandy loam which is fertile for paddy and betel vine cultivation. The field study was conducted in the Bhogarai area which is situated in the northern most part of Balasore district. Bhogarai revenue area is one of the most economically developed areas of Balasore district. This area is located at 21°.65' latitude and 87°.36' longitude. And elevation is 13m (altitude).The area is famous for production of pan, coconut and betel nut (Figure 1). The coastal Odisha experiences maritime climate and receives the south-west monsoon rainfall. The temperature of the coastal area is moderate. The climate of the Balasore district is characterized by hot summer and high humidity during rainy season, dry winter and low diurnal range of temperature throughout the year. Annual normal rainfall of the district is 1591 mm, which is almost 9% higher than normal rainfall of Odisha. This is due to tropical cyclones, which cause

major precipitation in the area. Cyclones are frequent in the district because of the close proximity to the Bay of Bengal. The district is generally affected by cyclones, which are caused when depressions originate in the Bay of Bengal over Andaman and Nicobar Islands and move towards east coast of India. More than 70.9% of rainfall is received between June to September through South-West monsoon. The study area generally experiences three distinct seasons viz., rainy (July–October), winter (November–February) and summer (March–June). Although the Bay of Bengal is very near to the study area (about 15 km), the maximum and minimum temperatures recorded at Balasore Metrological Station during 2014-2015, were 36.7°C in the month of May 2015 and 14.2°C recorded in the month of January 2015. In summer season the minimum daily temperature ranged from 23.3°C to 27.1°C, while in rainy season (July-October) minimum ranged from 24.1 to 25.5°C and in winter it ranged from 14.2 to 18.9°C. During the summer season the daily maximum temperature ranged from 32.8 ^{0}C to 36.7 ^{0}C. In winter the maximum temperature ranged from 24.9^{0}C to 29.3^{0}C, while in rainy season it varied from 30.1 to 31.7^{0}C. The rainy season starts in the experimental area during the middle of June. The average annual rainfall of the area is 675.6 mm. During the year 2014-15 the highest rainfall was recorded in the month of June 2015 i.e. 228.9 mm and lowest in the month of October (2.0 mm). Due to climate change very lowest rainfall was recorded in rainy season. It was observed that soil property in Bhogarai revenue area varies at different place. Two types of soil are seen in Bhogarai. i.e. sandy-loam and clay-loam. It was noticed that the production of *Piper betle* L. grows well in sandy-loam soil as compared to clay-loam soil.

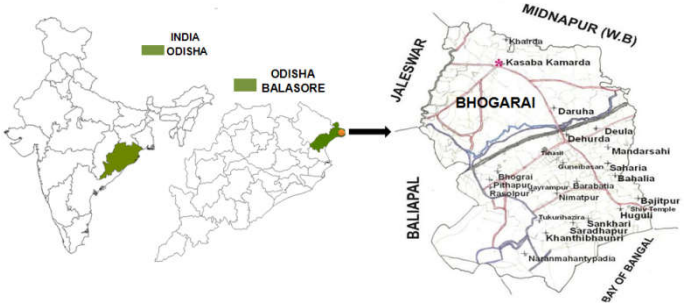

Figure 1 Map showing the study site of Bhogarai area of Balasore district, Odisha.

MATERIAL AND METHODS

Field observations and collection of plant materials

For the present study, different places of Bhogarai revenue area of Balasore district of Odisha, under study, were visited in different seasons during 2014-2015 to collect information about pan (*Piper betle* L.). The specimens i.e. leaves of pan were collected from the field of Bhogarai areas. During the collection of the specimens morphological forms along with ecological notes were documented. The diseases of the plant species were identified and reported, on the basis of plant specimen collected, personal observation and interviews (group, personal, electronic and telephonic) made during the study period. The rural areas were visited and discussion with farmers and labours was made and they were interviewed to collect information on cultivation, harvesting, processing, disease management, marketing, problems and current uses of the species. A questionnaire was developed to collect the required information.

Fresh pan leaves collected were brought to laboratory in a plastic bag. The specimens were identified, described and illustrated with the help of Flora Book "The Botany of Bihar and Odisha" [9] and other Indian Flora "The Flora of British India" [11] and research articles [1, 7, 8]. The plants were enumerated with their correct scientific name, family name, local Odia name and English name along with the botanical descriptions of the species.

Soil Carbon Analysis

Sandy-loam and clay-loam soils were collected separately from two different sites. And organic carbon was estimated by Walkey- Black rapid titration method (1934) [22]. 500 mg of soil sample (passed through sieve) were taken in a 500 ml conical flask. 10 ml of standard potassium dichromate solution was added to the soil sample and swirled, so that the dichromate solution completely wet the soil. 20 ml of concentrated sulfuric acid was added with intermittent swirling of the conical flask, after which the flask was kept for half an hour. Then the contents were diluted with 200 ml distilled water and 10 ml of ortho-phosphoric acid, half spoon of solid sodium fluoride powder and 1 ml of diphenyl-amine indicator were added. The contents were titrated with ferrous ammonium sulphate solution till the colour of the solution changed from dark blue to dark green. With each batch run, one blank was processed to which no soil was added. The total organic carbon of the soil was calculated as described by Walkey- Black (1934).

$$C\,\% = \frac{0.003\times(B-T)}{B/10}\times\frac{100}{W} = \frac{3(B-T)}{BW}$$

Where W = Weight of the soil taken in grams, B = Titrate value for blank, T= Titrate value with soil

Soil pH

20 g of soil sample was taken in 100 ml of beaker and 40 ml of distilled water was added to it with continuous stirring with a glass rod for about 30 minutes. The pH meter was standardized and the glass electrode was immersed in the soil-water suspension and the pH was read. Interpretation of pH are extremely acidic <4.5, very strongly acidic 4.6-5.0, strongly acidic 5.1-5.5, medium acidic 5.6-6.0, slightly acidic 6.1-6.5, neutral 6.6-7.3, mildly alkaline 7.4-8.0, strongly alkaline 8.1-9.0, very strongly alkaline >9.0.

Physiological Analysis

Photosynthetic pigment content

The chlorophylls occur in the chloroplasts as green pigments in all photosynthetic plant tissues and they are the essential components for photosynthesis. They are bound loosely to proteins and are readily extracted in organic solvents such as acetone or ether. There are five types of chlorophylls (Chl) in plants. Chlorophyll a (Chl a) and chlorophyll b (Chl b) occur in higher plants, ferns and mosses. Chlorophylls c, d and e are only found in algae and in certain bacteria. So, total chlorophyll contents were determined by following (Arnon 1949) [2, 10, 20]. For extraction and estimation of total chlorophyll, 100 mg leaf tissues were homogenized with 80 % acetone in clean

mortar and pestle and the volume was adjusted to 5 ml. The extracts were then centrifuged for 10 minutes at 5000 rpm and the supernatants were collected separately. After that the absorbance of the supernatant was taken at 645 nm (A_{645}) and 663 nm (A_{663}) in a spectrophotometer using 80 % acetone as the reference. The total chlorophyll content of the supernatant was calculated as described by Arnon (1949). The total chlorophyll content was calculated using the formula:

µg total chlorophyll / ml of extract = $8.02 \times A_{663} + 20.21 \times A_{645}$
Where A_{663} and A_{645} are the absorbances at 663 nm and 645 nm respectively.

Protein content

Lowry *et al.* (1951) method was followed to determine the protein content using Bovine Serum Albumin (BSA) solution as standard for estimation of protein content of test samples [15]. Briefly, 200 mg of leaf tissues were taken for extraction of proteins. The leaf tissues were homogenized with cold 50 mM sodium phosphate buffer (pH 7.5) in a glass mortar and pestle, kept in an ice bath and the volume of the homogenate was adjusted to 5 ml using the buffer. The homogenate were then centrifuged at 7000 rpm for 10 min at -4^{0}C. Equal volume of 20% (w/v) TCA was added to the supernatant and these were kept in a refrigerator for overnight to facilitate complete participation of soluble protein. The samples were then centrifuged for 15 min at 4000 rpm. The pellets were washed successively with 10% cold TCA (twice), ethyl alcohol (once), ethyl alcohol: chloroform (3:1, v/v, once), ethyl alcohol: ether (3:1, v/v, once) and finally with ether (once). The pellets were evaporated to dryness and were solubilized by re-suspending in 2 ml of 0.3 N NaOH for 16 hr at 37 °C. Finally the samples were centrifuged and supernatants were collected for protein estimation, after suitable dilution. The absorbance of the developed colour was read at 750 nm and the protein content was calculated by comparing with the standard curve calibrated with 0-200 µg of BSA (Sigma, fraction v) per assay.

Peroxidase activity

Peroxidase occurs in plants and in certain animal cells. It catalyzed the oxidation of many organic compounds by hydrogen peroxide (amines, phenols and hydroquinones etc.). The peroxidase activity was assayed by taking guaiacol as the reduced co-substrate, following the method developed by Kar and Feierabend (1984) [12]. For extraction and estimation of peroxidase activity, 200 mg leaf tissues were taken separately in clean mortar and the extraction process was carried out in an ice bath. The leaf tissues were homogenized with sodium phosphate buffer (pH 7.5) which was known as extraction buffer, in a mortar and pestle and the volume was adjusted to 5 ml. The homogenates were centrifuged in a cooling centrifuge machine at 10,000 rpm for 10 min at -4°C. The supernatants were collected as enzyme extract and were used for enzyme assay after suitable dilution. The assay mixture was composed of 0.5 ml of 30 mM guaiacol, 0.5 ml of 30 mM hydrogen peroxide, 1.5 ml of 0.1 M sodium phosphate buffer (pH 6.8) and 0.5 ml of enzyme supernatant. As soon as the enzyme was added to the assay mixture the reaction started immediately which was visually noticed by change in colour of the reaction mixture. This was due to formation of tetra-guaiacol in the reaction mixture. The rate of change in colour of the reaction mixture was immediately recorded in spectrophotometer at 470 nm. Peroxidase activity was calculated using the extinction co-efficient of 26.6 $mM^{-1}cm^{-1}$ for tetraguaiacol at 470 nm and was expressed *Katals* i.e. moles of tetraguaiacol formed per second due to peroxidase activity under assay condition.

RESULTS AND DISCUSSION

The pan species

The *Pana* species belongs to the family Piperaceae. The species is characterized as a dioecious, climbing shrub with woody root, stem stout with pinkish-strip along, node dilated. Petiole 2-2.5 cm long, leaf blade fleshy coriaceous, glabrous, greenish or yellowish, broadly ovate, 7-8.5 cm wide, 9-11 cm long, apex acuminate, base cordate. Veins 7-9, elevating beneath, 2 pairs basal, one pair arising from midrib. Spike cylindric, pendulous. peduncle 2-3 cm long. Bract orbicular. Stigmas 4-6, pubescent. Fruiting spike 3-5 cm long. Drupe embedded on rachis. Flowering and fruiting on year round [9, 5,1]. The two varieties of *Pana* cultivated in the study area are characterized as follows.

Piper betle L. var. Jaleswar (Family- Piperaceae)
In local vernacular it is known differently as *Bali Pana*, *Desi Pana*, *Mitha Pana*, *Birkuli Pana*. The *Bali Pana* leaves are long, thin; smell good, juicy and sweet in taste.
Piper betle L. var. Bangladeshi (Family- Piperaceae)

In local vernacular it is known differently as *Chandrakana Pana*, *Matiali Pana*, *Athila Pana*, *Raga Pana*. The *Chandrakana Pana* leaves are rounded and big, thick; smell good and pungent in taste. The most distinguishing feature of the *Chandrakana Pana* is the left and right lobes of leaf margin near the petiole, which circumscribe the petiole and tend to touch each other.

Soil organic carbon and pH

Table 1 Soil organic carbon and pH of two prevalent types of soil of Bhogarai area.

Types of soil	Surface soil		Depth soil (30 cm)	
	Organic carbon %	pH	Organic carbon %	pH
Sandy loam	2.80	6.13	1.67	5.82
Clay loam	2.00	6.76	1.28	6.31

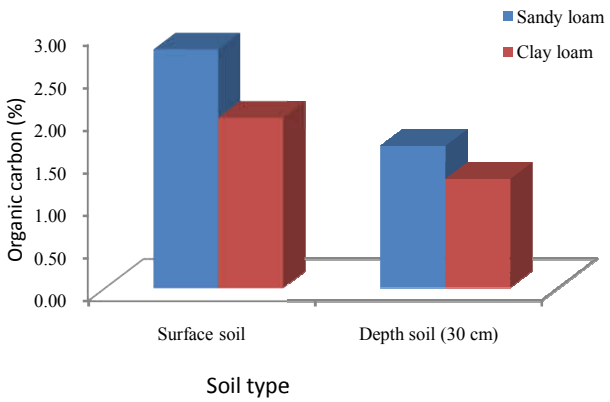

Figure 2 Soil organic carbon (%) available in two different types of soil of Bhogarai.

The soil organic carbon content and the pH of different soil samples found in different Pan cultivated fields of Bhogarai are

shown in Table 1. As shown in Figure 2, organic carbon percentage of sandy loam soil is higher than clay loam soil in both surface soil and 30 cm depth soil. Soil organic levels of both the type of soil found in the study area were higher than 2.0%, indicating that there are sufficient soil organic matter to maintain most soil functions.

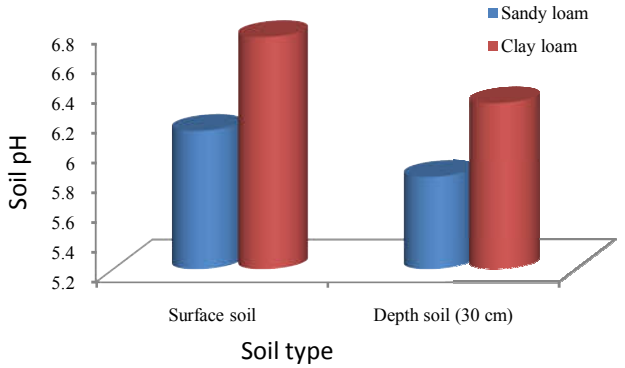

Figure 3 Soil pH available in two different types of soil of Bhogarai.

Figure 3, shows that pH of surface soil of sandy loam has slightly acidic where as clay loam has neutral and 30 cm depth soil pH has medium acidic in sandy loam soil where as in clay loam soil it has slightly acidic. pH varies in agricultural land because of load of manure in the soil. Though the soil of the study area is slightly acidic in nature, sufficient soil organic matter present in these soils can increase the soil buffering capacity. Even if the acidification pressure remains on the soil due to the existing agricultural practice, the healthy amount of soil organic matter will delay the process of soil acidification.

Physiological analysis

The results of the different physiological parameters including total chlorophyll content, protein content and peroxidase activity in the healthy and diseased leaves of *Pana* are shown in Table 2 and the results have been discussed under following headings.

Table 2 Total chlorophyll content, Protein content and Peroxidase activity of healthy and diseased *Pana* leaf.

Types of leaf	Total chlorophyll content (mg g^{-1} FW)		Protein content (mg g^{-1})		Peroxidase activity (µkatal g^{-1} FW)	
	Bali Pana	*Chandrakana Pana*	*Bali Pana*	*Chandrakana Pana*	*Bali Pana*	*Chandrakana Pana*
Healthy	2.513	2.065	3.9	1.7	0.003	0.004
Foot rot	1.410	1.373	1.2	1.2	0.008	0.005
Leaf spot	1.698	1.736	0.6	1	0.007	0.009
Powdery mildew	0.876	0.807	0.2	1	0.014	0.017

Photosynthetic pigment content (Total Chlorophyll)

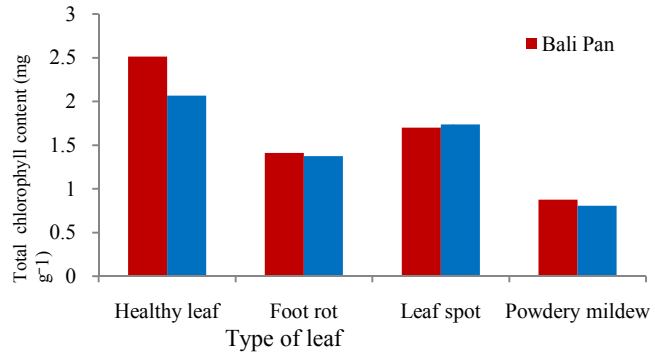

Figure 4 Total chlorophyll content of healthy and diseased leaf.

Table 3 Total chlorophyll content percentage in diseased leaves in comparison to the healthy leaves.

Variety	Healthy leaf (%)	Diseased leaf (%)		
		Foot rot	Leaf spot	Powdery mildew
Bali Pana	100	56	68	35
Chandrakana Pana	100	66	84	39

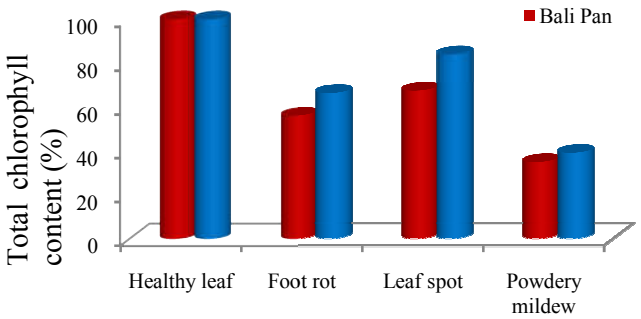

Figure 5 Total chlorophyll content percentage of diseased leaves in comparison to healthy leaves.

The results indicate that the healthy leaves of *Bali Pana* has higher chlorophyll content (Figure 4), however, from Figure 5, it can be seen that the *Chandrakana* variety is more resistant against all the three studied diseases. In *Bali pan*, the chlorophyll content of diseased leaves has been reduced to 35 % for Powdery mildew disease whereas in *Chandrakana* variety the corresponding decrease is 39 % (Table 3). Total chlorophyll content is an index of overall health and productivity of plant [19]. Chlorophyll content of plants varies from species to species; age of leaf and also with the pollution level as well as with other biotic and abiotic condition [13].

Protein content

Table 4 Protein content percentage in diseased leaves in comparison to the healthy leaves.

Variety	Healthy leaf (%)	Diseased leaf (%)		
		Foot rot	Leaf spot	Powdery mildew
Bali Pana	100	31	15	5
Chandrakana Pana	100	71	59	59

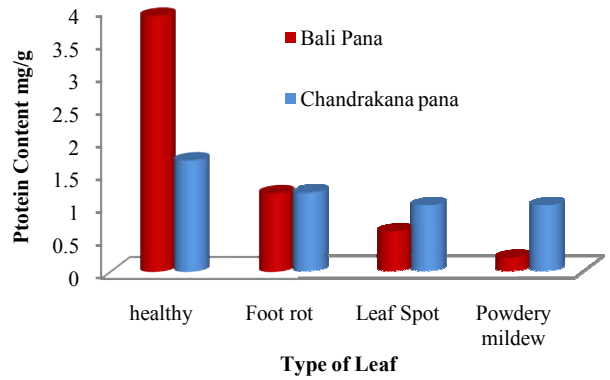

Figure 6 Protein content different leaves of two different variety of pan collected from Bhogarai.

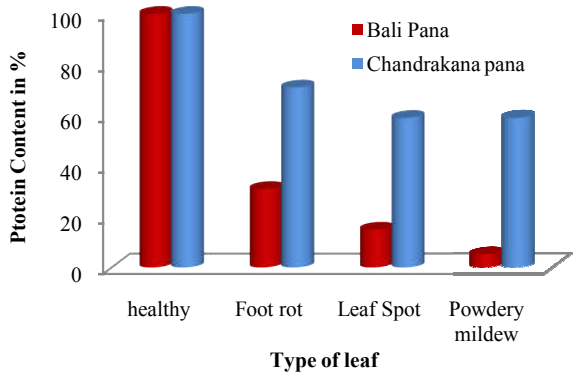

Figure 7 Protein content percentage of diseased leaves in comparison to healthy leaves.

Results of the study show that the healthy leaves of Bali pan have high protein content. i.e. 3.9 mg g^{-1} whereas the healthy leaves of *Chandrakana Pana* have low protein content i.e. 1.7 mg g^{-1} (Figure 6). However, in terms of disease resistance, *Chandrakana* variety shows better results (Figure 7). Protein content of the Powdery mildew affected leaves of *Bali* pan variety has reduced to 5 % of that of healthy leaves whereas, the corresponding value for *Chandrakana* variety is 59 % (Table 4). Proteins play a key role in different physiological processes of plant. Under different conditions of abiotic and biotic stress protein content of the plant tissue varies. Under stress conditions, different metabolic pathways of the plant stops and simultaneously different stress related proteins are expressed, which help the plant to survive in the stress period. Thus the portion content of a plant tissue can serve as an indicator of stress resistance.

Peroxidase activity

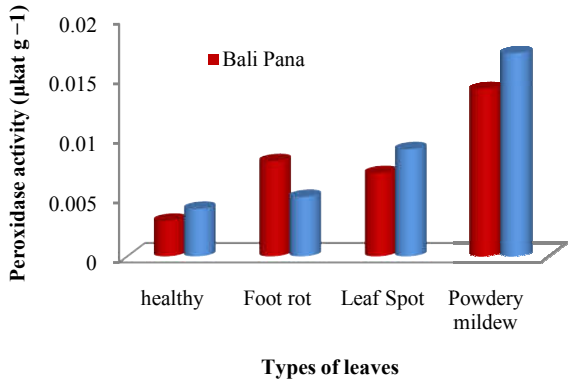

Figure 8 Peroxidase activity of healthy leaf tissue and diseased leaf tissue of *Bali Pana* and *Chandrakana Pana*.

Table 5 Peroxidase activity percentage in comparison to the diseased leaf.

Variety	Healthy leaf (%)	Diseased leaf (%)		
		Foot rot	Leaf spot	Powdery mildew
Bali Pana	100	125	225	425
Chandrakana Pana	100	267	233	467

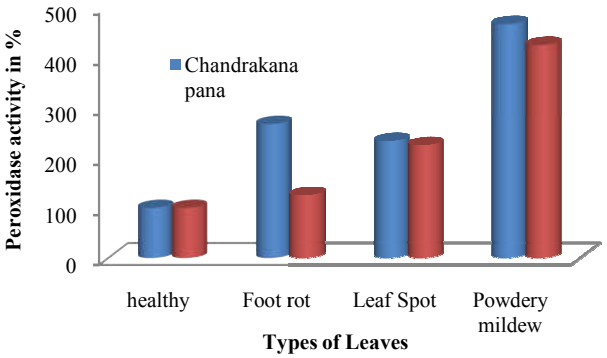

Figure 9 Peroxidase activity percentage in comparison to the diseased leaf.

As shown in Figure 8 and Figure 9 peroxidase activity in both healthy and diseased leaves of *Chandrakana* variety is more than that of the *Bali* pan variety. The leaf extracts of powdery mildew diseased plant show highest peroxidase activity with reference to other two studied diseases (Table 5). Peroxidases are known to play a role in increasing a plant's defenses against pathogens. Peroxidases are involved in many physiological processes in plants, including responses to biotic and abiotic stresses. Thus higher amount of peroxidase activity in the tissue extract indicate increased disease resistance.

In spite of tremendous economic potentiality of the betel crop, it remains neglected particularly by the scientists, technologists, administrators and policy makers as well. Consequently, statistical data of betel leaf is still scattered and messy while its agronomy remains to be a matter of personal experience gained through traditional farming practiced generation after generation. Awareness of plant protection practices to control pests and diseases are required to achieve good yield. There are needs to select new planting techniques with modern technology and to select disease resistant varieties. The betel vine cultivation has very much affected by diseases and outcome of the farmers were, big loss for betel vine cultivation. The farmers have not been able to identify the disease at an early stage to initiate preventive action due to the non availability of modern technology. Present study has made a pioneering step in identification of a disease resistant variety of pan for the local agro-climatic condition using a unique combination of eco-physiological analysis. This will not only help the farmers to have a better yield but also it will allow further research to develop disease resistant varieties of pan, an economic crop of the coastal regions of India and other South Asian countries in general and Bhogarai area in particular.

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