



**SHRI SHIVAJI EDUCATION SOCIETY, AMRAVATI'S**  
**SHRI SHIVAJI COLLEGE OF ARTS, COMMERCE AND SCIENCE, AKOLA (MS)**  
**Affiliated with Sant Gadge Baba Amravati University, Amravati (MS)**  
**UGC Status- College with Potential for Excellence (Phase II Completed)**  
**DST- FIST (Level "00") Support;**  
**Lead College Status by S. G. B. Amravati University, Amravati (MS)**  
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3.3.3

3.3.3 Number of books and chapters in edited volumes/books published and papers published in national/international conference proceedings per teacher during last five years (10)

Academic Year: 2018-19

Shri Shivaji Education Society, Amravati's

## SHRI SHIVAJI COLLEGE OF ARTS, COMMERCE AND SCIENCE, AKOLA



NAAC Re-Accredited with A grade with CGPA 3.24  
UGC Status of 'College with Potential for Excellence', DST-FIST level-0 Support

**Lead College status by S.G.B.A.U. Amravati**

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No. SSC/AKL/

Date 15-12-2021

### Declaration

This is to declare that the information, reports, true copies and numerical data etc. furnished in this file as supporting documents is verified by IQAC and found correct.

Hence this certificate.

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**3.3.3 Number of books and chapters in edited volumes/books published and papers published in national/ international conference proceedings per teacher during last five years (10)**

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# PROCEEDINGS



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**S.S.S.K.R.Innani Mahavidyalaya**

**Karanja (Lad), Dist. Washim (M.S.)**

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*In collaboration with*

**Department of Mathematics & IQAC**

**Sant Gadge Baba Amravati University, Amravati**

**Vidya Bharati Mahavidyalaya, Amravati**

**&**

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**ABSTRACTS SECTION**

## ASSESSMENT OF *IN VITRO* ANTACID ACTIVITY OF *FOENICULUM VULGARE*, *CORIANDRUM SATIVUM* AND *LEPTOSPERMUM SCOPARIUM* BY A MODIFIED ARTIFICIAL STOMACH MODEL

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### ABSTRACT

Some plants and fruits may consist of pharmaceutical ingredients suitable to treat certain diseases such as stomach acidity. Our aim was to verify the claim that extract of *Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum scoparium* can be used as an alternative for neutralizing stomach acidity. Therefore, for this purpose, the antacid activity of these aqueous extracts was evaluated by using artificial stomach model. This modified model of artificial stomach is useful for the determination of duration of consistent neutralization of the artificial gastric acid. The pH of these aqueous extracts and its neutralizing effect on artificial gastric acid was determined and compared with water and the active control sodium bicarbonate. The neutralization capacity *in vitro* was determined with the classical titration method. All treatments including that of aqueous extracts and sodium bicarbonate showed significant acid neutralizing effects when compared with water. The duration for consistent neutralization and antacid capacities of extract of *F. vulgare* ( $172 \pm 6$  min), *C. sativum* ( $154 \pm 5$  min) and *L. scoparium* ( $118 \pm 3.5$  min) were significantly higher than that of water ( $108 \pm 4$  min). The aqueous extract of *F. vulgare*, *C. sativum* and *L. scoparium* was consistently active in the artificial stomach model and possesses potent antacid effects.

**Key Words :** *Foeniculum vulgare*, *Coriandrum sativum*, *Leptospermum scoparium*, artificial stomach model, antacid activity.

### INTRODUCTION

A variety of herbal plants have been used worldwide to remedy various diseases. The healing aspects of herbal plants have been accepted in many cultures and have been known as alternative medicine. Some herbs may consist of pharmaceutical ingredients suitable to treat certain cases such as stomach acidity or ulcers (1).

*Foeniculum vulgare* also called fennel and commonly known as badi sop, is a flowering plantspecies in the carrot family. It is a hardy, perennial herb with yellow flowers and feathery leaves. Fennel seeds benefits in helps to regulate blood pressure, reduce water retention. According to ayurveda fennel seeds reduce all three Trodosha (Vata, Pita, and Kapha). The seeds have a cooling effect on the body. It is a good idea to consume a fennel seed drink during the scorching summer, to relieve heat from body. The oil found in the seeds is carminative in nature, hence it is used in massage blends, especially in Ayurveda to calm the nerves and promote mental clarity.

*Coriandrum sativum* also known as cilantro and commonly called as Dhaniya, is an annual herbs in the family Apiaceae. All parts of the plant are edible, but the fresh leaves and the dried seeds are the parts most traditionally used in cooking. In Ayurveda, coriander often recommended for

stomach related ailments, as it is known to ease digestion. It is indeed super seed and its regular use can bring about various health benefits.

*Leptospermum scoparium*, commonly called manuka, manuka myrtle, is a species of flowering plant in the myrtle family Myrtaceae. Some of the top manuka uses are and benefits include helps with SIBO, low stomach acid, acid reflux, treat burns, wound and ulcers, help allergies, improves sleep.

Hyperacidity is a condition that occurs in many diseases caused by an excessive formation of acid in the stomach associated with a burning sensation in the throat and heart area. This condition appears in many diseases ranging from gastritis and peptic ulcers to the currently named gastro-esophageal reflux disease (GERD) (2).

Acidity is a common gastrointestinal disease which may not be necessarily caused by a pathogenic infection. It is attributed to a functional disorder that can result due to a variety of reasons which is related to heartburn and gas formation in stomach. In acidity, gastro esophageal reflux disease which is commonly known as urdhva gata amalpitta in Ayurveda, there is a movement of gastric juices from the stomach into the lower esophagus [3]. This is a condition which occurs when acidic contents in stomach move upward

into the esophagus and make it dysfunctional. Gastric acid is a digestive fluid formed in the stomach having a pH of 1 to 2. It is a mixture of hydrochloric acid, large quantities of potassium chloride and sodium chloride. The acid in stomach plays a significant role in the digestion of proteins, by activating digestive enzymes and making ingested proteins unravel so that digestive enzymes can break down the long chain amino acids. General symptoms found in children are respiratory problems, inadequate weight, vomiting, coughing and turning down food. The symptoms shown by adults include long heartburn, chest and stomach pain, gas formation in stomach, inflammation in chest; gastro esophageal reflux, voice change and formation of ulcer in esophagus, pain during muscular contractions and pain in ears are some of the symptoms of acidity (4-7). Although there are a number of antacids and anti ulcer drugs, most of these have limitations, side effects and drug interactions. Herbal medicines generally deal with plants and its extracts for treatment of various ailments. These are usually considered to be safer and with no side effects (8). Peptic ulcer or Peptic ulcer disease (PUD) is defined as a break in the mucosal lining of the gastrointestinal tract (9). It occurs in that part of the gastrointestinal tract which is exposed to gastric acid and pepsin, i.e. the stomach and duodenum (10). The incidence of PUD varies with age, gender, geographical location and is associated with severe complications including hemorrhages, perforations, gastrointestinal obstruction, and malignancy. Thus, this clinical condition represents a worldwide health problem because of its high morbidity, mortality and economic loss (11). Antacids relieve the effects of the ulcer by neutralizing small amounts of excess stomach acid.

The experimentation was carried out by using a artificial modified artificial stomach model. Using this method we have screened the neutralization effect, duration of neutralization and neutralization by simple titration method.

## **MATERIALS AND METHODS**

### **Collection, authentication and extraction of fruit rinds**

The sources *Foeniculum vulgare*, *Coriandrum sativum*, *Leptospermum scoparium* was collected from the local shops of the Akola district region of Maharashtra, India and air dried under shade, powdered mechanically and stored in air tight

containers. The extract was prepared by keeping powder for incubation in distilled water for 24 hrs (20 gm powder in 80 ml distilled water) and water as a solvent and stored in a refrigerator for further use.

### **Chemicals and reagents**

Pepsin, sodium chloride, hydrochloric acid and sodium bicarbonate were obtained from Shri. Shivaji College of Arts Commerce and Science, Akola (MS), Maharashtra, India. All other chemicals were obtained from local sources and were of analytical grade.

### **Preparation of artificial gastric acid**

Two grams of NaCl and 3.2 mg of pepsin were dissolved in 500 ml distilled water. Hydrochloric acid (7.0 ml) and adequate water were added to make a 1000 ml solution. The pH of the solution was adjusted to 1.20.

### **pH determination of the extracts**

The pH of all three extracts was determined at temperatures ranging from 25°C to 37°C by pH meter (Spectronics, India). The pH values of the active control solution sodium bicarbonate (SB) and water was also determined for comparison.

### **Determination of the neutralizing effects on artificial gastric acid**

The freshly prepared test solutions of all three extracts (90 ml), water (90 ml) and the active control SB (90 ml) were added separately to the artificial gastric juice (100 ml) at pH 1.2. The pH values were determined to examine the neutralizing effects on artificial gastric juice. (Six experiments were performed for each solution.

### **Determination of the duration of consistent neutralization on artificial gastric acid using the modified artificial stomach**

The apparatus of the modified artificial stomach was made up of three elements: a pH recording system (R), a stomach (S) and a peristaltic pump (P). Each freshly prepared test sample (90 ml) was added to 100 ml of artificial gastric juice at pH 1.2 in the container of the artificial stomach at 37°C and continuously stirred (30 rpm) with a 2.5-cm magnetic stirring apparatus. Artificial gastric juice at pH 1.2 was pumped at 3 ml/min into the container of the artificial stomach, and pumped out at 3 ml/min at the same time. A pH meter was connected to continuously monitor the changes of pH in the container of the artificial stomach. The duration of the neutralization effect was determined when the pH value returned to its initial value (pH 1.2). Six experiments were



performed for each freshly prepared test solution, water and standard (SB).

**Determination of the neutralization capacity in vitro using simple titration method**

Each freshly prepared test sample (90 ml) was placed in a 250 ml beaker and warmed to 37°C. A magnetic stirrer (REMI, India) was continuously run at 30 rpm to imitate the stomach movements. The test samples were titrated with artificial gastric juice to the end point of pH 3.

**RESULTS AND DISCUSSION**

**The pH values of the test solutions at temperatures ranging from 25°C to 37°C**

The pH values of the test solutions at temperatures ranging from 25°C to 37°C. The pH values of the extracts of *Foeniculum vulgare*, *Coriandrum sativum*, and *Leptospermum scoparium* solutions at temperatures from 25°C to 37°C ranged from 4.02 to 3.92, 3.58 to 3.51 and 2.98 to 2.93, respectively. The pH values of water and SB solutions at temperatures from 25°C to 37°C ranged from 6.94 to 7.24 and 9.13 to 9.07, respectively. The results indicate that temperature did not affect pH significantly.

**Neutralizing effects on artificial gastric acids**

When 90 ml of the test solution was added to 100 ml of the artificial gastric juice (pH 1.2), the pH values of extracts of *Foeniculum vulgare*, *Coriandrum sativum*, and *Leptospermum scoparium* solutions were found to be 3.08 ± 0.04, 2.52 ± 0.08 and 2.08 ± 0.05, respectively. The pH values of water and SB solutions were 1.52 ± 0.02 and 2.11 ± 0.14, respectively. This result shows that the neutralizing effect of all three extracts was significantly better than that of water (Table 1).

**Table 1:** pH values with 90 ml water, standard and all three extracts added to 100 ml of artificial gastric juice

SN	Samples	pH values
1	Water	1.52 ± 0.02
2	Standard	2.11 ± 0.14
3	<i>Foeniculum vulgare</i>	3.08 ± 0.04
4	<i>Coriandrum sativum</i>	2.52 ± 0.08
5	<i>Leptospermum scoparium</i>	2.08 ± 0.05

**Duration of consistent neutralization effect on artificial gastric acids**

The durations for consistent neutralizing effects of extracts of *Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum scoparium* solutions

were 172 ± 6, 154 ± 5 and 118 ± 3, respectively. Those of water and SB solutions were 108 ± 4 and 180 ± 7, respectively. The duration of antacid action of SB was the longest, followed by the *Foeniculum vulgare*, *Coriandrum sativum*, *Leptospermum scoparium* which were significantly higher than that for water (Table 2).

**Table 2:** Duration of antacid effect for consistent neutralization of gastric acid

SN	Sample	Duration
1	Water	108 ± 4
2	Standard	180 ± 7
3	<i>Foeniculum vulgare</i>	172 ± 6
4	<i>Coriandrum sativum</i>	154 ± 5
5	<i>Leptospermum scoparium</i>	118 ± 3

**Physical neutralization capacity in vitro**

The active control SB and all three extracts, exhibited significant antacid potency. The neutralization capacities of extracts of *Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum scoparium* were significant as of standard.

*Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum scoparium* juice is traditionally used for the treatment of acidity. Therefore, the present study used the simple titration method and the modified artificial stomach model, which mimic the regular physiological functioning of a human stomach, to explore the antacid effects of this three extracts. The model of the 'artificial stomach' reproduces two major gastric dynamic functions. It allows interplay between one function (F-IN) corresponding to acid secretion and the other (F-OUT) corresponding to gastric emptying. This model is used to mimic gastric secretion and emptying in physiological situations. A 3 ml/min rate of 0.1 N HCl introduction corresponds to an hourly acid output of 18 mmol whilst arbitrarily chosen emptying rates represent slow, equal to input and fast emptyings. These parallel conditions are encountered in response to liquid meals in normal subjects. The lag-times before reaching selected pH values after introduction of the test solutions have been converted into antacid capacities (mmol H<sup>+</sup>), taking into account the amount of acid present in the 'stomach' at time 0 and the amount of acid introduced thereafter (12). Efficacious, intensive antacid therapy is often unacceptable because of the common side effects, especially altered bowel functions. Aluminum salts may cause constipation and magnesium salts cause diarrhoea. Sodium bicarbonate (SB) should be

avoided even though it is a potent neutralizer of acid because it contains significant amounts of sodium and may alter the systemic pH. In addition, antacid-drug interactions have been frequently reported for SB (13). The most clinically significant interactions occur with ferrous sulfate, tetracycline and quinolone antibiotics. Other interactions are potentially significant because they involve drugs with narrow therapeutic ranges. Considering the side effects and interactions of antacids, the natural and edible products such as *Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum Scoparium* should be looked to as an alternative for the treatment of PUD.

### CONCLUSION

In conclusion, this study aims the scientific validation of the traditional antacid claims of

*Foeniculum vulgare*, *Coriandrum sativum* and *Leptospermum Scoparium* and to promote its use in the form of a soft drink and as a culinary spice in people's diets. Besides, it is cheap, readily available to all strata of society, with medicinal properties attributed to it.

### ACKNOWLEDGEMENT

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### CONFLICT OF INTEREST STATEMENT

We declare that we have no conflict of interest.

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**ABSTRACTS SECTION**

## GAMMA RAY INDUCED VARIATION IN SEED CHARACTERISTICS OF *CICER ARIENTINUM* L. (VAR- VIJAY)

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### ABSTRACT

Gamma rays irradiated  $M_2$  population of *Cicerarietinum* L. (Var- Vijay) after post-harvest analysis revealed different variations in seed characters like, seed size, surface texture and seed coat color. Total eleven variants were noted with eight having different seed coat colors categorized in to four groups on the basis of seed size as normal, small, bold and extra bold two groups on the basis of surface texture and wrinkled or smooth surface. The seed yield of all the variants was noted as 100 seed weight, which was corresponding with seed size. The biochemical analysis of the seed mutants in the form of protein, iron and zinc content indicates that, dark seeds with dark colored seed coat have higher level of protein and minerals as compare to control.

**Key words:** Biochemical characters, *Cicerarietinum* L., gamma rays, post-harvest, seed characters, variation.

### INTRODUCTION

*Cicerarietinum* L., (Chickpea) is the second largest pulse crop in the world. It is one of most popular and chief source of plant protein along with mineral content like iron and zinc. Apart from this plant have some medicinal properties. Especially, seed grains with dark colored seed coat are reported to have more medicinal potential than normal faint color. On the other hand, the chickpea grains have significant level of phytic acid, which usually inhibit the actual content of proteins and minerals at consumers end. Chickpea variety-Vijay is one of the most preferred varieties by the farmers of Maharashtra state for its resistivity to wilt and comparatively high yield. Therefore, present study was directed to induce the variations in seed characters of variety- Vijay in relation to its protein and mineral content using gamma irradiation as mutagenic agent.

### MATERIAL AND METHODS

The germplasm of chickpea variety Vijay was procured from Mahatma PhuleKrishiVidyapithRahuri (MS). The dried, healthy seeds with 10-12% moisture content were irradiated with gamma rays with dose of 300, 400 and 500 Gy. The gamma ray (GR) irradiation facility ( $Co^{60}$  source) was made available from Bhabha Atomic Research Center, Trombay, Mumbai. The treated seeds were sown in October 2015 under field conditions at Departmental field of ShriShivaji College, Akola (MS) with spacing 15 cm within row and 30 cm between rows to raise  $M_2$  plants. The  $M_1$  plants were harvested individually and sown in October 2016 to raise  $M_2$  plant to row progenies. The untreated control was

sown on either side of each plot. The  $M_2$  progeny was closely monitored for morphological variations and harvested individually (Khan et al., 2005 and Kharkwal, 2003). The seed characters of the mutant population were observed during post-harvest analysis. The protein content of seed was analyzed as per Lowry et al., (1951) and Iron and zinc analysis was done by employing the method of Velu et al., (2006) and Velu et al., (2008) respectively.

### RESULTS AND DISCUSSION

On the basis of field observations and post-harvest data analysis, it was found that, gamma irradiated  $M_2$  population of Chickpea variety-Vijay has produced eleven different types of seed variants. The plants producing these seed variants are designated as mutant -1 to 11. These showed variation in seed coat color, seed size, seed surface texture, 100 seed weight and biochemical characters like protein content, iron and zinc content (Table-1).

The control seed coat color was dark brown with wrinkled surface. The identified mutant showed 08 different seed coat colors viz, brown, brown with black strips, light brown, black, green, golden yellow, yellow and yellow brown. These seeds with different seed coat colors were categorized into four groups according to their size as normal (control type), small, bold and extra bold and on the basis of surface texture as wrinkled/ rough and smooth (Table-01). The 100 seed weight (g) of the seeds was observed to correlate with seed size. The normal sized seed variant's 100 seed weight was ranging from 17.22 to 17.85 g, which is in the range of control, while in bold seeded it was 18.66 to 21.85 g and in extra bold seeds 24.28 g and



small seeds showed 16.22 g 100 seed weight (table-1). The protein content in seed coat color variants with dark color was found to range between 18.66 to 23.78 mg/g while that of mutants with light seed coat colors and control was 17.26 to 17.74 mg/g.

Considering biochemical characterization in terms of protein, iron and zinc content, the dark colored seeds were found to have more content as compare to light colored seed mutants and control. As far as mineral content is concern, we did not observe any strict correlation with other characters but, mutants with dark colored seed coat were found to have significantly higher level of both iron and zinc (Table-1 and Fig.1).

Some important reports on induced genetic visibilities in seed characters of different crops includes that of Pawar et al., (1979), Singh and Raghuvanshi (1991), Wani et al., (2013) Kashid and Subhash, (2015) and Gaffar et al., (2016). However, this is the first report demonstrating different degree of variation in chickpea seed characters correlated with their yield and biochemical characters.

### CONCLUSION

Gamma irradiation is a very efficient tool for mutation breeding to create genetic variability in crops that could be used to develop new varieties with desired characters like stress tolerance, high yield and high quality products. From the present report it is clear that lower doses of gamma radiations (300 Gy) could induce several variations in seed characters like seed coat color, size, surface texture, seed yield and biochemical such as proteins and minerals. Higher level of biochemical content could be correlated with dark seed coat color. These variations might be due to structural rearrangements in DNA upon exposure of lower doses of gamma radiations. The observed variations in seed characters could be used further to identify the genes responsible these changes at molecular level. Further, the stabilization of these characters could lead the development of new varieties with some novel characters.

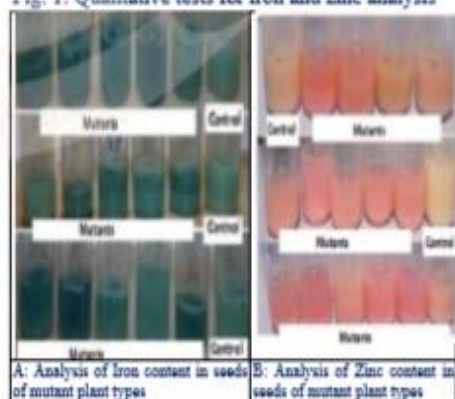
### ACKNOWLEDGEMENT

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**Table-1: analysis of different mutant plant types for variation in seed characters.**

Plant Types	Seed color	Seed size	Surface texture	100 seed wt. (g)	Protein (mg/g)	Iron (mg/100g)	Zinc (mg/100g)
Control	Dark Brown	Normal	Wrinkled	17.53 ± 0.62	17.72 ± 0.86	30.8-35.9	~37.0
Mutant -1	Black	Normal	Wrinkled	17.42	20.42	~60.0	50.0-65.0
Mutant -2	Brown with black strips	Extra bold	Wrinkled	24.28	23.78	30.0-39.9	38.0-49.0
Mutant -3	Green	Normal	Wrinkled	17.58	19.65	~60.0	50.0-65.0
Mutant -4	Golden yellow	Normal	Wrinkled	17.56	19.88	36.0-49.9	38.0-49.0
Mutant -5	Yellow	Normal	Wrinkled	17.22	17.52	36.0-49.9	~37.0
Mutant -6	Yellow brown	Bold	Wrinkled	19.21	18.66	30.0-39.9	38.0-49.0
Mutant -7	Brown	Bold	Wrinkled	19.86	19.02	36.0-49.9	38.0-49.0
Mutant -8	Dark brown	Bold	Smooth	21.33	19.56	~60.0	50.0-65.0
Mutant -9	Light brown	Bold	Smooth	21.83	19.83	30.0-39.9	38.0-49.0
Mutant -10	Brown	Normal	Wrinkled	17.82	17.74	30.8-35.9	~37.0
Mutant -11	Brown	Small	wrinkled	16.22	17.26	30.8-35.9	38.0-49.0

**Fig. 1: Qualitative tests for iron and zinc analysis**



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**ABSTRACTS SECTION**

## CHEMOPROFILING AND ANTIOXIDANT POTENTIAL OF A LESSER KNOWN *LEUCAS STRICTA*

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### ABSTRACT

*Leucas stricta* is one of the common, small, herbaceous member of Lamiaceae family. However, it was not focused for its medicinal potential as compared to its allied members. Present study is focused on its chemoprofiling using HPTLC and validating its antioxidant potential. It was observed that, this plant is rich in alkaloids, phenolic compounds, terpenoids, saponin and steroids. HPTLC analysis of methanolic extract of leaves of this plant showed several phenolic compounds like gallic acid, quercetin, catechin, apigenin and myrcetin. It was also found to have significant level of antioxidant potential as compare to standard antioxidant and positive check. Thus, the chemoprofiling of *L. stricta* validate that, it possesses antioxidant potential.

**Key words:** *Leucas stricta*, Lamiaceae, Chemoprofiling, HPTLC, Antioxidant.

### INTRODUCTION

*Leucas stricta* is a lesser known member of family Lamiaceae. It was found to be used as vegetable in South India (Muthu et al., 2006). However, it was not incorporated in the traditional healthcare systems of Indian sub-continent. The other members of the same family like *L. aspera* and *L. cephalotes* were routinely used by traditional as well as ayurvedic herbal healers. Probably, the ample availability of these counterparts, *L. stricta* remained un-attended. But, the scenario has changed over the period and now, these species are started declining. Therefore, it is essential to identify the other source with similar properties so that one can conserve the overexploitation of species under use. The present attempt was made to screen the *L. stricta* for its available phytochemicals and test its antioxidant potential.

### MATERIAL AND METHODS

The plant was collected from Narnala forest patches. The plant was identified taxonomically using local flora (Naik, 1998 and Karthikeyan & Singh, 2001) and a specimen copy was deposited in Department of Botany, ShriShivaji College, Akola (MS). The leaf material of the plant was shade dried for 8-10 days and then ground into fine powder and kept in airtight bottles until use. The preliminary phytochemistry of the powdered material was done using standard methods (Harborne, 1973 and Koche et al., 2010). Further, the methanolic powdered extract was used for HPTLC analysis. The HPTLC was done on CAMAG TLC unit using, Chloroform: Methanol (7.5:2.5) system on C18 column. The antioxidant

activity was measured as IC 50 following the method of (Blois, 1958).

### RESULTS AND DISCUSSION

The powdered material of leaves of *L. stricta* was found to rich in phytochemicals. Among the solvents used, most phytochemicals were observed to be extracted in methanol (table-1). It showed presence of alkaloids, phenolics, flavonoids, tannins, terpenes and steroids. It also showed significant amount of phenolic compounds and flavonoids (table-1).

Fig.1 showed HPTLC chromatogram of *L. stricta* methanolic leaf extract at 366 nm. It showed total 10 peaks. These peaks were interpreted on the basis of Rf values of separated compounds. Three peaks were found to correspond with the known phenolic compounds i.e. peak number 4 (Gallic acid, Rf value 0.30), Peak number 7 (Ferulic acid, Rf value 0.74) and Peak number 9 (Kaempferol, Rf value 0.87). Rest of the peaks might be of unknown compounds (table-2). When scanned at 540 nm, the HPTLC chromatogram of *L. stricta* methanolic leaf extract, it showed total 23 peaks (Fig. 2). When correlated the Rf values of these compounds with standard compounds, four phenolic compounds were identified (table-3). The identified peaks were – peak number 13 (Catechin, Rf value 0.39), Peak number 16 (Myricetin, Rf value 0.52), peak number 17 (Quercetin, Rf value 0.65), Peak number 19 (Vanolic acid, Rf value 0.73) and peak number 22 (Apigenin, 0.90).

The antioxidant potential of *L. stricta* extract was analyzed as DPPH scavenging activity and total antioxidant activity. In both cases, sample extract

was found to have significant activity more than standard antioxidant used for the study (table-4). Plants are rich in chemicals which are responsible its different biological properties, like antimicrobial, antioxidant, anti-inflammatory, diuretic, anthelmintic etc. It was now clear that, the plant with high level of phenolics and flavonoids showed antioxidant properties (Michalak, 2006; Prakash et al, 2009 and Kasote et al., 2015). Gupta and Sharma (2006) enlighten the area by summing the natural antioxidant plants used in traditional medicine. Recently Sulaiman et al., (2013) reviewed the natural antioxidants, their

analysis and evaluation. Further, Kamath et al., (2015) showed the direct relation between the phenolic content and antioxidant potential of the plant. Our report is positively correlated with these observations.

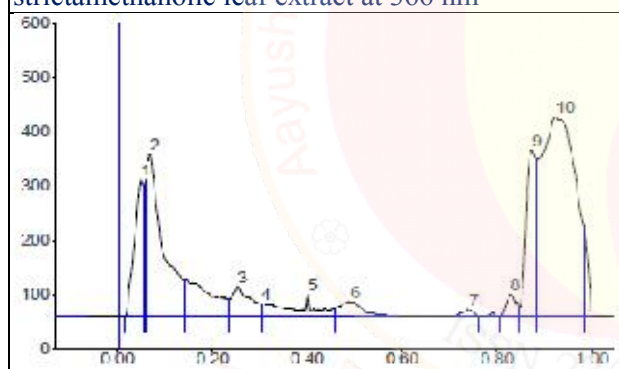
Form the study, it could be stated that, Leucas stricta is rich in chemical composition. Its HPTLC profile showed sizable amount of phenolics and flavonoids which could be correlated with the antioxidant potential of crude methanolic extract of L. stricta. Further, this plant could be a potential replacement for its counterpart plants for antioxidant property.

**Table-1: Preliminary phytochemical analysis of leaf powder of *L. stricta***

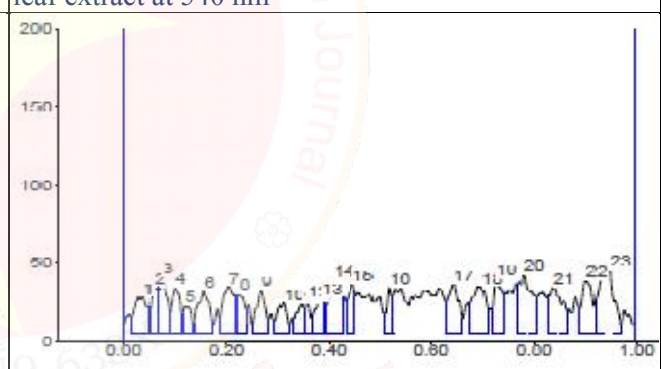
Leaf Powder	Test for phytochemicals					
	Alkaloids	Phenolics	Flavonoids	Tannins	Terpenes	Steroids
Aqueous Extract	-	-	+	+	-	+

Methanol Extract	+	(18.15 ± 0.33)	(08.69±0.05)	+	+	+
Ethanol Extract	+	+	+	-	-	+

**Fig. 1: HPTLC chromatogram of *L. stricta* methanolic leaf extract at 366 nm**



**Fig. 2: HPTLC chromatogram of *L. stricta* methanolic leaf extract at 540 nm**



**Table-2: HPTLC data of Methanolic leaf extract of *L. stricta* at 366 nm.**

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned compounds
1	0.01	0.2	0.05	252.3	17.68	0.06	241.7	4674.9	9.69	1. Unidentified
2	0.06	244.4	0.07	299.7	21.01	0.14	67.0	9679.5	20.06	2. Unidentified
3	0.24	33.5	0.25	55.5	3.09	0.30	22.7	1860.5	3.05	3. Unidentified
4	0.30	23.7	0.30	23.7	1.66	0.35	14.8	700.3	1.64	4. Gallic acid
5	0.38	12.1	0.40	40.8	2.80	0.41	12.1	383.0	0.79	5. Unidentified
6	0.46	16.3	0.49	27.2	1.91	0.53	6.6	1039.8	2.15	6. Unidentified
7	0.71	0.5	0.71	13.9	0.97	0.76	0.3	303.7	0.63	7. Ferulic acid
8	0.81	0.2	0.83	40.1	2.81	0.84	21.8	611.6	1.27	8. Unidentified
9	0.85	15.6	0.87	305.9	21.44	0.88	287.8	5640.4	11.69	9. Kaempferol
10	0.89	287.8	0.93	367.5	25.76	0.98	167.3	23279.7	48.23	10. Unidentified



**Table-3: HPTLC data of Methanolic leaf extract of *L. stricta* at 540 nm.**

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned compound
1	0.02	5.1	0.04	18.5	3.37	0.05	11.4	381.0	4.30	1. Unidentified
2	0.05	12.3	0.06	24.7	4.50	0.06	24.4	237.6	2.68	2. Unidentified
3	0.07	22.6	0.07	30.9	5.63	0.09	8.8	389.8	4.39	3. Unidentified
4	0.09	10.0	0.10	24.2	4.42	0.11	6.9	315.9	3.56	4. Unidentified
5	0.11	7.8	0.12	13.3	2.43	0.13	0.0	162.2	1.83	5. Unidentified
6	0.14	1.8	0.16	22.2	4.04	0.17	1.1	396.4	4.47	6. Unidentified
7	0.19	5.3	0.21	25.1	4.58	0.22	19.8	459.6	5.18	7. Unidentified
8	0.22	19.9	0.23	20.6	3.75	0.24	10.6	275.5	3.11	8. Unidentified
9	0.25	0.1	0.27	23.0	4.19	0.28	3.8	313.1	3.53	9. Unidentified
10	0.30	2.5	0.31	14.6	2.66	0.32	0.5	172.2	1.94	10. Unidentified
11	0.33	4.3	0.35	13.7	2.50	0.35	11.0	173.3	1.95	11. Unidentified
12	0.35	11.5	0.36	15.9	2.90	0.37	4.1	123.5	1.39	12. Unidentified
13	0.37	5.3	0.39	17.8	3.21	0.39	15.2	255.1	2.88	13. Catechin
14	0.39	13.1	0.41	28.9	5.27	0.43	17.8	542.4	6.12	14. Unidentified
15	0.44	16.2	0.45	26.5	4.83	0.45	19.0	321.2	3.62	15. Unidentified
16	0.51	8.0	0.52	24.4	4.45	0.52	19.3	196.4	2.21	16. Myricetin
17	0.63	16.0	0.65	26.6	4.84	0.66	3.8	481.3	5.43	17. Quercetin
18	0.68	13.7	0.70	25.1	4.58	0.71	10.2	574.5	6.48	18. Unidentified
19	0.72	10.5	0.73	29.8	5.44	0.74	20.8	430.5	4.85	19. Vanillic acid
20	0.77	26.6	0.78	33.2	6.05	0.81	19.2	766.4	8.64	20. Unidentified
21	0.83	19.2	0.84	24.4	4.45	0.87	10.0	529.1	5.96	21. Unidentified
22	0.89	11.1	0.90	29.4	5.36	0.92	12.8	483.5	5.45	22. Apigenin
23	0.92	14.4	0.95	35.9	6.55	0.97	0.7	889.3	10.03	23. Unidentified

**Table-4: Antioxidant activity of methanolic leaf extract of *L. stricta***

Leaf Extract	IC 50 value for antioxidant activity	
	DPPH scavenging activity	Total antioxidant activity
<i>Leucasstricta</i>	18.25	16.22
Standard	26.08	17.25

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**ABSTRACTS SECTION**

## GC-MS DETECTION AND DETERMINATION OF CHEMICAL CONSTITUENTS IN *CRATEVA RELIGIOSA* G. FORST. LEAVES

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### ABSTRACT

The present study was carried out to identify and analyze chemical constituents present in leaves of *Crateva religiosa* G. Forst. leaves by extracting them in solvents Chloroform, Dichloromethane and 50% Ethanol. Also Analytical, Physiochemical, Qualitative, Quantitative tests were performed for the understudy plant part. GC-MS analysis was also done for all the three extracts of *Crateva religiosa* G. Forst. leaves extracted in Chloroform, Dichloromethane and 50% Ethanol. The separated compounds were identified by comparing mass spectra with the NIST database. Retention time, Molecular name, Peak area percentage, Molecular weight and Molecular formula were ascertained to all the identified molecules. Molecular structures of the identified secondary metabolites were drawn by Marvin sketch Software.

**Keywords:** GC-MS, Chemical Constituents, Molecular structures

### INTRODUCTION

In the present time, natural products derived from plants are being tested for presence of new drugs with new modes of pharmacological action as inappropriate use of antibiotics has augmented the emergence of drug resistance to the micro-organisms and the extensive use of these synthetic drugs often cause considerable side effects. Recent studies are involved in the identification and isolation of new therapeutic compounds of medicinal importance from higher plants for specific diseases (Erturk *et al.*, 2006; Averineni, *et al.*, 2007).

*Crateva religiosa* G. Forst. also known as *Crateva adansonii* DC. is commonly called by various names like Sacred Garlic Pear; Temple plant; Varuna; Viawarna; Nirvala and belongs to the family Capparaceae. And its trade name is three leaved capper (Pullaiah, 2006). It is a rare medicinal plant in Maharashtra (Survase and Raut, 2011). It is an important medicinal plant and has been used in Indian Ayurvedic medicine for various ailments. A postal stamp was issued by the

Indian postal Department to commemorate this tree. In India, Some ethnic communities offer flowers to Baruna (God of rain) in the month of Chaitra “march and april” for better rainfall.

Leaves of this plant are are stomachic, tonic, aromatic, and sweet. The leaf juice in doses of ½ to 3 tolas is given in rheumatism mixed with coconut milk (or oil) and ghee (or butter); The fresh leaves bruised well with a little vinegar, lime juice or hot water, is applied to the skin as poultice or paste and takes 5-15 minutes to obtain rubefacient effect, if kept longer it acts as vesicant. Leaves are also used in healing of wounds and also applied to body swellings. The leaves are heated and applied to relieve backaches. The leaves are also used in a preparation to treat permanent retraction of the testis, blubs are used as an emetic and poison antidote. The Present study was undertaken to validate the ethnomedicinal uses and to know about the chemical composition of the Leaves of this plant which are of medicinal importance by using unique technique of GC-MS.

Gas Chromatography is a technique which is used for direct analysis of components existing in plants by separating the compounds by vaporizing them and same vaporized compounds are intrinsically detected by the flame ionization detector and electron capture detector (Leibler *et al.*, 1996). The spectrometric method such as mass spectrometry (MS) is coupled with Gas chromatography for the detection of separated compounds (Uzer *et al.*, 2005; Eisenhauer *et al.*, 2009). Mass spectrometry (MS) is the most sensitive and selective method for molecular analysis and can yield information on the molecular weight.

The plant has been already reported for having Wound healing (Ajali *et al.*, 2010); Anti-mutagenic activity (Chichioco-Hernandez *et al.*, 2009); Anti-inflammatory activity (Abdullahi *et al.*, 2012); suppressing various immune factors (Bani *et al.*, 2006); Antimicrobial activity (Agboke *et al.*, 2011; Wagay *et al.*, 2017); Antioxidant activity (Pandey *et al.*, 2013); and Antimycotic potential (Sahoo *et al.*, 2008). The chemical constituents of the understudy plant part are being reported for the first time by this study.

## **MATERIALS AND METHODS**

The plants were collected from Gandhigram village, Akola of Vidarbha region, Maharashtra and the morphological characters were noted which were then used for the identification of this plant. After morphological identification, plant herbarium was also authenticated by taxonomist Professor Dr. S.P. Rothe. The voucher specimens were deposited in the herbarium house of Department of Botany, Vidyabharati Mahavidyalaya, Camp, Amravati, Maharashtra, India. The Leaves of this collected plant were shade dried and then converted to powder form for further studies.

The procedures recommended in Indian Pharmacopoeia (Anonymous, 1966) and Gupta *et al.*, 2013 were followed for analytical or physiochemical study.

The extraction was done by using 10 grams of plant powder and 180ml of solvent viz. Chloroform, 50% Ethanol, and

Dichloromethane in the Soxhlet apparatus. The heating mantle temperature was set at evaporating temperature of the solvent and the cycles of soxhletion were done for about 6 hours. After extraction in Soxhlet apparatus for 6 hours the extracts were filtered and concentrated to 5ml using rotatory vacuum evaporator at room temperature. These concentrated samples were then used for further analysis.

The standard procedure for Qualitative analysis and quantitative analysis was followed (Wagay *et al.*, 2015).

The GC-MS analysis was carried out using gas chromatography-high resolution mass spectrometer. 2  $\mu$ l of the prepared extracts was employed for GC-MS analysis. The GC-MS analysis was carried using Shimadzu Make QP-2010 with column of 60 meter length, with 0.25 mm internal diameter and 0.32 thickness. Helium gas was used as carrier gas at constant flow rate of 1ml/minute. Injector temperature was set at 50  $^{\circ}$ C. the Oven temperature were programmed from 10  $^{\circ}$ C to 280  $^{\circ}$ C at 10  $^{\circ}$ C/minute to 200  $^{\circ}$ C then 10  $^{\circ}$ C/ 3 minutes to 250  $^{\circ}$ C ending with a 5 minutes isothermal at 280  $^{\circ}$ C. The sample was injected in split less mode.

Interpretation on mass spectrum of GC-MS was done using the National Institute Standard and Technology (NIST) database. The mass spectrum of compounds was compared with the spectral data of known compounds present in spectral libraries (NIST).The name, molecular weight and molecular formula of the identified molecules were ascertained by this analysis. The molecular structures of the important compounds were drawn by software Marvin sketch after identification.

## **RESULTS AND DISCUSSION:**

### **MORPHOLOGICAL CHARACTERS**

A small to moderate sized tree with a much-branched head. Bark grey, smooth horizontally wrinkled, wood is yellowish white, turning light-brown when old. Leaves deciduous, pinnately trifoliolate, clustered at the ends of branchlets with a common petiole 4-8 cm long at the summit of which are tree leaflets; ovate



lanceolate or obovate, acute or acuminate, attenuated at the base, entire, glabrous on both surfaces and reticulately veined. The lateral leaflets oblique at the base with a rather slender point at the tip; petiolules present. Flowers many, in dense terminal corymbs, greenish-white, pedicels long, Stout and glabrous. Sepals petaloid, small, ovate, and acute. Petals ovate or oblong (including the

claw). Stamens purplish longer than the petals, spreading. Gynophore nearly 5cm long, terete, smooth. Ovary ellipsoid; stigma flat. Fruit globose, woody, smooth or scurfy berry, with hard and rough rind, on the thickened gynophores. Seeds numerous, kidney-shaped, embedded in yellow pulp, nearly smooth & brown. Flowering occurs from March to May.



**Fig 1-3:** Morphological features of *Crateva religiosa* G. Forst. (1) Habit; (2) Bark; (3) Flower

**Analytical study**

S. No.	Parameter studied	%age value (w/w)	
1.	Moisture content	26.54 ± 0.721	
2.	Total Ash	20.77 ± 0.85	
3.	Acid soluble ash	29.68 ± 0.30	
4.	Acid insoluble ash	71.33 ± 1.22	
5.	Water soluble ash	28.08 ± 0.89	
6.	Water insoluble Ash	71.68 ± 0.28	
7.	<b>Extractive values in</b>	Chloroform	8.25 ± 0.15
		50% Ethanol	17.22 ± 0.87
		Dichloromethane	3.33 ± 0.64

**Table No 1:**

Analytical values of *Crateva religiosa* G. Forst. Leaves

Note: Percentage mean (n=3) ± SD

**Powder study**

**Table No 2:** Organoleptic evaluation of *Crateva religiosa* G. Forst. Leaves

S. No.	Particulars	Observation
1.	Colour of powder	Greenish grey
2.	Odour	Odorless
3.	Taste	Sweet
4.	Texture	Smooth

**Table No 3:** Fluorescent behavior of *Crateva religiosa* G. Forst. Leaf powder on treatment with different chemical reagents Leaves

S. No.	Powder + Reagent used	Leaves	
		Visible Light	UV Light
1.	Powder as such	Green	Fluorescent green
2.	Powder + Conc. H <sub>2</sub> SO <sub>4</sub>	Black	Greenish Black
3.	Powder + Conc. HNO <sub>3</sub>	Orange	Orange red
4.	Powder + Conc. HCl	Black	Greenish black
5.	Powder + 10% NaOH	Yellow	Yellowish green
6.	Powder + 1 N HCl	Light yellow	Fluorescent green
7.	Powder + Iodine solution	Dark reddish brown	Dark black
8.	Powder + 5% FeCl <sub>3</sub>	Dark brown	Light brown
9.	Powder + KI	Dark brown	Yellowish green
10.	Powder + 1 N HNO <sub>3</sub>	Light yellowish brown	Fluorescent green
11.	Powder + 1 N H <sub>2</sub> SO <sub>4</sub>	Yellowish white	Greenish transparent
12.	Powder + Ethyl acetate	Light green	Fluorescent green

### Phytochemical studies

#### i) Qualitative phytochemical Analysis

**Table No 4:** Qualitative Phytochemical Screening of *Crateva religiosa* G. Forst. Leaves

S. No	Constituents	Chemical tests	LEAVES			
			W	D	E	C
1	Alkaloids	Wagners test	++	++	++	++
		Mayers test	++	--	++	--
2	Flavonoids	Sodium hydroxide test	++	--	++	--
		Lead acetate test	++	--	++	--
3	Glycosides	Killer killiani test	--	--	--	--
		Fehlings test	++	--	++	--
4	Phenols	Phenols test	--	--	--	--
5	Saponins	Frothing / Foam test	++	--	--	--
6	Steroids	Salkowaski test	--	--	--	++
		LB test	--	--	++	--
7	Tannins	Ferric chloride test	--	--	--	--
8	Terpenoids	Salkowaski test	++	--	--	++

Note '+'= Present and '-'= Absent

Where, W= Water; D= Dichloromethane; E= 50% Ethanol; C= Chloroform respectively.

The qualitative phytochemical screening of *Crateva religiosa* G. Forst. in four extracts i.e., Water, Dichloromethane, 50% Ethanol and Chloroform showed that there was prominently presence of phytoconstituents like Alkaloids, Flavonoids, Phenols, Glycosides, Saponins, Steroids and Terpenoids. However, Tannins were absent in all the extracts of *Crateva religiosa* G. Forst leaves. Water and 50% Ethanol extracts showed presence of

most phytoconstituents followed by Chloroform and Dichloromethane.

#### ii) Quantitative Phytochemical Analysis

The crude content of major phytochemical compounds in *Crateva religiosa* G. Forst. were also determined using the standard procedures, for Alkaloids (Harborne, 1973), Flavonoids (Bohm and Kocipai- Abyazan, 1994), Saponins (Obdoni & Ochuko, 2001) and Phenolics (Vermerris *et al.*, 2006). It was found that among all the five tested

phytochemicals, the plant showed higher level of Flavonoids then Alkaloids which is also closely followed by Saponins. The content of Phenols was in least percentage in the *Crateva religiosa* G. Forst.

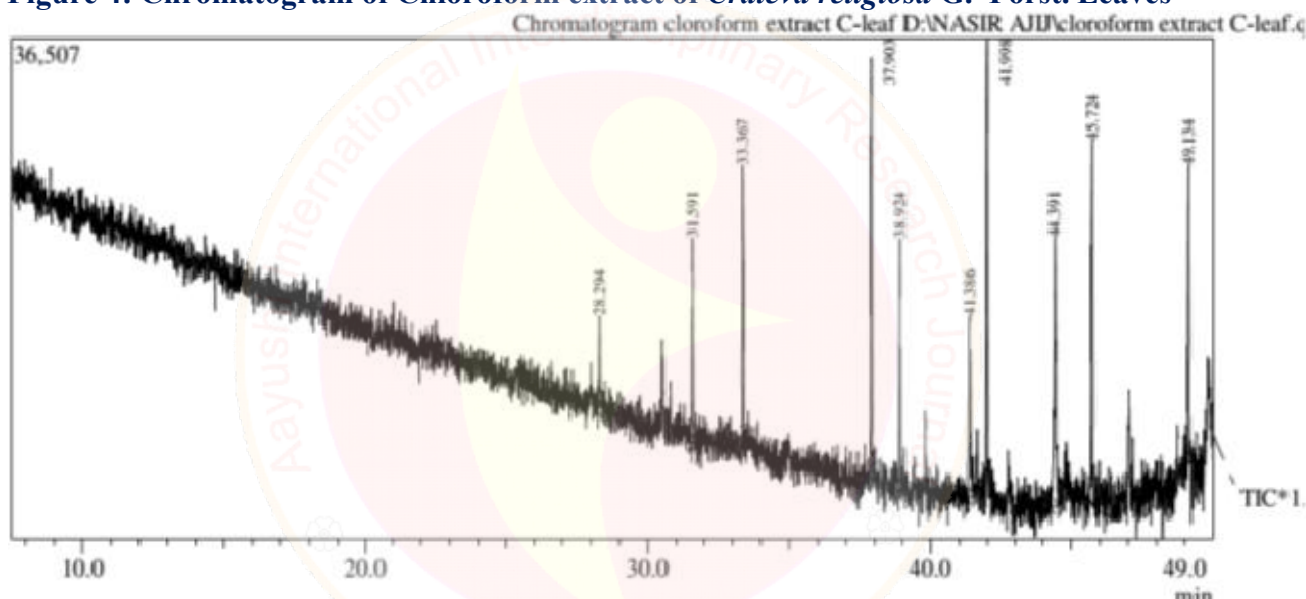
1.	Flavonoids	7.86 ± 0.46
2.	Alkaloids	0.88 ± 0.056
3.	Saponins	0.96 ± 0.28
4.	Phenols	1.16 ± 0.10

Note: Percentage mean (n=3) ± SD  
 Phenols: expressed as gallic acid equivalents (GAE/g)  
 Flavonoids: expressed as quercetin equivalent (mg/g)

**Table No 5: Quantitative Phytochemical analysis of *Crateva religiosa* G. Forst. Leaves**

S. No.	Phytochemicals	% of Crude Content (g / 100 gms of dry sample)
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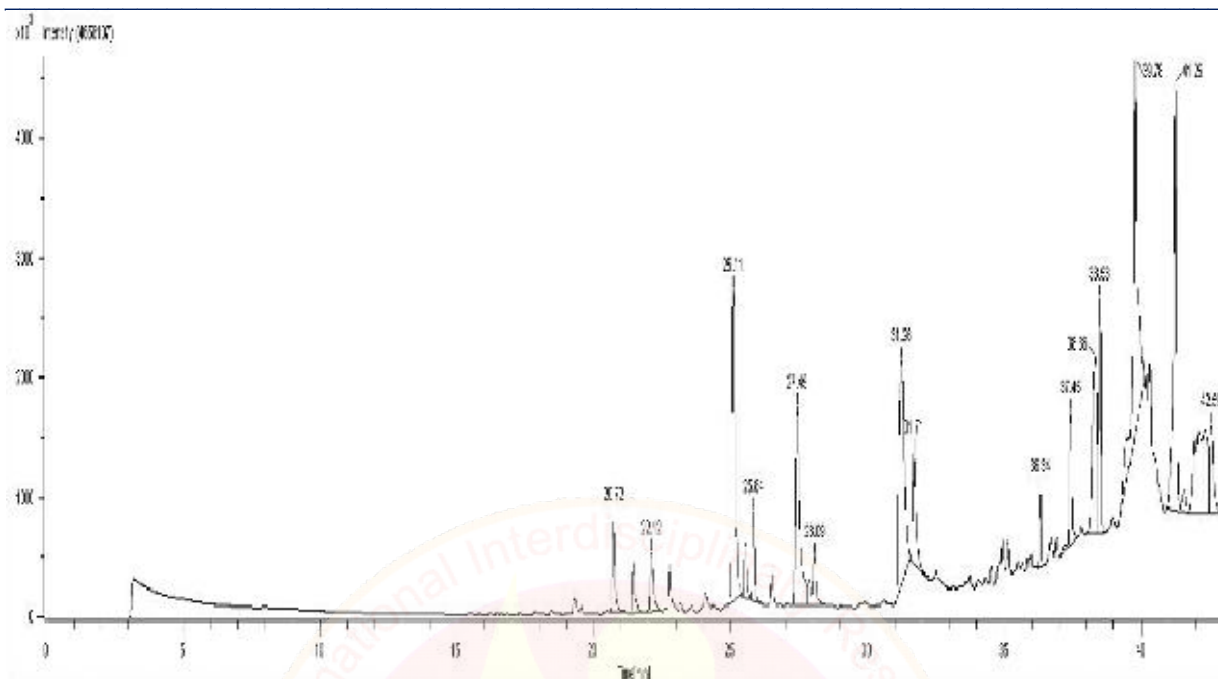
**Figure 4: Chromatogram of Chloroform extract of *Crateva religiosa* G. Forst. Leaves**



**Table No 6: Compounds identified in the Chloroform extract of *Crateva religiosa* G. Forst. Leaves**

S. No.	RT	Name of Compound	Peak Area %	MW	MF
1.	28.294	1-Tetradecene	2.58	196	C <sub>14</sub> H <sub>28</sub>
2.	31.591	Phenol,2,4-bis(1,1-dimethylethyl)-	6.82	206	C <sub>14</sub> H <sub>22</sub> O
3.	33.367	1-Pentadecene	7.63	210	C <sub>15</sub> H <sub>30</sub>
4.	37.903	1-Pentadecene	13.46	210	C <sub>15</sub> H <sub>30</sub>
5.	38.924	Z-2-Dodecen-1-ol	8.06	184	C <sub>12</sub> H <sub>24</sub> O
6.	41.386	n-Hexadecanoic acid	9.55	256	C <sub>16</sub> H <sub>32</sub> O
7.	41.998	5-Eicosene, (E)-	12.73	280	C <sub>20</sub> H <sub>40</sub>
8.	44.391	Dihydrogeraniol	17.29	156	C <sub>10</sub> H <sub>20</sub> O
9.	45.724	5-Eicosene, (E)-	13.46	280	C <sub>20</sub> H <sub>40</sub>
10.	49.134	1-Docosene	8.42	308	C <sub>22</sub> H <sub>44</sub>

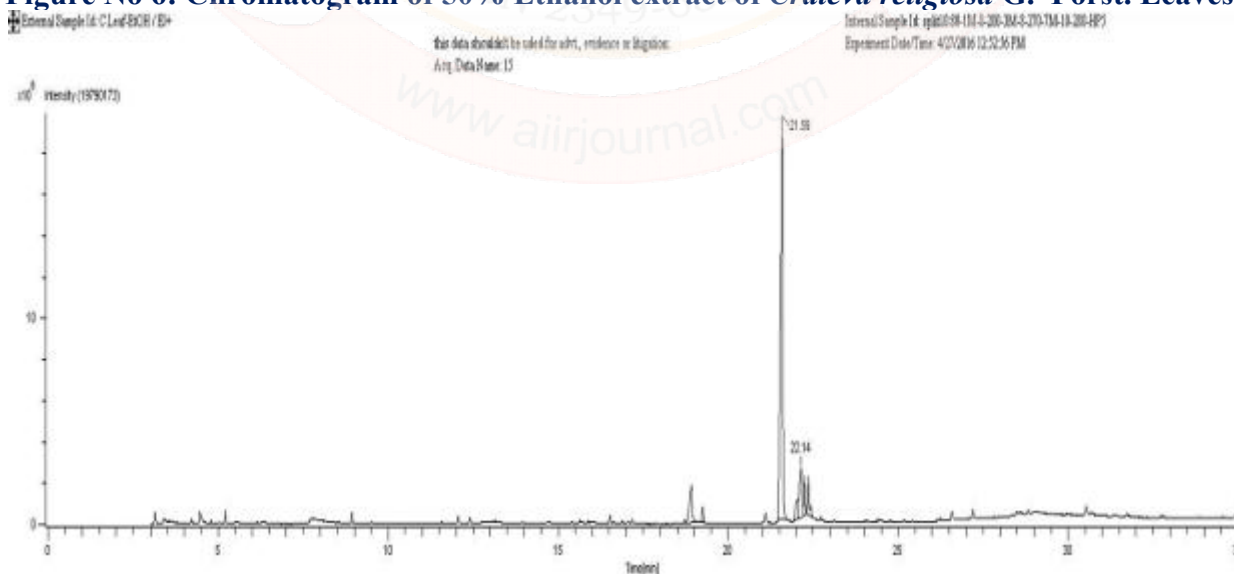
**Figure No 5: Chromatogram of Dichloromethane extract of *Crateva religiosa* G. Forst. Leaves**



**Table No 7: Compounds identified in the Dichloromethane extract of *Crateva religiosa* G. Forst. Leaves.**

S. No.	RT	Name of Compound	Peak Area %	MW	MF
1.	20.72	Cyclolongifolene oxide, dihydro	2.45	218	C <sub>15</sub> H <sub>22</sub> O
2.	22.12	Ar-tumerone	1.82	216	C <sub>15</sub> H <sub>20</sub> O
3.	31.71	Octadecanoic acid	3.63	284	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>
4.	38.53	Heptacosane	4.95	380	C <sub>27</sub> H <sub>56</sub>
5.	40.33	Methyl 5-[2-(3-furyl)ethyl]-5,6,8a-trimethyl-3,4,4a,5,6,7,8,8a-octahydro-1-naphthalenecarboxylate	3.76	330	C <sub>21</sub> H <sub>30</sub> O <sub>3</sub>
6.	41.26	Tetracosane, 1-bromo-	12.52	416	C <sub>24</sub> H <sub>49</sub> Br

**Figure No 6: Chromatogram of 50% Ethanol extract of *Crateva religiosa* G. Forst. Leaves**





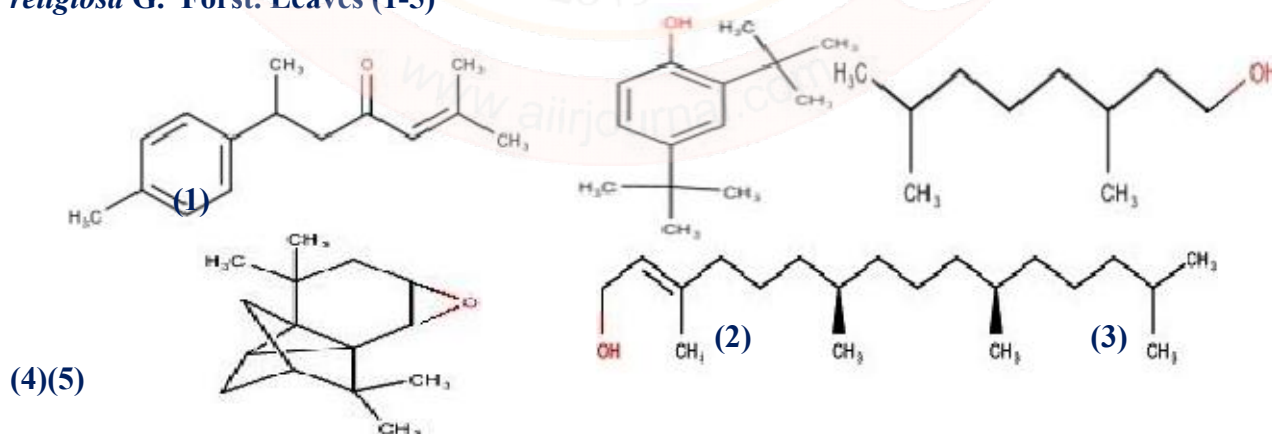
**Table No 8: Compounds identified in the 50% Ethanol extract of *Crateva religiosa* G. Forst. Leaves**

S. No.	RT	Name of Compound	Peak Area %	MW	MF
1.	18.91	n- Hexadecanoic acid	7.77	256	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>
2.	19.25	Hexadecanoic acid	1.74	284	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>
3.	21.59	Phytol	68.66	296	C <sub>20</sub> H <sub>40</sub> O
4.	22.14	9,12,15-Octadecatrienoic acid, [Z.Z.Z]-	9.23	278	C <sub>18</sub> H <sub>30</sub> O <sub>2</sub>
5.	22.25	n-Propyl 9,12-octadecadienoate	3.71	322	C <sub>21</sub> H <sub>38</sub> O <sub>2</sub>
6.	22.36	Ethyl,9,12,15-octadecatrienoate	4.79	306	C <sub>20</sub> H <sub>34</sub> O <sub>2</sub>

**Table No 9: Secondary metabolite profile of *Crateva religiosa* G. Forst. Leaves**

S. No	Name of Compound	Category
1.	Ar-tumerone	Sesquiterpene / Terpenoid
2.	Phenol,2,4-bis(1,1-dimethylethyl)-	Phenol
3.	Dihydrogeraniol	Monoterpenoid / Terpenoid
4.	Cyclolongifolene oxide, dihydro	Sesquiterpene / Terpenoid
5.	Phytol	Terpenoid

**Molecular structures of the important secondary metabolites identified in the *Crateva religiosa* G. Forst. Leaves (1-5)**



Ar-tumerone (1) has been reported for acting as an Antidote for snake bite (Ferreira *et al.*, 1992), anti-apoptotic activity (Aratanechemuge *et al.*, 2002).

Phenol,2,4-bis(1,1-dimethylethyl)- (2) has been reported for its Anti-bacterial activity (Kuppuswamy *et al.*, 2013), Antifungal and Antioxidant activity (Raja *et al.*, 2011) Antiinflammatory activity (Sanitha *et al.*,



2015) Anticancerous activity (Pereira *et al.*, 2009).

Dihydrogeraniol (**3**) has been reported to cure Skin infections (Kelm *et al.*, 2000), Also used for Fragrances and cosmetics, Contact allergen, P-miticide, use regulated in EU, non-corrosive for skin (Terratox™ Explorer Database).

Cyclolongifolene oxide, dihydro (**4**) is mostly component of essential oil (Paritala *et al.*, 2014; Mahmoudi and Nosratpur, 2013).

Phytol (**5**) has been reported to have Anti-inflammatory activity (Ogunlesi *et al.*, 2009), Antimicrobial and diuretic activity (Rajeswari *et al.*, 2012) Anti-cancer activity (Himaja *et*

*al.*, 2014), Cancer preventive activity (Sermakkani *et al.*, 2012), Antimalarial activity and also used for Joint dislocation, Hernia (Dandekar *et al.*, 2015).

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**ABSTRACTS SECTION**



## MICROPROPAGATION STUDIES ON *SCHLEICHERA OLEOSA* (LOUR). BELONGING TO FAMILY SAPINDACEAE FROM VIDARBHA REGION.

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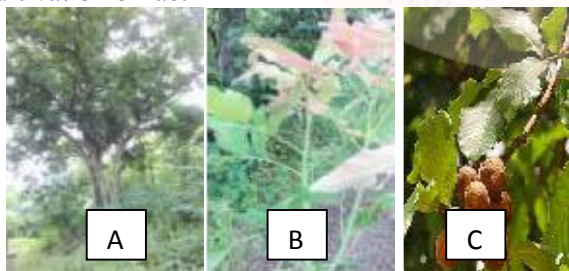
### ABSTRACT

*Schleichera oleosa* (Lour.) belongs to the family Sapindaceae. It is a large deciduous tree. The timber is suitable where hardness, bending strength and toughness are required. It is about 70 percent heavier than teak tree. The wood of this tree is commonly used for oil and sugar mills, cart wheels, and agricultural implements. Oil extracted from the seed of kusum tree, called 'kusum oil', is a valuable component of true Macassar oil used in hairdressing, culinary and lighting purposes and in traditional medicine. At present situation the plant become rare in natural forest therefore micropropagation study is carried out for the conservation of this plant. The Murashige and skoog's (MS) basal medium with different growth regulators showed best result.

**Key words:** MS medium: Murashige & Skoog Medium, 2, 4-D:2, 4-diphenylphenoxyacetic acid, BAP:6-benzylaminopurine, NAA: Naphthaleneacetic acid, KN: Kinetin, lit: litre, M: mole.

### INTRODUCTION

*Schleichera oleosa* (Lour) commonly known as kusum. In local area this plant known as Mokha. Deciduous trees. Flowering and fruiting is from March-June. This tree utilized for multipurposes with both commercial importance and tribal sentiments which is worth conserving. Kundu et,al(2011)Its provides good shade. This plant produce excellent quality lac resin.(Roonwal ML,1962)Different plant parts of *Schleichera oleosa* (Lour)are used as tribal food, animal feed, seed oil and timber.It is also important source of traditional medicines (Dipnarayan Saha,2012).The plant has been traditionally used for the treatment of pain inflammation, itching, ulcers.(Shrinivas K. et, al.2011).The bark of kusum is often used to control tissue swelling by voids.(D.Saha et,al,2010),The major used of this tree is for cultivation of lac.



A. Tree of *Schleichera oleosa* B. New leaves C. Friut of *Schleichera oleosa*

### MATERIAL AND METHODS

The experimental plant material was collected from Vanoja forest Dist. Akola. The complete experiment was performed under aseptic and controlled condition. Apart from the well-equipped

tissue culture laboratory .In the present study, one of the most widely employed Toshio Murashige and Folke Skoog (MS) media (1962) was chosen as a basal media throughout the experiment. The MS medium was supplemented with certain adjuvant like vitamins, auxins and cytokines etc. for initiation of growth, establishment of cultures, maintenance and multiplication of culture and morphogenesis in the explants under aseptic conditions.

The media was prepared in sterile double distilled water by adding desired supplements such as, concentrated stock solutions of macronutrients, micronutrients, iron and organic supplements with various combinations and concentrations of plant growth regulators. Plant Growth Regulators BAP,kinetin and NAA were added after the MS basal media attains room temperature. The Hydrogen ion concentration (pH) of the media was adjusted to 5.8. The Sucrose 30gm was added as carbon source and dissolved properly. Finally 8gm agar was added to the media and volume made upto 1 liter by using double distilled water. The media was then boiled to melt the agar.The medium was then poured in tissue culture autoclaved test tubes. Then test tube containing media was allowed to stand for solidification of media. Contamination was checked after 24 hours.The culture bottles containing MS media are ready for inoculation of explants.

The procedure was performed in laminar air flow cabinet to maintain aseptic conditions throughout the experiment. The explants that were used are nodal part, inter nodal part, leaves of *Schleichera oleosa* (Lour) were washed thoroughly with running tap water and then with sterile double

Effect of different plant regulators on various explant of <i>Schleichera oleosa</i> (Lour)						
Sr.No.	Explant	Plant Growth Regulators	Conc. mg/lit	Shoot length(cm)		Average Length
				30-40 Day	50-60 Day	70Day
1	node	IBA + BAP	2 + 1.5	Callus formation	Multiple shoot Induction	+++
2	node	IBA + BAP	0.05+0.1	No callus	No callus	-
3	leaf	BAP + 2,4-D	1.5+ 0.5	Callus formation	Multiple shoot Induction	+++
4	leaf	BAP + 2,4-D	1.5+2	Weak callus	-	++
5	internode	NAA+2,4-D	0.5+2	No callus	-	-
6	internode	BAP+ kinetin	1.0+1.0	Callus formation	Multiple shoot Induction	+++
7	shoot tip	kinetin NAA	+ 3+2	No callus	-	-
8	shoot tip	kinetin NAA	+ 1+0.5	Callus formation	Multiple shoot Induction & rooting	+++

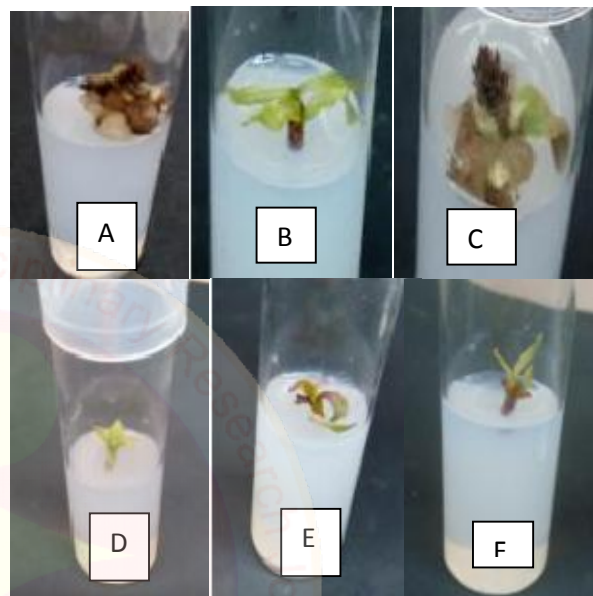
distilled water. The various explants were surface sterilized with 70% alcohol for nearly two minute. Then the explants were immersed in 0.1% mercuric chloride (HgCl<sub>2</sub>) for one minute, followed by rinsing with sterile double distilled water for 3-4 times. Then the explants were soaked by placing them on sterile filter papers (tissue paper) Then the explants were cut into small pieces with sterile scalpel under laminar air flow and inoculate on MS medium containing different concentrations of growth regulators. Burner was used to avoid contamination during handling.

### RESULT AND DISCUSSION

In combination of IBA+BAP(2+1.5)mg/lit, BAP+2,4-D(1.5+ 0.5)mg/lit, BAP+ kinetin (1.0+1.0 )mg/lit, kinetin + NAA (1+0.5)mg/lit ,node, leaf, internode, shoot tip explants respectively were inoculated and the initiation of callus and induction of multipal shoot and rooting was noted . A satisfactory response of initiation and elongation of shoot was observed .After nine weeks of culture on the rooting medium, the plantlets with well developed shoots and roots

were acclimatized in the greenhouse. During the first two weeks in the greenhouse, air relative humidity was maintained. Under these conditions maximum plantlets survived and resumed normal growth.

**Table. Effect of different plant regulators on various explant of *Schleichera oleosa* (Lour)**



**Figures.A&B Callus induction. C,D,E,F initiation and elongation of shoot and Roots Conclusion**

To conclude from this study, while using different combinations of growth regulators at different concentrations it was concluded that different explants responded distinctly. Callusing was observed in node ,shoot tip, internode and leaf explants when supplemented with different growth regulators. Shooting was observed in shoot tip explants when media was supplemented with growth regulators in different combinations at different concentrations such as kinetin + NAA(1+0.5 mg/lit), BAP + 2,4-D (1.5 + 0.5 mg/lit), and IBA + BAP (2 + 1.5 mg/lit). And shoot tips showed good shoot initiation when inoculated in media supplemented with BAP + 2,4-D (1.5 + 0.5 mg/lit).Both shooting and rooting was obtained in media containing the growth regulators in combination of IBA + BAP at a concentration of 2+1.5 mg/lit.

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**ABSTRACTS SECTION**



## SYNTHESIS OF N-GLUCOPYRANOSYL SUBSTITUTED 1, 3, 4-THIADIAZOLIDINE DERIVATIVES AND ITS APPLICATION AS ANTIMICROBIAL AGENTS

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### ABSTRACT

A series of 2-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-4-phenyl-5-substituted benzoylimino-1, 3, 4-thiadiazolidines have been synthesized by the interaction of 1-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-3-phenyl amino thiocarbamide with various substituted benzoyl isocyanodichlorides. The synthesized compounds were structurally confirmed by analytical and IR,  $^1\text{H}$  NMR and Mass spectral analysis. In the present investigation the In-vitro antimicrobial activity of compounds has been evaluated by using several bacteria and fungi. All compounds studied shows satisfactory antimicrobial activity.

### INTRODUCTION

Heterocyclic compounds are found to exhibit anti-inflammatory, anti-parasitic, anti-tubercular, antidiabetic activity<sup>1-3</sup>. In recent years, there has been increasing interest in the synthesis of heterocyclic compounds by cyclization of appropriate linear compounds. Organosulfur compounds play an important role in modern organic synthesis. Recently in our laboratory there are various reports on sugar heterocyclic possessing antimicrobial and antifungal activities<sup>4-6</sup>.

In view of applications of these compounds in various fields, we report the synthesis of a series of 2-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-4-phenyl-5-substituted benzoylimino imino-1, 3, 4-thiadiazolidines (**3a-h**) have been synthesized by the interaction of 1-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-3-phenyl amino thiocarbamide (**1**) with various substituted benzoyl isocyanodichlorides (**2a-h**).

The structures of the products were confirmed by the spectral (IR,  $^1\text{H}$  NMR and Mass<sup>8-15</sup>) and elemental analysis (Table 1).

### EXPERIMENTAL

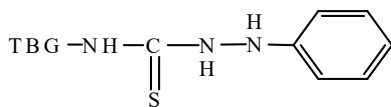
Melting points of all synthesized compounds were determined using open capillary tube on Mac digital melting point apparatus and were uncorrected. The IR spectrum was recorded in KBr Disks on SHIMADZU IR affinity – 1 – FTIR spectrometer. The NMR spectrum was recorded in Bruker DRX – 300 instruments operating at 300 MHz using  $\text{CDCl}_3$  solution with TMS as internal standard. The mass spectrum was recorded on a

THERMO Finnigan LCO Advantage max ion trap Mass Spectrometer. Specific rotations were measured on Equip-Tronics EQ-801 Digital Polarimeter. Thin layer chromatography (TLC) was performed on silica gel G for TLC (Merck) and spots were visualized by iodine vapours.

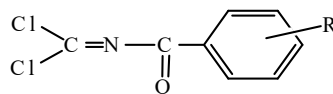
### GENERAL METHODS

1. Synthesis of 1-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-3-phenyl amino thiocarbamide (**1**): It was prepared by refluxing Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl isothiocyanate and phenyl hydrazine in benzene for 1 hr.
2. Preparation of substituted benzoyl isocyanodichlorides (**2a-h**)<sup>7</sup>: It was prepared by passing excess amount of gaseous chlorine into the solution of substituted benzoyl isothiocyanates in chloroform.
3. 2-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-4-phenyl-5-substituted benzoylimino-1, 3, 4-thiadiazolidines (**3a-h**)  
Mixture of 1-Tetra-O-benzoyl- $\beta$ -D-glucopyranosyl-3-phenyl amino thiocarbamide (**1a**) (0.002M, 1.49gm) and benzoyl isocyanodichloride (**2**) (0.002M, 0.268gm) was reflux in chloroform for about 3 hr. The chloroform was distilled off and the sticky mass triturated several times with petroleum ether (60-80 °C) to afford solid (**3a**). It was purified by ethanol-water.  
Similarly, when the reaction was extended to other substituted benzoyl isocyanodichlorides (**2b-h**) the corresponding 1, 3, 4-thiadiazolidines (**3b-h**) has been synthesized.

**SCHEME:**

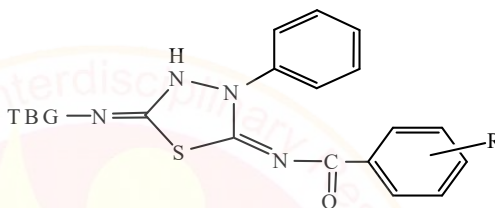


1-Tetra-*O*-benzoyl- $\beta$ -D-glucopyranosyl  
3-phenyl amino thiocarbamide (**1**)



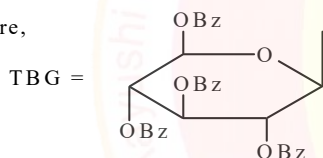
Substituted benzoyl isocyanodichlorides (**2**)

Reflux  
Chloroform  
3 hr



2-Tetra-*O*-benzoyl- $\beta$ -D-glucopyranosyl-4-phenyl-5-substituted benzoylimino-  
1,3,4-thiadiazolidines (**3a-h**)

Where,



Bz = COC<sub>6</sub>H<sub>5</sub>

R = a) H, b) *o*-Cl, c) *m*-Cl, d) *p*-Cl, e) *o*-methyl, f) *m*-methyl, g) *p*-methyl h) *p*-methoxy.

**SPECTRAL ANALYSIS**

**3a:- IR(KBr cm<sup>-1</sup>):** 3061 (Aromatic C-H), 2972 (Aliphatic C-H), 1735 (C=O), 1600 (C=N), 1313 (C-N), 1269 (C-O), 1070 (Characteristics of glucose), 713 (C-S); **<sup>1</sup>H NMR (CDCl<sub>3</sub>, ppm):**  $\delta$  7.9-7.2 (35H, m, Aromatic protons), 6.12-4.29 (7H, m, glucopyranosyl protons), 6.57-6.56 (1H, s, hump N-H); **Mass (m/z):** 874, 730, 619, 517, 457, 335, 287, 227, 105.

**3d:- IR(KBr cm<sup>-1</sup>):** 3059 (Aromatic C-H), 2954 (Aliphatic C-H), 1730 (C=O), 1651 (C=N), 1600 (C=C), 1315 (C-N), 1273 (C-O), 1070 (Characteristics of glucose), 713 (C-S).; **<sup>1</sup>H NMR**

(CDCl<sub>3</sub>, ppm):  $\delta$  7.89-7.40 (34H, m, Aromatic protons), 4.34-6.11 (7H, m, glucopyranosyl protons), 6.34 (1H, s, N-H), 2.39-1.9 (3H, s, CH<sub>3</sub>); **Mass (m/z):** 909 (M<sup>+</sup>), 745, 729, 667, 579, 335, 231, 108.

**3h:- IR(KBr cm<sup>-1</sup>):** 3061 (Aromatic C-H), 2960 (Aliphatic C-H), 1735 (C=O), 1620 (C=N), 1600 (C=C), 1313 (C-N), 1278 (C-O), 1070 (Characteristics of glucose); **<sup>1</sup>H NMR (CDCl<sub>3</sub>, ppm):**  $\delta$  7.89-7.40 (34H, m, Aromatic protons), 5.75-3.74 (7H, m, glucopyranosyl protons), 6.84 (1H, s, N-H), 3.42 (3H, s, O-CH<sub>3</sub>); **Mass (m/z):** 905 (M<sup>+</sup>), 723, 619, 579, 457, 257, 231, 105

**Table 1: Physical Data of compounds 3(a-g)**

Sr. No.	Products	m.p. (°C)	Yield (%)	R <sub>f</sub> Value	Elemental Analysis %		[ $\alpha$ ] <sub>D</sub> <sup>31</sup> (c, in CHCl <sub>3</sub> )
					Found (Required)		
					N	S	
1.	3a	179	74	0.86	6.39 (6.40)	3.65 (3.66)	+81.2° (0.97 in CHCl <sub>3</sub> )
2.	3b	185	72.94	0.92	6.08 (6.16)	3.45 (3.52)	+84.1° (0.91 in CHCl <sub>3</sub> )
3.	3c	174	85.21	0.87	6.07 (6.16)	3.48 (3.52)	+42.2° (0.96 in CHCl <sub>3</sub> )

4.	3d	178	70.50	0.79	6.18 (6.16)	3.49 (3.52)	+52.6° (0.95 in CHCl <sub>3</sub> )
5.	3e	190	75.25	0.85	6.28 (6.33)	3.60 (3.60)	+85.1° (0.94 in CHCl <sub>3</sub> )
6.	3f	181	76.27	0.92	6.29 (6.30)	3.61 (3.60)	+122.2° (0.9 in CHCl <sub>3</sub> )
7.	3g	171	80.21	0.81	6.32 (6.30)	3.59 (3.60)	+52.50° (0.96 in CHCl <sub>3</sub> )
8.	3h	196	76.57	0.78	6.06 (6.18)	3.52 (3.53)	+76.2° (0.94 in CHCl <sub>3</sub> )

### ACKNOWLEDGEMENT

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**ABSTRACTS SECTION**



## MONITORING AIRBORNE FUNGAL MYCOFLORA IN COLLEGE LABORATORY

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### ABSTRACT

Air is important part of environment. Various airborne particles are present in the environment. Air pollution is the most serious problem to human health. Aerobiological studies are important for providing qualitative and quantitative information about airborne fungal organism. The present study was conducted to analyse the mycoflora from Botany laboratory were investigated between June 2015 to May 2016, In this study, 15 species was isolated and identified from the selected laboratory. The Petri-plate expose method using PDA (Potato dextrose agar) CZA (CzapekDox Agar) media in petri-plate. 15 species were identified rest which are not identified were kept as unidentified fungi. *Aspergillusniger*(18.33%) and *Aspergillusfumigatus* (20.45%) These are the dominant species than rest of the fungi.

**Keywords:** PDA, CZA, Petriplate, Mycoflora, *Aspergillusniger*

### INTRODUCTION

The term aerobiology was first coined by the American plant pathologist "Fred cambellmeier" in 1930. So the term aerobiology came in use since 1930 to denote the airborne fungal spores, pollen grains and other airborne microorganisms. The outdoor environment is never completely free from the incidence of microbial prop gules, which are collectively called as "air spora". The term "air spora" is suggested by Gregory in 1952. Airborne fungi, the most important group in the air, have been well known as the cause of contamination problems in the environment and in human activities. Normally, fungi as saprophytes, play a significant role as primary decomposers of substrates in various ecosystems (Nazim et al. 2012). Air is a natural medium for certain very minute particle including many mycoflora. Fungal spores constitute a significant fraction of airborne bioparticles (Meraj et al 2000, Durugbo et al 2013). Investigation on aeromycoflora in libraries was carried out in past by many workers (Ghosh et al 2014, Sinha et al 1998). Airborne mycoflora are largely determined by topography, meteorological parameters, vegetation and biotic factors including human activities (Lacey 1981). However, airborne fungi are involved in the respiratory system infections. For example, polypoid chronic rhinosinusitis has been caused by *Bipolarisspicifera* (Buzina et al. 2003), while *Aspergillusfumigatus* is associated with respiratory symptoms in patients with asthma (Fairs et al. 2013). Accordingly, the aeroallergen from

*Alternaria sp.* is one of main factors causing allergy in human respiration (Cordon & Millington 2001). Further, some groups of airborne fungi can grow inside office furniture and lead to exposure to volatile organic compounds (VOCs) or occupation-associated cancer epidemics due to aflatoxicosis (Gedikoglu et al. 2012). Aero mycology deals with the study of air borne fungi and their spores. Fungi have both beneficial and nuisance effect on our lives. They destroy our food. Fabrics, leather and other articles. They are also responsible for causing large number of diseases in the plants like Rust, Smut Blight, and Mosaic etc.

### MATERIAL AND METHODS

The present study was conducted in college laboratories by petri plate expose method. The petri plate expose method is uses for isolation of fungal species by using PDA (Potato dextrose agar) and CZA (CzapekDox Agar) media at monthly intervals with petri-plate from June 2015 to May 2016. Petri-plate were expose in 10 minute of Botany laboratory and incubate at  $30 \pm 10C$ . for 4-5 days. For the species identification, specimen microscopic slide were prepared with the help of glycerine jelly as mounting media and lacto-phenol cotton blue as the standard stain.

### RESULT AND DISCUSSION

Present investigation was undertaken to analysed , a total 15 species was isolated and identified the identified species were *Aspergillusniger*, *A. fumigatus*, *A. flavus*, , *A. nidulans*,

*Alternaria alternata*, *Alternaria tenuissima*, *Curvularia lunata*, *Penicillium citrinum*, *Fusarium spp.*, *Rhizopus spp.*, *Mucor spp.*, *Chaetomium spp.*, *Helminthosporium spp.*, *Geotrichum spp.*, *Drechslera spp.* According to the expose petri-plate samples the total percentage of samples were recorded in Table 1. *Aspergillus niger* (21.44%), *A. fumigatus* (21.70%) and *Curvularia lunata*, (17.57%) were of high occurrence. *A. flavus* (5.42%), *Penicillium citrinum* (7.23%), *Alternaria alternata* (5.16%), *Alternaria tenuissima* (5.42%) and *Fusarium spp.* (4.13%) were of moderate occurrence and remaining seven are low occurrence. *A. nidulans* (1.29%), *Rhizopus spp.* (1.80%), *Mucor spp.* (1.29%), *Chaetomium spp.* (1.55%), *Helminthosporium spp.* (1.03%), *Geotrichum spp.* (1.29%) and *Drechslera spp.* (0.77%). *Aspergillus niger* and *A. fumigatus* are highest dominating airspora in selected laboratory.

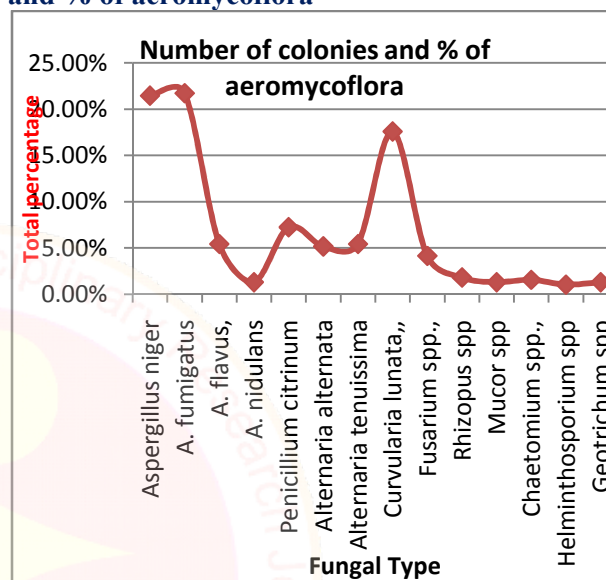
Table 1: Number of colonies and % of aeromycoflora

Sr.No.	Fungal Type	Fungal colonies	Total percentage
1	<i>Aspergillus niger</i>	83	(21.44%),
2	<i>A. fumigatus</i>	84	(21.70%)
3	<i>A. flavus</i> ,	21	(5.42%)
4	<i>A. nidulans</i>	05	(1.29%)
5	<i>Penicillium citrinum</i>	28	(7.23%),
6	<i>Alternaria alternata</i>	20	(5.16%)
7	<i>Alternaria tenuissima</i>	21	(5.42%)
8	<i>Curvularia lunata</i> ,	68	(17.57%)
9	<i>Fusarium spp.</i> ,	16	(4.13%)
10	<i>Rhizopus spp</i>	07	(1.80%)
11	<i>Mucor spp</i>	05	(1.29%)
12	<i>Chaetomium spp.</i> ,	06	(1.55%)
13	<i>Helminthosporium spp</i>	04	(1.03%)
14	<i>Geotrichum spp</i>	05	(1.29%)
15	<i>Drechslera spp</i>	03	(0.77%)
<b>Total</b>		387	-

Contribution percentage of individual species as per the standard formula:

$$\% \text{ of contribution} = \frac{\text{Total no. of colonies of one species}}{\text{Total no. of colonies of total species}} \times 100$$

### Graphical representation of number of colonies and % of aeromycoflora



### CONCLUSION

From the above discussion it was concluded that, various type of fungal species was identified and isolated. Out of which some are harmful and cause health problem and respiratory disorder. It is may be because of favourable condition for the growth of fungi.

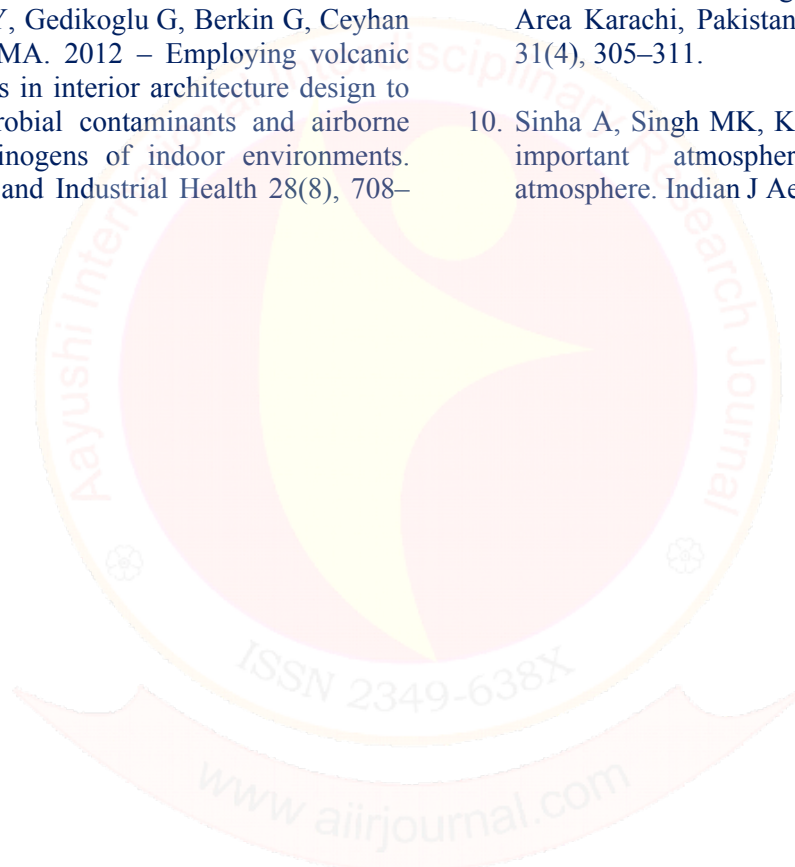
### ACKNOWLEDGEMENTS

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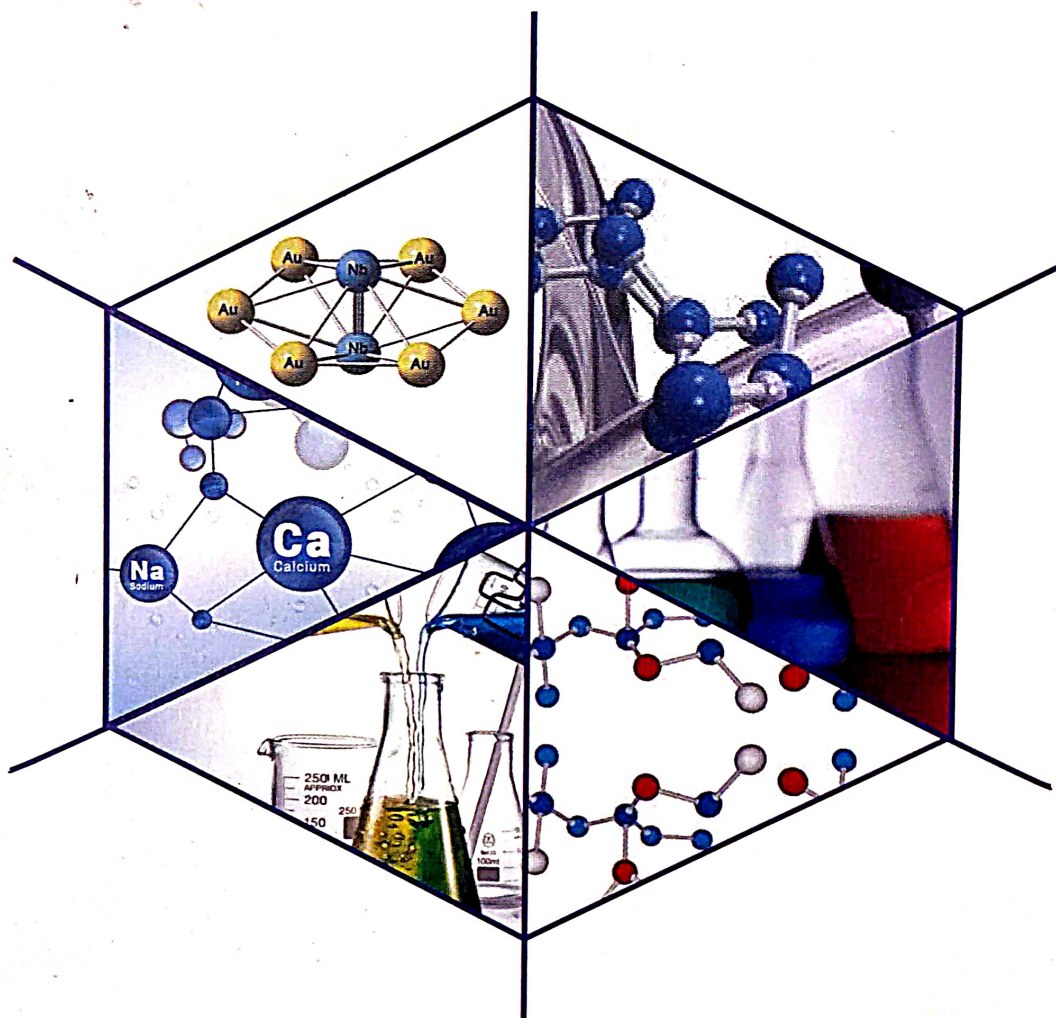
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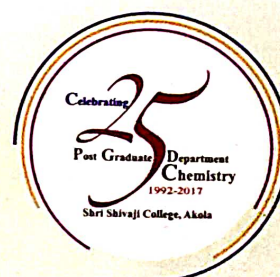


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of the reaction and purity of the synthesized chalcones was monitored by thin layer chromatography with F252 silica gel precoated aluminium sheets using petroleum ether – ethyl acetate (9:1) as a developing solvent and spots were visualized with near UV light. The IR spectra were recorded on SHIMADZU SPECTROPHOTOMETER using KBr pellets and expressed in  $\text{cm}^{-1}$ . The NMR spectra were recorded on BRUKER ADVANCE (400 FT-NMR) spectrophotometer in DMSO using tetramethyl silane as an internal standard and chemical shift values are expressed in  $\delta\text{ppm}$ . The MASS spectra were recorded on Waters UPLC-TQC MASS Spectrometer.

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## Plant Electricity and Few Electrochemical Reactions

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### Abstract:

Different researchers from various fields are trying their best to introduce special methods of converting the chemical energy produced by plants through the process of photosynthesis into electricity. Such method of non-conventional energy conversion could be a vast source of renewable energy in near future. It is today's demand to investigate and develop various types of renewable, sustainable, pollution free and highly efficient energy sources. Throughout this research paper, few fundamental investigations for generating an electrical energy (potential difference) from living plants are practiced. The energy is generated by embedding the sensors and cells at various points in the plant to allow flow of electrons using reduction and oxidation reactions. Multiple tests has been conducted using different type of electrodes, cells and sensors, and attempted to determine the characteristics of the generating system. Though the research work in such a field is in infancy, the researcher was able to produce the potential difference of about 4.5 volts using such a way. For optimum output, lots of new types of materials has been tested and used in combination to give better performance for the development of such type of plant energy. We, the researchers, expects that the green and clean energy will be utilized for few low power electrical and electronic appliances for their operation in near future.

Many plants material contents various type of organic and inorganic materials which are absorbed by their leave and root systems. These chemicals and minerals can be utilized as an electrolyte to occur the electrolysis process. The electrodes and cells can be prepared using various types of materials like Copper, Aluminum, Zinc, Lead, Iron, Carbon, Steel, Silver, Gold, Tungsten, Brass and Platinum. Also, different type of shapes and sizes of the electrodes as well as cells were tested for the optimum output of voltage. The sap flow present inside the plant itself, was taken as an electrolyte needed for the chemical process. Few electrochemical reactions are discussed throughout this research paper.





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**ABSTRACTS SECTION**



## APPLICATION OF BIONANOFERTILIZER IN COMBINATION FOR NEW PERSPECTIVE IN PRECISION AGRICULTURE

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### ABSTRACT

Attentive use of nanotechnology can assure the food safety through improving agricultural production. The present study reports the effect of combination of Copper and Iron nanoparticles (Bionanofertilizer) on germination percentage and germination rate of wheat seeds. The nanoparticles were synthesized by adding the 1mM solution of CuSO<sub>4</sub>, FeSO<sub>4</sub> separately into 24hr old supernatant of *Bacillus* sp. (Bioreduction). To study the germination test, the one set of wheat seeds were soaked in 10, 20, 30, 40 and 50µg/ml concentrations of Cu and Fe nanoparticles separately and the other set of wheat seeds were soaked in same concentrations of combination of Cu and Fe nanoparticles. The separate and combinatorial application of nanoparticles of concentrations 30 and 40µg/ml respectively showed increased in number of seminal roots and germination percentage of wheat as compared to control. The antifungal activity of separate Cu and Fe nanoparticles and the combination of nanoparticles were checked against some pathogenic fungi (*A. flavus*, *F. oxysporum*, *C. lunata* and *Phoma*) of plants. Among the two, the combinatorial nanoparticles showed better results of seed germination and antifungal activity and will prove to be the better bionanofertilizer to augment the agricultural yield.

**Key words:** FeNPs, CuNPs, Antifungal activity, Bionanofertilizer, seed germination.

### INTRODUCTION

Nanotechnology is the emerging field of science and has various applications in many fields. Nanotechnology is the science, engineering and technology dealing with the particle size of 1 to 100 nm. The large surface area, high surface energy and quantum confinement are the unique properties of nanoparticles. Due to these unique features the nanoparticles may result in different environmental fate and behaviors than their bulk counterparts (Singh *et al.*, 2015).

Different types of nanoparticles can be synthesized by various physical, chemical, biological and hybrid methods. The use of toxic chemicals in physical and chemical methods greatly limits their biomedical applications. Therefore to expand the use of nanoparticles there is a need of development of reliable, nontoxic and eco-friendly methods for synthesis of nanoparticles and the use of microorganisms to synthesize nanoparticles is one of the best options to achieve this goal. The chemical methods are able to produce large quantities of nanoparticles with a defined size and shape in a relatively short time, but they are complicated, outdated, costly, inefficient and produce hazardous toxic wastes which are harmful to environment and human health also. The biological method limits the use of expensive

chemicals and it is also environment friendly and it is supported by further approach that majority of bacteria inhabit ambient conditions of varying temperature, pH and pressure. The nanoparticles generated by biological process have high catalytic reactivity, greater surface area and improve contact between the enzyme and metal salt (Xiangqian Li *et al.*, 2011).

The use of nanoparticles in agriculture promotes the efficiency of plants to uptake more nutrients, endures environmental pressure and increases the resistivity against diseases (Jhazab *et al.*, 2015). Nanotechnology has potential to provide food security by enhancing crop production through precision farming, efficient utilization of water, protection against diseases, new material for pathogen detection and protection of the environment (Razzaq *et al.*, 2016). Plants grown in nutrient medium provided with nanosilver can uptake and accumulate nanoparticles (Lee *et al.*, 2012). The application of Iron nanoparticles improved agronomic traits of soyabean (Sheykhbaglou *et al.*, 2010). The present study was carried out to explore the role of combination of iron and copper nanoparticles as a bionanofertilizer to enhance wheat growth, germination percentage.

## MATERIAL AND METHODS

### Synthesis of nanoparticles

The CuNPs and FeNPs were synthesized by biological method. The 100ml, 1mM solution of CuSO<sub>4</sub> and FeSO<sub>4</sub> were separately inoculated with 100ml of 2 days old supernatant of *Bacillus sp.* The two mixtures then incubated at room temperature for 48 hrs in dark condition. After 48 hrs the synthesis of CuNPs and FeNPs were observed by change in the colour of initial solution to dark bottle green and dark brown respectively. The 1mM solutions of CuSO<sub>4</sub> and FeSO<sub>4</sub> were taken as a control.

### Characterization of nanoparticles

The synthesized CuNPs and FeNPs were characterized by UV-visible spectrophotometry and XRD.

### Germination test

To study the germination test the wheat seeds were sterilized with ethanol for near about 5 min and then the one set was soaked in a separate solution of nanoparticles (FeNPs and CuNPs) of concentrations 10, 20, 30, 40 and 50 µg/ml. The second set of wheat seeds was soaked in a mixture of Fe and Cu nanoparticles. According to concentrations the soaked seeds were placed in a respective sterilized petri plates containing filter

paper. The germination percentage was calculated after 6 days by using following formula:

Germination percentage = (Number of seeds germinated/ Total number of seeds)×100

## RESULTS AND DISCUSSION

Effect of FeNPs, CuNPs and its combination on germination percentage and number of seminal roots are presented in Table 1. The FeNPs and CuNPs individually showed the 80% germination at the same concentration i.e. 30 µg/ml. As compared to individual nanoparticles the combination of FeNPs and CuNPs had pronounced effect on germination percentage. It showed 90% germination at the concentration of 30µg/ml. Both the NPs had pronounced effect on the development of more number of seminal roots at the concentration of 30µg/ml as compared to control (fig.1). Further increase in concentration of FeNPs, CuNPs and its combination, significantly decreased the germination percentage and number of seminal roots also.

Table1: Comparative study of FeNPs, CuNPs and its combination on germination percentage and number of seminal roots

Concentrations	Germination % using FeNPs	No. of seminal roots	Germination % using CuNPs	No. of seminal roots	Germination % using combination	No. of seminal roots
10 µg/ml	60	3	60	3	60	3
20 µg/ml	70	4	70	3	70	5
30 µg/ml	80	4	80	5	90	6
40 µg/ml	60	4	70	4	70	4
50 µg/ml	40	2	40	2	40	2

The control (distill water) shows 70% germination but the germinated seeds are infected with fungi

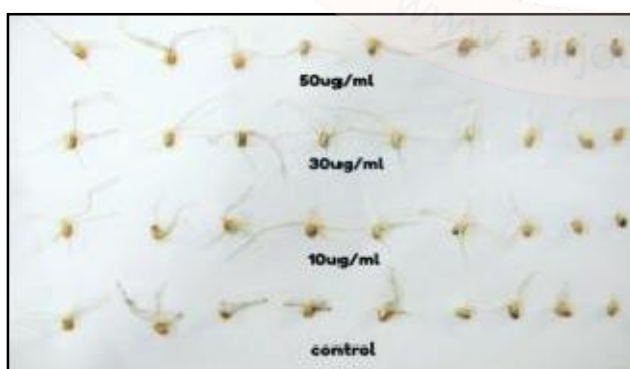


Fig.1 Effect of combination of FeNPs and CuNPs on number of seminal roots

The higher concentrations were found to inhibit the germination percentage in the percentage in the present study. Similar effect of AgNPs on

germination percentage have been reported. At higher concentration the AgNPs showed inhibitory effect on the germination percentage of wheat (Razzaq *et al.*, 2016). Most study had done on effect of AgNPs on various plant species. Contrary to this, present study investigated the effect of FeNPs and CuNPs. AgNPs significantly enhanced the leaf area, yield and N, P and K use efficiency (Jhazab *et al.*, 2015).

The antifungal activity of separate Cu and Fe nanoparticles and its combination is shown in Table 2. The combination of nanoparticles showed better results of antifungal activity against plant pathogenic fungi (*Fusarium oxysporum* and *Aspergillus flavus*) as compared to the individual nanoparticles. The nanoparticle also gives the good results against bacteria also. The FeNPs showed good antibacterial activity against *E. coli*

and *S. aureus* (Pande *et al.*, 2015). The Ag and Cu nanoparticles have not affected the growth and development of *P. herpotrichoides*, suggesting that the effect of nanoparticles is determined by the

plant's responses to the pathogen rather than the phytotoxic action of the Cu or Ag nanoparticles (Belava *et al.*, 2017).

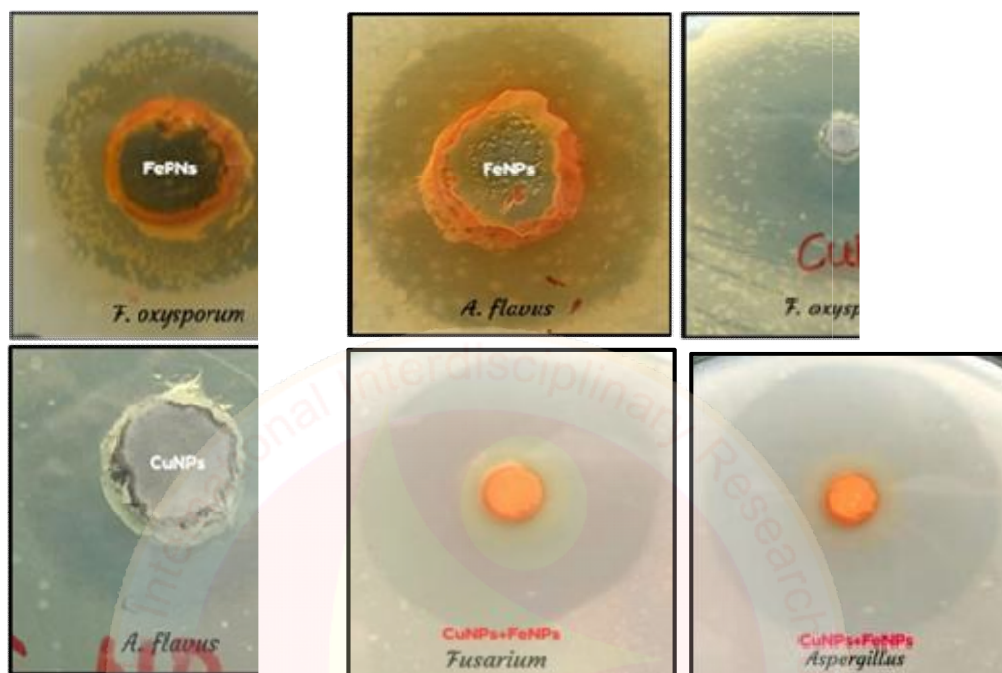


Fig 2: Antifungal activity of Cu and Fe nanoparticles and its combination against plant pathogenic fungi.  
 Table2: The antifungal activity of Cu and Fe nanoparticles and its combination against plant pathogenic fungi.

Nanoparticles	Zone of inhibition in mm	
	<i>Fusarium oxysporum</i>	<i>Aspergillus flavus</i>
FeNPs	17mm	14mm
CuNPs	13mm	18mm
Combination	23mm	20mm

### CONCLUSION

The combination of Fe and Cu nanoparticles produced good effect on the germination

percentage and number of seminal roots in wheat at particular concentration (30µg/ml) as compared to control and individual nanoparticles. The high concentration of nanoparticles was found to inhibit the germination percentage and number of seminal roots. Therefore a particular concentration of nanoparticles is needed for good results according to plant species. The exposure of wheat seeds to nanoparticles also prevents the wheat seeds from fungal infection. The combination of Fe and Cu nanoparticles showed good antifungal activity against plant pathogenic fungi (*Fusarium oxysporum* and *Aspergillus flavus*) as compared to individual nanoparticles. The present study reveals that the attentive use of combination of seed applied nanoparticles enhanced the germination percentage and number of seminal roots in wheat and will prove to be the good bionanofertilizer

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**ABSTRACTS SECTION**

## MORPHOLOGICAL VARIATION OF THRIPS INSECT *ELAPHROTHRIPS PROCER* (SCHMUTZ) (THYSANOPTERA:PHLAEOTHRIPIDAE)

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### ABSTRACT

Detailed study of insect *Elaphrothrips procer* (Schmutz) was conducted from larva to adult. During developmental stages the morphological variations were observed in the stages of larva I, larva II prepupa, pupa I and pupa II. On the basis of morphology, the male thrips is stronger than female and shows developmental variation than that of the female. Male thrips shows minor and major form i.e. normal, gynaecoid forms and overgrown, oedymorous form respectively. The male is longer than female, dark in colour and having elongated tubular abdomen than female. The size of the body length of the adult male is 7107  $\mu\text{m}$  and female is 4984  $\mu\text{m}$  long.

**Key words:** *Elaphrothrips procer*, developmental stages, gynaecoid, oedymorous, antenna

### INTRODUCTION

Thrips are occur on the tender, succulent parts of the plants, or under the barks of dead and drying twigs or among decaying leaves of grass. They enjoy a wide range of distribution, habits and ecological habitat. Though most of them are phytophagous, very few are predaceous feeding on mites, scales, psocids. Some of them produce and inhibit plant galls. While mycophagous or fungus feeding thrips are more common.

*Elaphrothrips procer* (Schmutz) (Family-Phlaeothripidae) is a mycophagous thrips. They are found within the curved folds of fungal infected dry leaves and feeds on fungal spores and generally occurs on the fungus infected dry leaves of *Butea monosperma* plant during humid seasons of the year. They have peculiar, piercing and sucking type of mouth parts with vestigial right mandible. Adults have two pairs of narrow fringed wings with long hairs and abdomen is long elongated with tube like anal segment.

### MATERIALS AND METHODS

The thrips were collected from their host plant *Butea monosperma* dry fungal infected leave during the humid periods of the year when they mostly occur near the roadside of highway and field area.

The adult male, female, larvae and eggs were kept in large plastic bowls along with fungus infected dry leaves. Then they transfer to the separate plastic rearing bowl to avoid overcrowding and food limitation. For protection bowls were covered by muslin cloth. Light 12:12 and temperature ( $25\pm 1^{\circ}\text{C}$ ) were maintained. Relative humidity

maintained at 80% by keeping wet filter paper in the rearing bowl, some time wet cotton plug also used.

Developmental stages and adults were dehydrated with ethanol grades (30%, 50%, 70%, 90%, and Absolute) then after cleared in xylol, embedded in DPX for mounting onto slides. In the case of specimens long preserved in 70% alcohol, before mounting to put them in 96 % alcohol to which ethyl acetate (1 part of ethyl acetate to 10 volume of water) has been added so as to make the materials soft.

For study of external morphology, slides of whole mounts of larvae, pupae and adults were used. All observations were made under compound microscope (MLX series Magnus) and stereo zoom microscope (Magnus) for better and detailed study of the sample. Computerised micro measurements were also done by using image analysis software (Olympus).

### RESULTS AND DISCUSSION

Larva and the adult have similar structure of the antennae, the antennal setae, sensorium and the mouth parts in both adapted for the fungus feeding. The integuments are sclerotinized at the head, thorax and abdominal terminal segment IX and X.

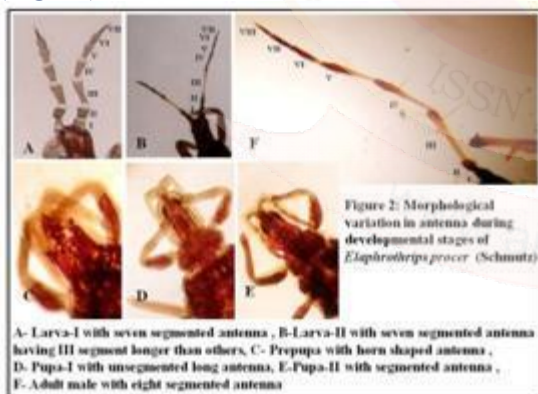
**Larva I:** Body is elongated, red in colour. Body measurement is about  $1.85 \pm 0.03 \mu\text{m}$  in length. The head is cylindrical and legs are grey with red pigments. The abdominal terminal segments IX and X are dark in colour (**Fig.1B**). The antennae are greyish in colour with total seven segments, the basal single scape, the middle single

pedicel and the distal large five jointed filaments flagellum (Fig.2A).



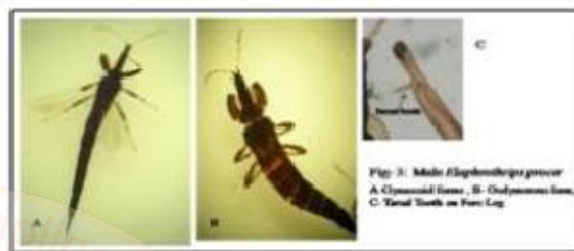
**Larva II:** Body is reddish in colour elongate in shape tapering towards the posterior end. The length of body is measures about  $3.78 \pm 0.12$  mm (Fig.1C). Antennae are seven segmented (Fig.2B). The abdomen has ten segments with rudiment eleventh segment bearing the whorl of setae. The abdominal segment IX and X is dark in colour.

**Prepupa:** Body is bright red in colour and cylindrical in shape tapering posterior. The body measures about  $2.54 \pm 0.17$  mm in length. The last abdominal segment is slightly darkish. Body is covered with a thin soft integument. The appendages show glassy texture (Fig. 1D). The antenna is short horn-like and unsegmented, situated on the frontolateral side with pairs of long thin setae at the front of each antenna. Antennae are distinctly clear and somewhat transparent (Fig.2C).



**Pupa I:** The body is red coloured, cylindrical, elongated in shape broader anteriorly and gradually tapering towards the posterior end. The body length measures about  $4.07 \pm 0.02$  mm. The Head is oblong in shape. Head bears a dark pigmented spot just behind the eyes (Fig.1E). The antennae lie along the lateral side of the head capsule. Antennae segmentation is not shown (Fig. 2D). The abdominal segments are similar to that of prepupa except in size.

**Pupa II:** Body is cylindrical more elongated than the prepupa and Pupa I but in shape similar to that of pupa I. The body length is measures about  $5.63 \pm 0.06$  mm in length. Pupa II is similar to prepupa and pupa I except the size (Fig.1F). The antennae are situated on the lateral sides of the head capsule. Antenna is segmented and elongated. In front of the antenna there are four pairs of elongated setae (Fig.2E).



**Adults:** On the basis of morphology, the male thrips is stronger than female and shows developmental variation than that of the female (Fig 3 G and H). Male thrips shows minor and major form i.e. normal, gynaecoid forms and overgrown, oedymorous form respectively (Fig.3 A and Fig.3 B). The male is longer than female, dark in colour and having elongated tubular abdomen than female. The male posses tarsal tooth on foreleg (Fig. 3.C) whereas in female tarsal tooth is absent. The size of the body length of the adult male is  $7107 \mu\text{m}$  and female is  $4984 \mu\text{m}$  long.

Pinet *et al.*, (2003), Mound and Morris (2003), Mound (2007) identified and classified the thrips on morphological account and explain the Thysanoptera phylogeny. Priesner (1928), Pesson (1951), Snodgrass (1954), Davies (1961), Stannard (1968), Ananthakrishnan (1969), Heming (1973), Moritz, (1997) termed first pupal instar as prepupa or propupa as it represent a very a short transitional stage and indistinctly developed wings pads and relatively smaller than that of the next stage. In the life history of *Elaphrothrips procer* (Schmutz) the prepupal stage has been lie in between the larva II and pupa I. Therefore, the following terminology is used in *Elaphrothrips procer* (Schmutz) i.e. first stage larva I, second larva II, prepupa, pupa I and pupa II similar with *Elaphrothrips greeni* Bagnall (Watane, 1985) In the larvae of *Bactridothrips brevitubus* the antennae are six segmented (Haga, 1974) while in *Haplothrips verbasci*, *Frankliniella fusa* (Heming, 1975) and *Elaphrothrips greeni* the antennae are seven segmented (Watane, 1985). In *Elaphrothrips procer* (Schmutz) the antennae are seven segmented and Prothorax is bilobed due to



presence of sclerotization, which are the common features of the thrips, *Haplothrips niger* (Loan and Holdaway 1955); *Bactridothrips brevitubus* (Haga, 1974); *Haplothrips verbasci* (Heming, 1978) and also in *Elaphrothrips greeni* (Watane, 1985). The abdominal segments IX, X and XI are indistinct in *Bactridothrips brevitubus* (Haga, 1974), while in *Elaphrothrips greeni* (Watane, 1985) except segment XI, other segment IX and X are distinct. *Elaphrothrips procer* (Schmutz) agree with the characters of abdominal segment present in *Elaphrothrips greeni*.

## CONCLUSION

*Elaphrothrips procer* (Schmutz) shows an intermediate type of development as the young ones resemble the adults in some characters. They are hemimetabolous type, while remaining three stages tend towards the holometabolous type of development as in the pupal stages. The change from the hemimetabolous type to the holometabolous type is achieved by the interpolation of a transitional prepupal stage. Presence of eight segmented antenna in adult and seven segmented antenna in larva-I and larva-II while showing intermediate prepupal stages are became the characteristic feature of the Thysanopteran, Phlaeothripidae insects.

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**ABSTRACTS SECTION**

## FEEDING EFFICIENCY OF MALE AND FEMALE *POECILIA RETICULATA* IN DIFFERENT WATER SAMPLE

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### ABSTRACT

Predation potentiality of a biological control agent, the guppy fish, *Poeciliareticulata* (=Lebistesreticulatus), was studied over the mosquito larvae under laboratory conditions. The third instar larvae of *Anopheles mosquito* were used as prey. The average consumption rate was 41.0 per day per fish. Feeding efficiency of female fish than male fish was comparatively more in pond water, drainage water and ground water. Comparatively more feeding efficiency observed in pond water than drainage and ground water. The female fish was more active (57.3) than male ones (29.7). The study suggests that this fish could be used, after careful field trial, as a promising and sustainable biological control agent in controlling malarial disease and other mosquito-borne diseases.

**Keyword :** predation potentiality, biological control, *Poecilia reticulata*.

### INTRODUCTION

Natural predators are used in biological control. This type of control is an environmentally friendly preventative method to control, among others, populations of pest organisms (Becker, 2006). Several organisms have proved to be an effective predators against mosquito larvae such as larvivorous fish, mosquito of the genus dragonflies and damselflies, cyclopoids copepods, nematode *Bacillus thuringiensis* H14 (WHO, 1986). Biological control of mosquitoes was very popular during the early part of the 20th century, but this type of control has been replaced with the insecticidal control due to easy availability of chemicals such as organochlorides and organophosphates. However, because of problems with insecticide resistance and greater awareness of environmental contamination, there has been renewed interest in biological methods (Service, 2000). Among the popular bio-control agents against mosquito populations are the larvivorous fish or mosquito fish: *Gambusia affinis* and *G. holbrooki* (Kumar and Hwang 2006). Fish have been widely used in public health, since as early as 1903. One of the most successful and widely used biological control agent against mosquito larvae is the mosquito fish *Gambusia affinis*. Fish other than *Gambusia* which has received the most attention as a mosquito control agent is *Poecilia reticulata*, the common guppy. All the states have been advised to upscale the use of fish as biological control method in rural areas. Guppy is a small member of the family *Poeciliidae* and like all other members of the family. The guppy, *Poecilia reticulata*, is a small ovoviviparous *Poeciliidae* fish which lives in fresh and brackish

waters of tropical regions. These fishes are self-perpetuating after its establishment and continuous to reduce mosquito larvae for long time. The cost of introducing larvivorous fish is relatively lower than that of chemical control. Use of fish is an environment friendly method of control. Larvivorous fish such as *Gambusia* and *Poecilia* prefer shallow water where mosquito larvae also breed. It is a very sturdy fish and survives in all types of water bodies. It tolerates high degree of pollution with organic matter. Reported to breed throughout the year at about four weeks interval after maturity. However breeding season will depend on climatic conditions. In warmer climate it may breed from April to November (Irianto and Austin 2002).

Guppies (*Poecilia reticulata*) are a fish species with attractive colours, bred and exported to other countries by ornamental fish companies. The species was discovered in the middle of the 19th century. The two main aspects that determine the trade and prosperity of the ornamental fish industry are health and nutrition of fishes. Success of larval rearing depends mainly on the availability of suitable food that is readily consumed. Control of mosquitoes through insecticide application has been the common practice in India for many decades. However, due to many reasons such as the increased number of spraying cycles, build up of resistance to insecticides by mosquitos (WHO, 1980), killing of non target species, non biodegradability and accumulation of insecticides in the environment, and higher cost for stronger and safer insecticides have proven that this control method is ineffective. Today, the mosquito control programmes are geared as integrated approaches

where environmental management and biological control methods being the key components.

Biological control of mosquitoes with predatory organisms in most instances is the most acceptable from environmental standpoint, and has been applied in many situations with reasonable success (WHO, 1995) and vector mosquitoes. Guppies (*Poecilia reticulata*) are a fish species with attractive colours, bred and exported to other countries by ornamental fish companies. The species was discovered in the middle of the 19<sup>th</sup> century. The two main aspects that determine the trade and prosperity of the ornamental fish industry are health and nutrition of fishes (Tovar Ramirez *et al.*, 2002; Waché *et al.*, 2006).

The aquarium fishes are rapidly gaining importance due to their immense commercial value in the export trade, the world over. Guppy fish body is short, cylindrical, compressed, head and body with scales, pre maxillaries slightly protractile; mandible prominent, teeth on jaws, plate dentate. Young males are easily distinguished from the females by their beautiful orange red and black dots all over the body and fins. The most striking feature of sexual behaviour in this species is the manner in which the males persistently pursue the females. Structurally, a viviparous fish (bearing living young) does not differ essentially from and oviparous (egg-laying) fish are present in the female's body in both cases. But, with the viviparous fish, the eggs hatch just before being laid. Guppies in spite of their tiny size, an useful citizens of whatever pond or stream they inhabit as one of their favourite foods is the "Mosquito wiggler".

Mosquito borne diseases continue to be a major problem in almost all tropical and subtropical countries. They are responsible for the transmission of the pathogens causing some of the most life – threatening and debilitating diseases of man, like malaria, yellow fever, dengue fever, chikungunya, filariasis, encephalitis, *etc.*

Environmental protection agencies have banned or placed severe restrictions on the use of many pesticides, which were formerly used in mosquito control programmes, and there are now fewer adulticides available than there have been for the last 20 years. Further, manufacturers themselves have withdrawn some insecticides due to the high cost of carrying out traditional tests now as per the government norms, in addition to the fact that the production of crop pesticides for the agricultural market is much more lucrative. Chemical pesticides and insecticides failed to give sustained

control of vector population and their over use resulted in development of resistance, elimination of natural enemies with subsequent imbalance in the ecosystem. To rationalize the use of insecticides with minimum impact on the environment it is now advocated to suppress the target population through integrated approach including the use of bio-control agents. Mosquitoes can live on almost every continent and habitat. There are 3,500 described species of mosquitoes in the world (Sathe and Tingare, 2010). The harmful effects of chemicals in mosquitoes as well as on non-target populations, and the development of resistance to these chemicals in mosquitoes along with recent resurgence of different mosquito borne diseases have prompted us to explore simple sustainable methods of mosquito control. Biological control, particularly using larvivorous fish, was important to malaria control programmes in the 20th century, particularly in urban and periurban areas as for immediate use in developed and developing countries. It has a very positive role to play in the integrated control methodologies in which both pesticides and fish or other biotic agents have their own roles.

Biological control refers to the introduction or manipulation of organisms to suppress vector populations. A wide range of organisms helps to regulate mosquito populations naturally through predation, parasitism and competition. As biological mosquito control agents, larvivorous fish (*i.e.*, those that feed on immature stages of mosquitoes) are being used extensively all over the world since the early 1900s (preDDT era). During the pre DDT era, control of mosquitoes and mosquito vectors of different mosquito borne diseases was under taken mainly by environmental management, pyrethrum space spraying, use of Paris green, oiling with petrol products and introduction of larvivorous fish. Recognizing the high larvivorous sustain in such environments (*i.e.*, they eat the prey, when introduced) for long periods.

It is important to have a sound knowledge of predator's prey selective patterns and particularly of its mosquito larval selection in the presence of alternate natural prey. In addition, the predator's adaptability to the introduced environment and overall interaction with indigenous organisms need to be considered prior to introduction prey and the feed of fish and their nutrient value is one of the most important factors in production cost and health of fish. Dengue is a disease of increasing



public health importance, with over 2.5 billion people at risk globally (Gubler 2002). Over 100 tropical countries are endemic for dengue and report increased epidemics, including more of the severe form of the disease, dengue haemorrhagic fever (DHF).

The larvivorous fish, *Poecilia reticulata* is an effective biological control agent of mosquitoes in different habitats in India. These fishes have adapted to the sewage drain conditions and are involved in regulating the populations of different species of pest..

The introduction of guppies for mosquito control is based on the assumption that they will consume sufficient numbers of water-borne larvae to substantially reduce mosquito populations, and ultimately reduce the incidence of malaria in humans.

Guppies (*Poecilia reticulata*) are a fish species with attractive colours, bred and exported to other countries by ornamental fish companies. Guppy is a small member of the family *Poeciliidae* (Female 4-6 cm long and male 2.5 – 3.5 cm long and like all other members of the family (Haskins et al., 1961). Morphological characters indicate that the genetically improved *Poecilia reticulata* has higher number of fin rays in caudal fin, dorsal fin and in pectoral fin (D7-8, C 21-26, P 8-12) than that of wild guppy (C 20-22, D7, P 7-10). Condition factor of laboratory reared wild males was found to be significantly higher ( $p < 0.05$ ) than that of the wild caught males. Males of wild guppy became sexually mature within 34-50 days after birth, while first spawning of females started 90 days after birth. The male to female ratio of the offspring was 1:3. The spawning interval was 19 - 45 days.

Guppy feeding behaviour is also of particular interest because it offers an opportunity to examine the consequences of strongly developed sexual dimorphism on the organization of foraging. In this species, females are much larger than males. The aquarium fishes are rapidly gaining importance due to their immense commercial value in the export trade, the world over. Guppy fish body is short, cylindrical, compressed, head and body with scales, pre maxillaries slightly protractile; mandible prominent, teeth on jaws, plate dentate. Young males are easily distinguished from the females by their beautiful orange red and black dots all over the body and fins. The most striking feature of sexual behaviour in this species is the manner in which the males persistently pursue the females.

Therefore for considering whole aspects present investigation made in feeding efficiency of male and female *Poecillia reticulata* in three different water sample.

## REVIEW OF LITERATURE

### International status

Natural predators are used in biological control. This type of control is an environmentally friendly preventative method to control, among others, populations of pest organisms (Becker 2006). Several organisms have proved to be an effective predators against mosquito larvae such as larvivorous fish, mosquito of the genus dragonflies and damselflies, cyclopoids copepods, nematode *Bacillus thuringiensis* H14 (WHO, 1986). Biological control of mosquitoes was very popular during the early part of the 20th century, but this type of control has been replaced with the insecticidal control due to easy availability of chemicals such as organochlorines and organophosphates. However, because of problems with insecticide resistance and greater awareness of environmental contamination, there has been renewed interest in biological methods (Service, 2000). Among the popular biocontrol agents mosquito populations are the larvivorous fish, or mosquito fish: *Gambusia affinis* and *G. holbrooki* (Kumar and Hwang 2006). This species however is ineffective for the control of mosquitoes in what is termed as “container habitats”, that is mosquito breeding places such as phytotelmata, tree holes, bamboo nodes and discarded containers (Kumar and Hwang, 2006).

America and Asia are commonly found in ornamental ponds in Composita and elsewhere (Rozendaal 1997). Guppies grow to about 6cm in length, and females produce 40-50 offspring following a one month generation period.

Biotic interactions such as competition and predation have been reported to be capable of regulating the number of mosquito populations by reducing the number of larvae that survive through larval development and by increasing the larval and pupal duration times (Knight *et al.*, 2004).

### NATIONAL STATUS

In India review of the bio-environmental methods for malaria control with special reference to the use of larvivorous fishes and composite fish culture in central Gujarat, India were reported by Rajni Kant *et al.*, 2013. Until recently, the ecological role of environmental managers has been more concentrated on preventing damage

from pollution rather than proposing sustainable solutions to different global and local problems faced by human societies.

Vector-borne diseases such as malaria, filariasis, Japanese encephalitis, dengue and many other arboviral diseases are emerging and resurging as serious public health problems and cause a huge medical, financial burden and social disarray in developing and under-developed countries (Sathe, 2011). This is due to drastic demographic changes, poor sanitary condition as a consequence of rapid unplanned urbanization, change in agricultural practices, failure to maintain previously effective control measures, absolute control strategies, progressive spread of drug resistance and lack of identification of epidemic risk indicators (Das and Amalraj, 1997). Chemical pesticides and insecticides failed to give sustained control of vector population and their overuse resulted in development of resistance, elimination of natural enemies with subsequent imbalance in the ecosystem. To rationalize the use of insecticides with minimum impact on the environment it is now advocated to suppress the target population through integrated approach including the use of bio-control agents. Mosquitoes can live on almost every continent and habitat. There are 3,500 described species of mosquitoes in the world (Sathe and Tingare, 2010).

One of the multiple possibilities of applying ecological theories for human welfare is the use of our knowledge about the effects and mechanisms of predation and competition within various kinds of permanent and temporary aquatic habitats. By manipulating particular trophic levels, desired changes can be achieved in a system (He *et al.*, 1994). Biological control means the use of different kinds of living things, and/or their derivatives to eliminate pest populations. Many biological control agents disperse by themselves enabling them to spread and build up viable populations (Caltagirone, 1981; Bellows, 2001; Headrick and Goeden, 2001).

During the pre-DDT era, control of mosquitoes and mosquito vectors of disease was undertaken mainly by environmental management, pyrethrum space spraying, use of Paris green, oiling with petrol products and larvivorous fish. Recognizing the high larvivorous potential of *Gambusia affinis*, this fish species was purposely introduced from its native Texas (southern USA) to the Hawaiian Islands in 1905. In 1921 it was introduced in Spain, then from there into Italy during the 1920s and later to 60 other countries. Beginning in 1908,

another larvivorous fish, *Poecilia reticulata* (guppy), a native of South America, was introduced for malaria control into British India and many other countries. The introduction of the use of DDT in indoor residual spraying for malaria control around the mid-1940s led to the gradual decline in the use of environmental management and biological control methods except in a few programmes in Russia

Biological control is defined as the action of predators, parasites (parasitoids) or pathogens in maintaining the density of another organism at a lower average than would occur in their absence. Biological control of vectors is an essential and effective means for controlling transmission of several mosquito-borne diseases such as malaria, filariasis, JE, dengue fever, etc. Among the numerous mosquito predators, fish have been used since the early 1900's for mosquito control and are the most commonly used biological control agent for the vectors of malaria in many countries. Use of larvivorous fishes in the field of mosquito control is well documented. The present study was conducted to assess the bio-control potential of four exotic fish is *Poecilia reticulata* which are well adapted in the native environmental conditions of India and are widely cultured in most of the fresh water biomes throughout the country. Our investigation was also aimed at analyzing the efficacy of co-existing bio agents of Anopheline mosquitoes, the relationship of predatory efficacy with reference to prey density and search area, and the efficacy of the four fish species under field conditions.

In India such type of work were done such as Vulnerability of the mosquito larvae to the guppies (*Poecilia reticulata*) in the presence of alternative preys were studied by (Manna *et al.*, 2008). Bio-control of mosquito larvae through the black molly, *Poeciliaspheps* by (Sumithra *et al.*, 2014). Use of larvivorous fish like *Gambusia affinis* and *Poecilia reticulata* different mosquito breeding habitats (Ghosh *et al.*, 2011; Haqetal., 1993; Fletcher *et al.*, 1992) in mosquito control has well established. However, reports on operational level scaling up or large scale use of fishes are scanty (Ghosh *et al.*, 2005; Haq *et al.*, 2003). They may prove to be a good option to insecticides in vector control where breeding habitats of malaria vectors are confined. Black mollies are voracious, feeding on mosquito larvae in breeding sites like drains and tanks. Bloodworms, micro worms, fruit flies, Daphnia and chopped up earthworm are other examples of



suitable food for molly. Mosquitoes are known vectors for transmission of disease throughout the world, including the pathogens responsible for encephalitis, equine infectious anaemia, malaria, yellow fever, dengue and filariasis (Pant *et al.*, 1981; Haas and Pal 1984; Lichtenberg and Getz 1985). The present work describes the predatory efficacy of *Poecilia reticulata* against mosquito larvae under laboratory conditions using multiple alternative prey and habitats conditions.

### **MATERIAL AND METHOD**

Feeding efficiency of male and female fish were used in the present study and analysed in the laboratory condition and for this study we used predator fish, is in the form of *Poecilia reticulata* as a biological control agent for the mosquito larvae. The predator fish were collected from the malaria department of Akola and mosquito larvae of *Anopheles*, *Culex* and *Ades* were collected from water pots from college campus and nearby stagnant water-body located in and around Akola city. After collection of prey and predator they were separately kept in the stocking aquarium in the laboratory for acclimatization. After acclimatization experimental sets were prepared for analysing feeding efficiency of male and female *Poecilia reticulata* separately in the three different water sample. For this study six aquarium were used at a time and this aquarium get filled with up to the 20 liter mark with pond water, drainage water and tap water separately and in each aquarium we kept male and female fish and released the mosquito larvae for checking feeding efficiency in the different water sample of male and female fish for 24 hours. Such types of experiment were conducted in six replica in the laboratory.

At a time in each aquarium 20 number of mosquito larvae were introduced against predator fish. Both male and female predator fish introduced separately in each experimental sets. The time of introduction of the predator and prey for each experiment that of each replicate was recorded, and the mosquito larvae, alive or dead, remained after consumption by the guppy fish were counted after 24 hours. The consumption rate of each guppy per day was calculated accordingly from each of the replicate i.e. total number of mosquito larvae introduced at the beginning of experiment minus total number of mosquito larvae consumed per day per predator fish.

Three types of water were used namely, drain water, tap water, pond water were used separately for each experiment.

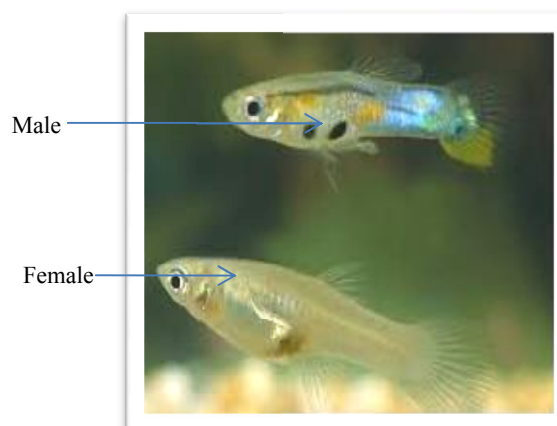
### **COLLECTION AND MAINTENANCE OF PREDATORS**

Guppies (*Poecilia reticulata*) were collected from the local Malaria department of Akola district of the state of Maharashtra, India. The fish were 3.1-4 cm in length. They were acclimatized for seven days in laboratory in holding aquarium containing de-chlorinated water.

### **MORPHOLOGY OF POECILIA RETICULATA**

The guppy (*Poecilia reticulata*), also known as million fish or rainbow fish, is one of the world most widely distributed tropical fish, and one of the most popular fresh water aquarium fish species. It is a member of poeciliidae family and, like almost all American members of the family, is live bearing. Guppies whose natural range is in Northeast South America, were introduced to many habitats and are now found all over the world. They are highly adaptable and thrive in many different environmental and ecological conditions. Male guppies, which are smaller than the females, have ornamental caudal dorsal fins, while females are duller in colour. Wild guppies generally feed on a variety of food sources, including benthicalgae and aquatic insect larvae. Guppies are used as a model organism in a field of ecology, evolution and behavioural studies.

Kingdom : Animalia  
Phylum : Chordata  
Class : Actinopterygii  
Order : Cyprinodontiformes  
Family : Poeciliidae  
Genus : *Poecilia*  
Species : *reticulata*



Sexual polymorphism exhibited in the guppy

(above male, below female)

Guppies exhibit sexual dimorphism. While wild-type females are grey in body colour, males have splashes, spots, or strips that can be any of a wide variety of colours. The size of guppies vary, but males are typically 1.5-3.5cm long, while females are 3-6cm long.

### OBSERVATION AND RESULT

Feeding efficiency of female *Poecilia reticulata* in tap water 334, pond water 397, and drain water 358 were observed in 30 days whereas male *Poecilia reticulata* consumed 147 in tap water pond water 215 and in drain water 180 respectively (table 1). Statistical analysis study shows that average number of larvae consumed by both male and female fish. Average number mosquito larvae consumed by a female guppy is 11.1 in tap water, 13.2 in pond water and 11.9 in drain water. This study shows that feeding efficiency of female fish is more in pond water than that of tap and drain water similarly male also show similar type of feeding efficiency in the three different water sample (table 1) This experimental analysis shows that female fish consume more no.

of mosquito larvae than that of the male fish. Similar types of result were also reported by (Saleeza SNR, Norma- Rashid Y, Sofian –Azirun 2011). During this study we also recorded some physico-chemical parameter of collected water sample. In this case we noted pH and temperature of water sample. pH of water sample were found in tap water 9.11; in pond water 8.66 and in drain water 7.8 were recorded during the study period. Temperature of the water sample were found to be 23°C in tap water; 20°C in pond water and 24°C in drain water were recorded.

In the present study feeding efficiency of male and female *Poecilia reticulata* were made in the three different water sample collected from pond, drainage and tap water. Six experiment set were designed for this we use glass aquarium. This study was made from 1 June to 30 March. Predation potentiality of a biological control agent, the guppy fish (*Poecilia reticulata*) was studied over the mosquitos larvae under laboratory conditions.

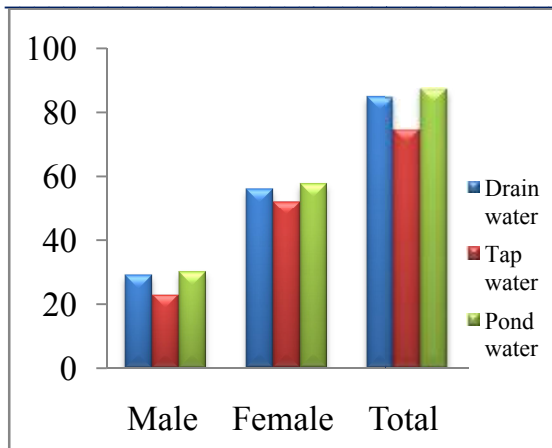
Table 1: Feeding efficiency of *Poecilia reticulata* in different water sample in laboratory condition.

<i>Poecilia reticulata</i>	Type of water	No. of days	Total no. of larvae released	Total no. of larvae consumed	Average no. of larvae consume
Female	Tap	30	600	334	11.1
	Pond	30	600	397	13.2
	Drain	30	600	358	11.9
Total			1800	1089	12.6
Male	Tap	30	600	147	4.9
	Pond	30	600	215	7.1
	Drain	30	600	180	6.3
Total			1800	542	6.1
Gr and total			3600	1631	54.3

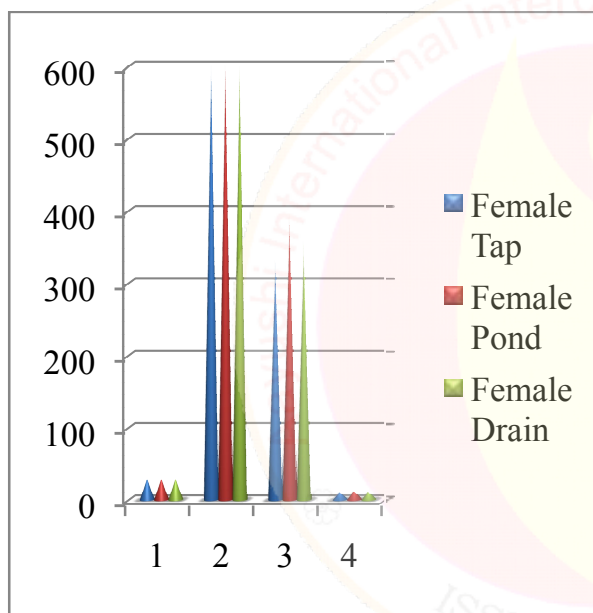
Observation on predation activities of guppies (*Poecilia reticulata*) on the larvae of three species of mosquito, namely *Anopheles*, *Aedes aegypti*, and *Culex quinquefasciatus* was carried out under laboratory conditions. Male and female guppies were used as predators for predation experiments on the 4th instars of mosquito larvae. The daily feeding rates comparing male and female guppies

on mosquito larvae were different; the female guppies consumed more mosquito larvae than male guppies did.

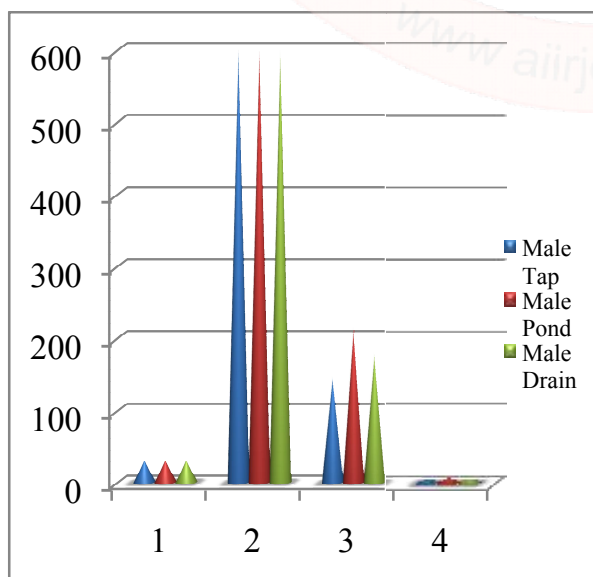
#### Graph No 1: Average no. of larvae consumed per day per fish



Graph No 2 : Average no. of larvae consumed by female in different water type per day per fish



Graph No. 3 : Average no. of larvae consumed by male in different water type per day per fish



It has been observed that a single predator fish (*Poecilia reticulata*) consumed an average of 54.3 mosquito larvae per day (24 hours). A female guppy fish consumed an average of about 12.6 per day per fish mosquito larvae. Male guppy fish consumed an average of about 6.1. From above number it clears that female guppy is consume more mosquito larvae than male. This indicate that the female guppy is more active than the male in feeding mosquito larvae.

### DISCUSSION

The efficient larvivorous fish, guppy, being viviparous and capable of surviving and increasing its population in polluted water, could be a better candidate for control of mosquitoes. In this study, it was observed that female guppies (*Poecilia reticulata*) ate mosquito larvae more than male guppies did. The advantage of using fish as a biological control agent is that fish has a good adaptation to its new environment.

The study also suggests that the guppy fish, which is available in drains and pools in Akola city, could be manipulated for use as a promising and sustainable biological control agent against the mosquito vectors of filariasis and other mosquito-borne diseases, of course, after a careful field trial. A number studies have indicate that Guppies of the *Poecilia reticulata* species are good predators, as they can control mosquito larvae population (Manna *et al.*, 2008; Seng *et al.*, 2008; Anogwih and Makanjuola, 2010; Gosh *et al.*, 2011).

### SUMMARY

The feeding efficiency of male and female *Poecilia reticulata* were analyzed in laboratory condition. For this purpose six set of aquarium of three different water sample are used in which male and female *Poecilia reticulata* introduced separately in each aquarium. From this study we were found that the feeding efficiency of female is more than that of the male. In specially in pond water the feeding efficiency of male and female *Poecilia reticulata* is increase in pond water than that of the drain and tap water.

### CONCLUSION

On the basis of above observation and result it is concludes that feeding efficiency of female *Poecilia reticulata* is more than male in every water sample. In specially pond water the feeding efficiency of female is more. So release more *Poecilia reticulata* in pond water around of urban and rural areas for control of mosquito population.



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**ABSTRACTS SECTION**

**DIVERSITY AND ABUNDANCE OF DRAGONFLIES AND DAMSELFLIES (ORDER - ODONATA) OF BHIVAPUR RANGE IN POHARA – MALKHED RESERVE FOREST, MAHARASHTRA (INDIA)**

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**ABSTRACT**

The present work is the result of survey on Dragonflies and Damselflies (Order - Odonata) fauna of Bhivapur Range of Pohara – Malkhed Reserve forest. This study was carried out for one year from June 2012 to May 2013. Total 39 Species of Dragonflies and Damselflies belonging to six families were recorded (Lebellulidae, Coenagrionidae, Ashnidae, Gomphidae, Platycenemididae and Lestidae). Among the families, lebellulidae is the most dominant and diverse one having 15 genera and 20 species. Beside this, in rainy season the diversity and abundance is somewhat higher as compared to the other seasons. The present area is very much diverse in Dragonflies and Damselflies with their higher abundance in all the seasons of the year with somewhat variations.

**Keywords:** Diversity, dragonflies and damselflies, Bhivapur range, abundance

**INTRODUCTION**

Dragonflies and Damselflies are the best flier insects of the world. They are the beautiful insect which generally flies just close to the water sources. Their color, size and their diurnal and often prominent behavior make them a popular group for both professional and amateur entomologists. Eight super families, 29 families and some 58 sub-families of dragonflies for approximately 600 genera and 6000 named species have so far been described all over the world (Silsby, 2001).

India is very much diverse with 500 named species of Dragonflies and Damselflies (Subramanian, 2005). Dragonflies and Damselflies are predaceous, amphibiotic and hemi-metabolous insects, which occurs in all kinds of freshwater habitats either permanent or temporary. Dragonflies and Damselflies show two totally different life styles, the egg and larval stages are

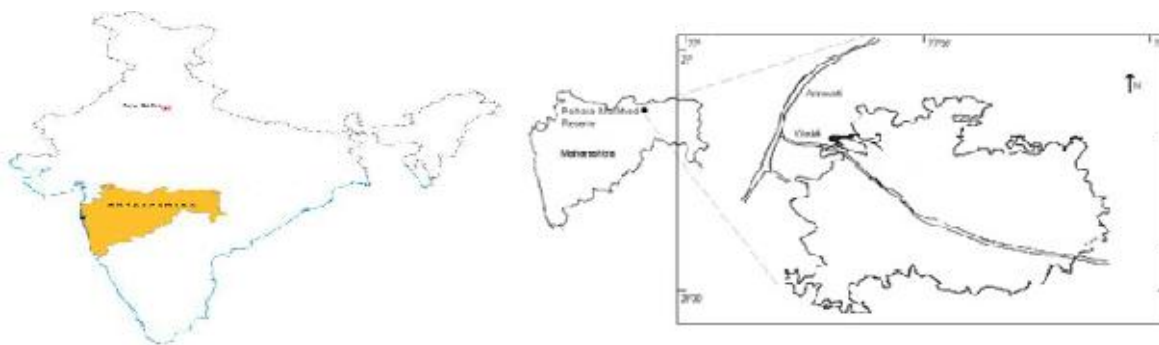
aquatic while the adults are terrestrial (Silsby, 2001).

The present Reserve Forest is much neglected for such type of study. Hence, the objective of this study was to conduct a survey on diversity of Dragonflies and Damselflies species with their seasonal abundance.

**MATERIAL AND METHOD**

**Study area:**

Bhivapur range is the part of Pohara – Malkhed Reserve Forest, Amravati, which is the one end of Reserve. Pohara – Malkhed Reserve Forest lies at 20°57'N and 77°57'E, cover the total area of 80 km<sup>2</sup> (Fig 1b). Many workers recorded various migratory Birds from the present forest. The vegetation of Reserve forest consist dry-deciduous, evergreen semi-evergreen, grassland and teak, salai, Khair and Palas plantation (Wadatkar and Kasambe, 2002).



**Fig.1a: India showing Maharashtra State Fig.1b: Study Area: Pohara – Malkhed Reserve Forest, Maharashtra**

**Observations and Photography:**

The presented work was done for one year from June 2012 to May 2013. Specimens of Dragonflies and damselflies were observed in field with careful note on their habitats. Most specimens were identified in the field by visual observations. The photography was done by using digital camera made by Nikon model no. L810. The specimens of unidentified species were collected by standard insect collecting net and then further their identifications were carried out in laboratory using Binocular microscope and by using the available literature the British Fauna of India by Fraser (1933, 1934 and 1936).

**Methodology:**

Different standard transects of 500 meter long were drawn in the study area. Specimens were observed and counted by walking along transects. The width of each transect was 5 meter. For minimize the error, the study area was divided into five different parts and the data from all parts were summarized to calculate the mean.

**Data analysis:**

Computer generated program Biodiversity Pro was used to calculate the Diversity indices like Shannon H', Shannon Hmax, Shannon J',

Simpsons Diversity (D), Simpsons Diversity (1/D), Richness.

**RESULTS AND DISCUSSION**

The present investigation for one year from June 2012 to May 2013 results that 39 species of Dragonflies and Damselflies belonging to 25 genera and six families. Observed families are Lebellulidae, Coenagrionidae, Ashnidae, Gomphidae, Platycenemididae and Lestidae. Results obtained cleared that the most dominant family is the Lebellulidae which contains 15 genera and 20 species. Also it is cleared that the list diverse families are Ashnidae, Gomphidae, Platycenemididae and Lestidae which contains two, one, two and one species respectively. Manwar et al, 2012 also perform similar investigation from Chattri lake region of same reserve forest and recorded 22 species.

Table 1 shows the taxonomic nomenclature, families and Abundance of each species from each season from the study area during the study period in Pohara range in Pohara – Malkhed Reserve Forest. Table 2 below listed the calculated Species richness and Species diversity.

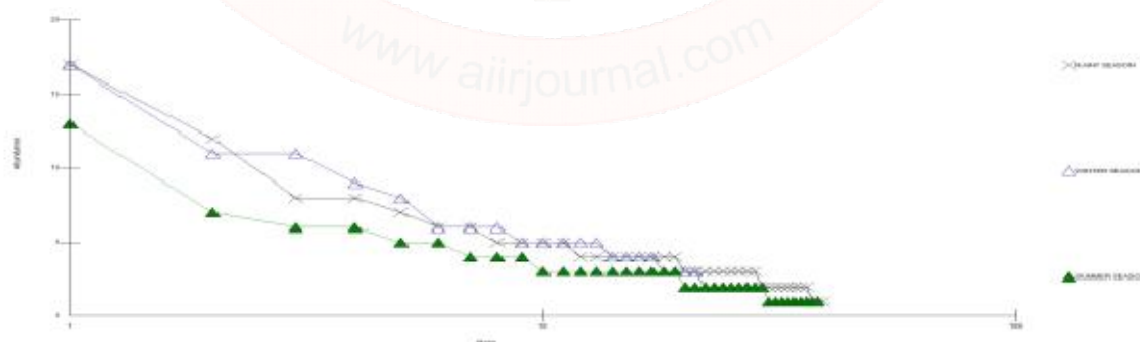
**Table1: Complete list of dragonflies and damselflies species found in Bhivapur range in Pohara – Malkhed Reserve Forest, Maharashtra with scientific names with family and season wise abundance of each species.**

Family	NAME OF SPECIES	RAINY	WINTER	SUMMER
Lebellulidae	<i>Acisoma panorpoides</i> (Rambur, 1842)	04	05	03
	<i>Aethriamanta brevipennis</i> (Rambur, 1842)	03	01	00
	<i>Brachydiplax sobrina</i> (Rambur, 1842)	03	01	01
	<i>Brachythemis contaminata</i> (Fabricius, 1793)	0	03	02
	<i>Crocothemis servilia</i> (Drury, 1770)	02	05	03
	<i>Diplacodes lefebvreii</i> (Rambur, 1842)	03	03	02
	<i>Orthetrum chrysis</i> (Selys, 1891)	04	05	02
	<i>Orthetrum Sabina</i> (Drury, 1770)	07	08	04
	<i>Orthetrum taeniolatum</i> (Schneider, 1845)	04	01	03
	<i>Pantala flavescens</i> (Fabricius, 1798)	14	12	04
	<i>Potamarcha congener</i> (Rambur, 1842)	01	00	02
	<i>Rhodothemis rufa</i> (Rambur, 1842)	01	00	00
	<i>Rhyothemis variegata</i> (Linnaeus, 1763)	03	00	04
	<i>Tholymis tillarga</i> (Fabricius, 1798)	01	02	01
	<i>Tramea limbata</i> (Desjardins, 1832)	02	02	02
	<i>Tramea virginia</i> (Rambur, 1842)	02	02	01
	<i>Trithemis aurora</i> (Burmeister, 1839)	02	02	03
	<i>Trithemis festiva</i> (Rambur, 1842)	02	02	02

	<i>Trithemis pallidinervis</i> (Kirby , 1889)	08	06	05
	<i>Urothemis signata</i> (Rambur, 1842)	03	01	01
<b>Gomphidae</b>	<i>Ictinogomphus rapax</i> (Rambur, 1842)	05	05	05
<b>Ashnidae</b>	<i>Anax immaculifrons</i> (Rambur, 1842)	02	02	02
	<i>Anax guttatus</i> (Burmeister, 1839)	04	03	03
<b>Coenagrionidae</b>	<i>Aciagrion pallidum</i> Selys, 1891	02	02	01
	<i>Agriocnemis femina</i> (Brauer, 1868)	01	00	02
	<i>Agriocnemis pieris</i> (Laidlaw, 1919)	01	04	03
	<i>Agriocnemis pygmaea</i> (Rambur, 1842)	04	18	08
	<i>Agriocnemis splendidissima</i> (Laidlaw, 1919)	02	02	01
	<i>Agriocnemis species</i>	04	01	01
	<i>Ceriagrion coromandelianum</i> (Fabricius, 1798)	12	11	11
	<i>Ischnura aurora</i> (Brauer, 1865)	03	03	03
	<i>Ischnura senegalensis</i> (Rambur, 1842)	05	11	06
	<i>Pseudagrion decorum</i> Rambur, 1842	04	00	01
	<i>Pseudagrion microcephalum</i> (Rambur,1842)	05	05	05
	<i>Pseudagrion rubriceps</i> (Selys, 1876)	04	04	02
	<i>Rhodischnura nursei</i> (Morton, 1907)	04	06	03
<b>Lestidae</b>	<i>Lestes elatus</i> (Selys,1862)	02	02	01
<b>Platycenemididae</b>	<i>Copera marginipes</i> (Rambur, 1842))	03	05	02
	<i>Copera vittata</i> (Selys,1863)	04	01	01

**Table 2: Diversity indices for different seasons of the study period in Bhivapur range in Pohara – Malkhed Reserve Forest, Maharashtra.**

Index	RAINY SEASON	WINTER SEASON	SUMMER SEASON
Shannon H'	1.503	1.426	1.484
Shannon Hmax	1.591	1.544	1.58
Shannon J'	0.944	0.924	0.939
Simpsons Diversity (D)	0.033	0.041	0.033
Simpsons Diversity (1/D)	30.187	24.557	30.57
Species richness	37	39	39



**Fig 2: Species rank Abundance plot for different seasons of the study period.**

Simpsons Diversity (D), Simpsons Diversity (1/D), Shannon H', Shannon H<sub>max</sub>, Shannon J' and Richness were calculated by comparing the data obtained from different seasons of the study period. In rainy season there is highest value of

Shannon H' which is 1.503 and the list is seen in the April which is 1.426. The almost same orders are also seen in Shannon J', Simpsons Diversity, Richness indices (table 2). The values of Simpsons Diversity index (1/D) and Shannon J, which is



very high in all seasons of the study period, indicate that the present study area contain high diversity, smaller values of Simpson index (D) also indicate the same result. According to the abundance of each species they were ranked which is shown in the Fig 2.

#### **CONCLUSION**

From the present survey we conclude that the Bhivapur range of Pohara - Malkhed Reserve forest is much rich in Dragonflies and Damselflies diversity. This rich diversity of Pohara range is may be due to large amount of water availability, floral diversity and less encroachment of human

activities. But some protection plans have to make in the forest to protect and build up the diversity.

#### **ACKNOWLEDGEMENT:**

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# Agro waste dyes- A sustainable source of natural dyes for textiles

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**Abstract:** Renewed international interest has arisen in natural dyes due to increased awareness of the environment and health hazards associated with the synthesis, processing and use of synthetic dyes. Textile processing industry is one of the major environmental polluters as the effluent from these industries contains a heavy load of chemicals including dyes used during textile processing. There are two main ways to limit the environmental impact of textile processing. One is to construct sufficiently large and highly effective effluent treatment plants, and the other way is to make use of dyes and chemicals that are environment friendly. Natural dyes, when used by themselves have many limitations of fastness and brilliancy of shade. However, when used along with metallic mordants they produce bright and fast colors. Therefore, instead of using unsustainable technology for producing colors one can use mild chemistry to achieve almost similar results. The rich biodiversity of our country has provided us plenty of raw materials, yet sustainable linkage must be developed between cultivation, collection and their use. Natural dyes can produce special aesthetic qualities, which combined with the ethical significance of a product that is environmentally friendly, gives added value to textile production as craftwork and as an industry.

Key words: Biodiversity, mordants, effluent, natural dyes.

## Introduction:

There are two types of agricultural crop residues. Field residues are materials left in an agricultural field or orchard after the crop has been harvested. These residues include stubble (stems), leaves, and seed pods. The residue can be ploughed directly into the ground, or burned first. Good management of field residues can increase efficiency of irrigation and control of erosion. Process residues are material left after the crop is processed into a usable resource. These residues include husks, seeds, molasses and roots. They can be used as animal fodder and soil amendment, fertilizers and in manufacturing. In India, the crop residues are traditionally utilized as animal feed as such or by supplementing with some additives. However, crop residues, being unpalatable and low in digestibility, cannot form a sole ration for livestock. Crop



residues are low-density fibrous materials, low in nitrogen, soluble carbohydrates, Minerals and vitamins with varying amounts of microbial breakdown. To meet the nutritional requirements of animals, and supplementing with green fodders (leguminous/non-leguminous) and legume (sun hemp, horse gram, cowpea, gram) straws.

Natural dyes can be broken down into two categories: substantive and adjective. Substantive, or direct dyes, become chemically fixed to the fiber without the aid of any other chemicals or additives, such as indigo or certain lichens. Adjective dyes, or mordant dyes, require some sort of substance, (usually a metal salt) to prevent the color from washing or light-bleaching out. Most natural dyes are adjective dyes, and do require the application of a mordant (the metal salt) solution to the fibers at some point in the dyeing process. Aluminum and iron salts were the most common traditional mordants, with copper, tin and chrome coming into use much later. In rural areas where these metals were not widely available, plants were also used as mordants, especially those that have a natural ability to extract such minerals from the earth, such as club moss.

Agricultural wastes are basically unusable substances which may be either liquid or solid produced as result of cultivation processes such as fertilizers, pesticides, crop residues and animal waste. Agricultural waste management is part, of the ecological cycle in which everything is cycled and recycled such that an interdependent relationship is maintained in the eco-system. By waste management, all the plant wastes are placed at the right place and right time for the best utilization in order to convert into useful product and pollution control. Globally, 140 billion metric tons biomass is generated every year from agriculture.

### **Objectives:**

1. To extract natural dye from agro waste.
2. To dye cotton fabric using two mordants.
3. To observe the color fastness of the dyed samples.

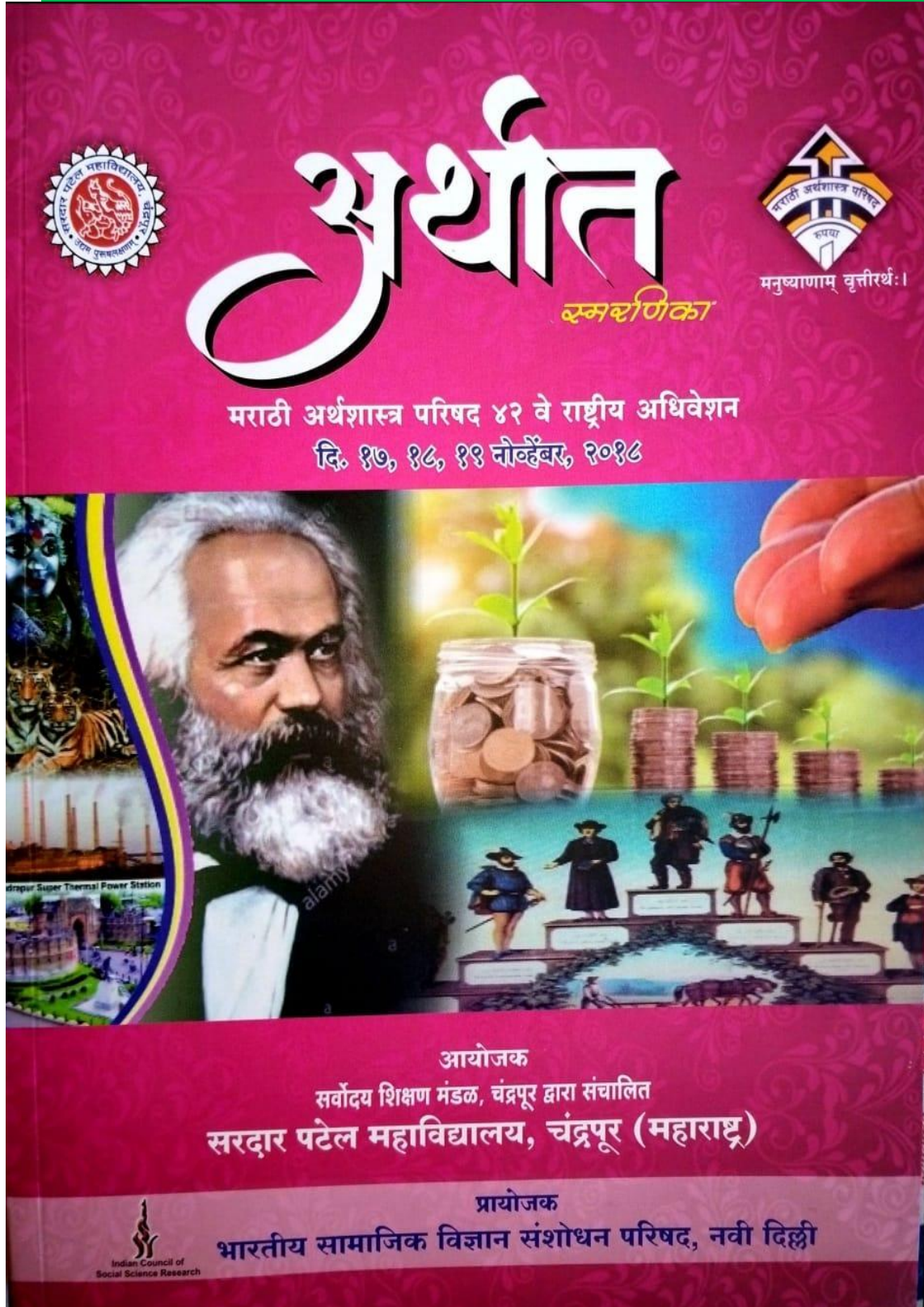
**Methodology:** Experimental and Exploratory Research Design was used for the study

### **1] Collection of Material:**

- i. Skin of Cajanus cajan (Pigeon pea-Tur) it is a waste product after harvesting. It is a kharif crop of a duration of 8 months. The fruit come after three months, which develops completely in four months. It requires 15 days for drying on the plant itself. While harvesting the skin and pie is separated. Usually the skin is a animal feed after drying.



E) Publications other than journal articles (books, chapters in books):



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## कार्ल मार्क्स आणि भौतिकवाद

डॉ. रामेश्वर मा. भिसे

प्राचार्य, श्री शिवाजी महाविद्यालय अकोला

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### गोष्टवारा

कार्ल हेनरिख मार्क्स जर्मन दर्शनिक, जर्मण अर्थशास्त्री, तत्वज्ञ, इतिहासकार, राजनीतिक सिद्धांतकार, समाजशास्त्री, पत्रकार आणि वैज्ञानिक समाजवादाचे प्रणेते. मार्क्सने तत्कालीन राजकीय लढ्यांमध्ये भाग घेतला. मार्क्सने प्रत्यक्ष वस्तुस्थितीचा अभ्यास करून आपली शास्त्रीय समाजवादी कल्पना मांडली आहे. कार्ल मार्क्सच्या समाजवादाला शास्त्रीय समाजवाद म्हणतात. कार्ल मार्क्सचा जन्म ५ मे १८१८ मध्ये जर्मनीमध्ये झाला. त्याचे शिक्षण बॉन आणि बर्लिन विद्यापीठात झाले. कायदेशास्त्र, इतिहास आणि तत्वज्ञानाचा अभ्यास त्यांनी केला बर्लिन विद्यापीठात असतांना हेगेलच्या तत्वज्ञानाचा प्रभाव त्याच्यावर पडला. १८४१ मध्ये त्यांनी पी.एच.डी प्राप्त केली. मार्क्सने आपला जगप्रसिद्धी ग्रंथ 'दास कॅपिटल' पहीला खंड इ.स. १८६० मध्ये प्रसिद्ध केला. इंग्लंडमध्ये औद्योगिक क्रांती आणि भांडवलशाहीचा विकास झाला त्याच इंग्लंडमध्ये राहून भांडवल शाही विरोधी लेखन करण्याचे कार्य मार्क्सने केले. मार्क्सचे विचार शिक्षण क्षेत्र, राजकीय क्षेत्र, कामगार लढायांमध्ये अजुनही आदर्श मानले जातात. कार्ल मार्क्सच्या शास्त्रीय समाजवादात द्वंद्वात्मक भौतिकवाद, ऐतिहासिक भौतिकवाद, अतिरिक्त मुल्य सिद्धांत, वर्गसंघर्ष, राज्यविहीन व वर्गविहीन समाज या साऱ्यांचा समावेश होतो.

उत्पादन व्यवहार, उत्पादनशक्ती, उत्पादनसंबंध, त्यातील अंतर्विरोध, खाजगी मालकी, शासनसंस्था, विचारसरणी, क्रांती इत्यादी मूलभूत संकल्पनांचा विचार करून

मार्क्सने मानवाचा उत्पादन व पुनरुत्पादनाचा व्यवहार, त्यातील उत्पादनशक्ती व उत्पादन संबंध यांचे द्वंद्वात्मक नाते यांच्या आधारे इतिहासाचा तसेच त्याच्या काळातील वर्तमानाचा अभ्यास केला. या अभ्यासातूनच त्यांनी निष्कर्ष काढला की मानवी इतिहास हा वर्गलढ्याचा इतिहास आहे. व उत्पादनसाधनांवरही भांडवली खाजगी मालकी हे आजच्या सर्व प्रश्नांचे मूळ आहे.

समाजाच्या विविध अवस्थांना नावे वेगवेगळी असली, तरी समाजातील विविध वर्गांचे असणारे संबंधाचे स्वरूप मात्र तेच आहे. एक शोषक वर्ग आणि दुसरा शोषित वर्ग, मार्क्सच्या मते, आज जी भांडवलशाही अस्तित्वात आहे., तीदेखील याच अवस्थामधून जाईल. भांडवलशाही नष्ट होऊन जी नवीन समाजव्यवस्था निर्माण होईल ती वर्ग. मार्क्सच्या मते, आज जी भांडवलशाही अस्तित्वात आहे, तीदेखील याच अवस्थामधून जाईल. भांडवलशाही नष्ट होऊन जी नवीन समाजव्यवस्था निर्माण होईल ती वर्गविरहित समाजरचना असेल. वर्गविरहित समाजव्यवस्थेत उत्पादन सामाजिक गरजेनुसार केले जाते, उत्पादन साधने समाजाच्या मालकीची असतात. कोणीही मालक असणार नाही. त्यामुळे शोषणाचा प्रश्नच निर्माण होणार नाही. वर्गविरहित समाज अवस्थेनंतर विकासाची नंतरची अवस्था म्हणजे साम्यवादाची अवस्था म्हणजे साम्यवादाची अवस्था होय. या अवस्थेत प्रत्येकाला गरजेनुसार काम आणि क्षमतेनुसार वेतन मिळेल.

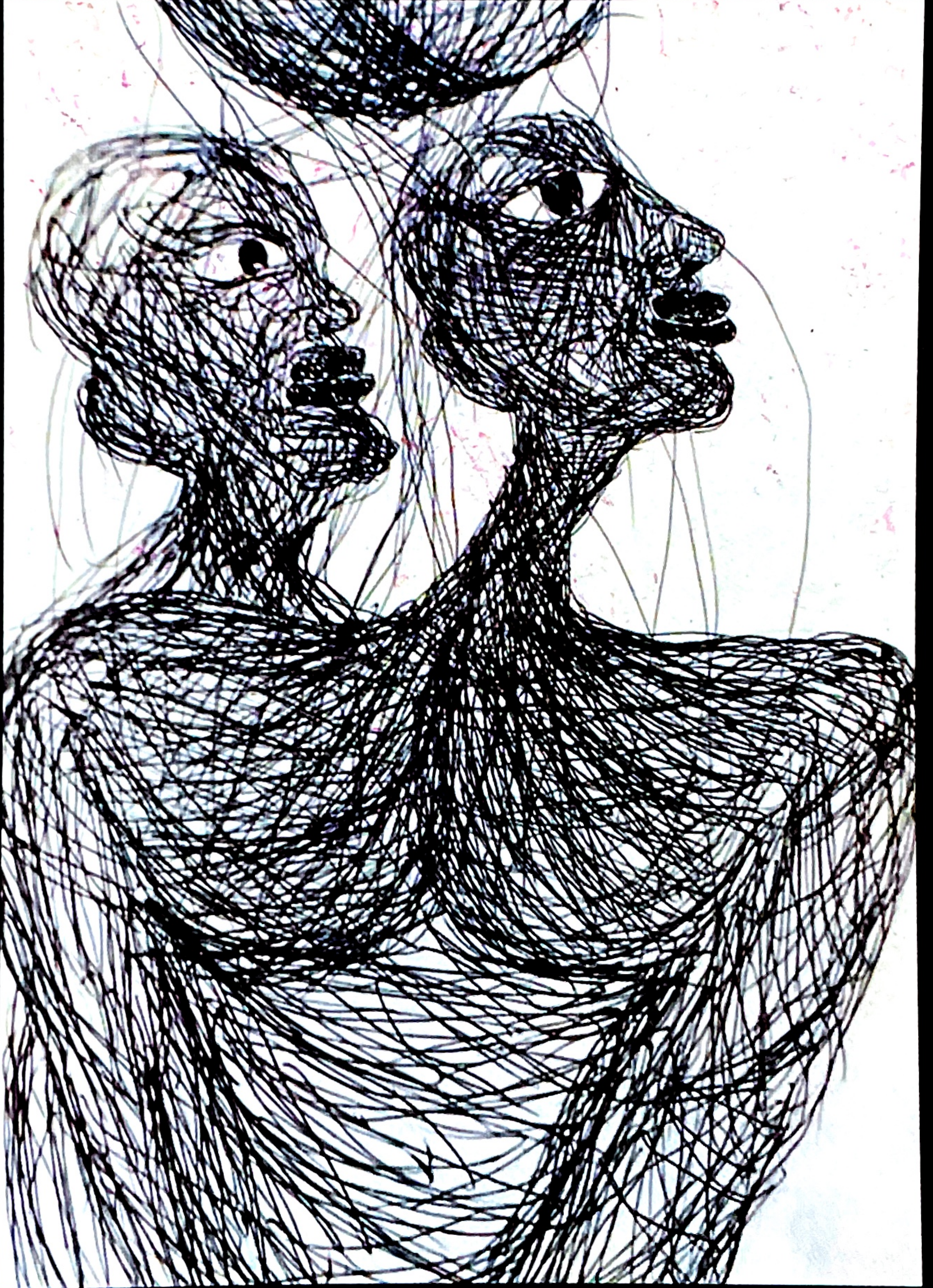
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नवी  
जावळस

आणि इतर एकांकिका

प्रा.संजय पोहरे



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आणि इतर एकांकिका  
प्रा. संजय सुखदेव पोहरे**

**सेवा प्रकाशन, अमरावती**



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आणि इतर एकांकिका  
प्रा. संजय सुखदेव पोहरे

**Nave Gaonkus**  
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बाबासाहेब आंबेडकरांनी दिलेली सामाजिक  
आणि त्या सामाजिक जाणिवेतून निर्माण  
ली मानवतावादी जीवनदृष्टी,  
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**भारतीय महिला**

*Indian Women in Historical Perspective*



डॉ. सूर्यकांत कापशीकर





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## प्रभा अत्रे का संगीत कला में योगदान

डॉ. सोपान सिताबराव वतारे  
संगित विभाग, श्री शिवाजी महा. अकोट

इतिहास साक्षी है उन गतिविधीयोका जिनके महान योगदान से स्वतंत्रता की लड़ाई के साथ - साथ संगीत - साहित्य, राजनिती खेल आदी क्षेत्र में महिलाओं ने अपना नाम रौशन किया। ऐसीही एक महान महिला संगीत विदूशी डॉ. प्रभा अत्रे का संगीत कला में योगदान भारतीय संगीत में विभिन्न कलाकारों ने समय - समय पर अपना महत्वपूर्ण योगदान प्रदान किया है। इसी क्रम में उत्तर भारतीय शास्त्रीय संगीत कलाकार पद्भूषण डॉ. प्रभा अत्रे अन्वेशक एवं सिध्द कलाकारों की परम्परा में अपना विशिष्ट स्थान रखती है। आपके कार्यक्रमों ने लोकप्रियता के अनेक कीर्तिमान स्थापित किए हैं। यह प्रतिभाशाली विविधांगी कलाकार गत लगभग ६० वर्षों से शास्त्रीय संगीत के मंच पर शोभायमान रही है और अपनी गान प्रवणता से जन - जन के हृदय को जीतती तथा अल्हादित करती चली आ रही है।

वर्तमान में किराना घराने की आप वरिष्ठ महिला गायिका हैं जो इस घराने का प्रतिनिधत्व कर रही हैं। इस संदर्भ में देश की लब्ध प्रतिष्ठित गायिका डॉ. प्रभा अत्रे सामाजिक सांगीतिक कार्यकर्ता एवं प्रचारक के रूप में निरन्तर सक्रिय रही हैं। आपने कलाकार के रूप में ही नहीं वरन् एक आयोजिका, लेखिका, गुरु एवं सामाजिक कार्यकर्ता के विविध रूपों में भी संगीत के विकासात्मक दृष्टिकोण को ध्यान में रखते हुए, देश में ही नहीं वरन् विदेशों में भी संगीत की लोकप्रियता बढ़ाकर सामाजिक सांगीतिक कार्यकर्ता के रूप में अपनी अहम् भूमिका निभाई है। उन्होंने शास्त्रीय, उपशास्त्रीय एवं भजनों के अंतर्गत लगभग ४५० बंदिशों की, रचनाएं की तथा इन बंदिशों को 'स्वरांगिनी' एवं 'स्वरंजनी' पुस्तक के रूप में संगीत समाज को प्रदान किया है एवं इन समस्त बंदिशों को सी.डी. में प्रस्तुत कर संगीत प्रेमियों एवं कलाकारों को नवीन सामग्री प्रदान की प्रभाजी ने न केवल भारत में अपितु विदेशों में भी विविध कार्यक्रमों, प्रशिक्षणों एवं व्याख्यानों द्वारा संगीत का प्रचार - प्रसार करने में अपना योगदान प्रदान किया है। आपने कॅनडा, अमेरिका, नेदरलैण्ड, स्विट्जरलैण्ड आदि देशों में कई स्थानों पर कार्यक्रम, कार्यशालाएं, व्याख्यान प्रदर्शन कर संगीत के प्रचार - प्रसार में अपना योगदान प्रदान किया है।

आप भारत में ही नहीं अपितु विदेशों में भी संगीत प्रेमियों को संगीत शिक्षा प्रदान करती रही हैं। अब तक आपने देश - विदेश के लगभग २५० शिष्यगणों को शिक्षा प्रदान की है और करती आ रही हैं। प्रभा जी ने भारतीय संस्था संस्कार भारती के माध्यम से भी कई व्याख्यान, सेमिनार, कार्यशालाएं, कार्यक्रमों एवं प्रशिक्षण द्वारा भारतीय संस्कृति में संगीत को बढ़ावा देने, तथा



उसे सुरक्षित रखने का कार्य किया। आपने कुछ सामाजिक, शैक्षिक तथा सांस्कृतिक संस्थाओं में संगठन सदस्य के रूप में भी कार्य किया। भारतीय शास्त्रीय संगीत, अन्य कलाओं के संरक्षण एवं उनके विकासात्मक दृष्टिकोण हेतु सन् २००२ में डॉ. प्रभा अत्रे फाउण्डेशन की स्थापना की। लेकिन फाउण्डेशन की स्थापना के पूर्व सन् १९६५ से ही कई कार्यक्रम आयोजित किए हैं।

आप लगभग पन्द्रह वर्षों से कई भारतीय एवं अन्तराष्ट्रीय संगठनों से भी जुडी हुई हैं। भारतीय शास्त्रीय संगीत को साधारण जनता में लोकप्रिय बनाने के लिए आपने मन्दिरों एवं आश्रमों में भी निरन्तर कार्यक्रम प्रस्तुत करती रही हैं तथा विशिष्ट कारणों से वे कई कार्यक्रमों में बिना मानधन के भी प्रस्तुति देती हैं। प्रभा जी विदेशों में भी अनेक विश्वविद्यालयों में मानद अध्यापिका (अपेपजपदह चतवमैवत) के रूप में अध्यापन कार्य करती हैं। आपने आकाशवाणी, दूरदर्शन कार्यक्रम, व्याख्यान प्रदर्शन कार्य एवं उत्कृष्ट कार्यक्रमों द्वारा लोकप्रियता एवं प्रशंसा अर्जित की है तथा इस तरह से आपने भारतीय सांगीतिक कला व संस्कृति को विदेशों में भी लोकप्रिय बनाया है। इस प्रकार प्रभा जी ने एक कलाकार होने के अतिरिक्त लेखिका, आयोजिका, गुरु एवं सांगीतिक, सामाजिक कार्यकर्ता आदि विविध पहलुओं द्वारा भी भारतीय सांगीतिक संस्कृति की धरोहर को सुरक्षित एवं लोकप्रिय बनाने में महत्वपूर्ण भूमिका निभाई है। संगीत में इस अमूल्य योगदान के लिए आपको शतशः नमन।

#### संदर्भ सुची

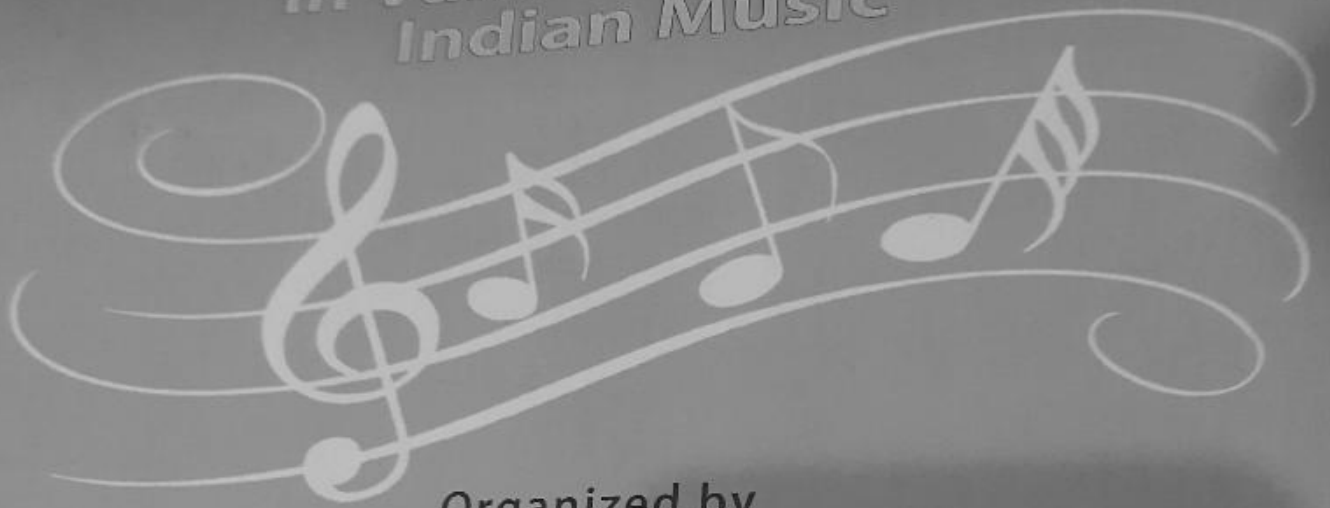
- १) संगीत कला विहार मासिक अंक : फेब्रुवारी २०१२
- २) संगीत पत्रीका



# **National Seminar**

On

**Role of Literature  
in Various Forms of  
Indian Music**



Organized by

**UNIVERSITY OF MUMBAI,  
DEPARTMENT OF MUSIC**

**Wednesday, 10th January, 2018**

**-Venue-**

**Shahir Amar Sheikh Auditorium,  
V.V.Bhavan, University of Mumbai  
'B' Road, Churchgate, Mumbai**



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# Role of Literature in Various Forms of Semi Classical Music

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## Abstract :

This paper provides an overview about role of literature in various forms of semi classical music like Thumari, Dadara, Kajri, Savant, Chaiti, Hori, Barmasa, Thumari and Bhajan and Thumari and Gazal.

These semi classical songs are basically in regional literature dialects of Hindi such as Awadhi, Bhojpura, Mizapuri etc. The dialect of Thumari is soft and tender than any other forms and it allows making colloquial usage of words to sound them elastic, for example – 'Paani' becomes 'Paniyaa' and 'Piyaa' turns into 'Piu' or 'Piyaruwa', etc.

Indian Classical Music or rag sangeet as we know is based upon (swarapradhan gayki) musical notation played major role in singing music and semi classical or light music is based upon shabdapradhan gayki means words played major role in singing of music.

Semi classical composition of Thumari in other languages such as Rajasthani, Marathi and Bengali.

## Introduction :

Music is so much a part of our lives that its presence in literature can help readers relate to fictional situation and characters. Also characters who love music are often creative people (as is the case with real-life music lover)

Thumari stands as an important and dominant genre in Indian music along with Dhrupad, Khayal and Tappa; as a well-accepted genre by all performers, musicologists and audience. With keeping its unique character intact as a musical form, Thumari has its own idiom, scholastic tradition, aesthetics and mannerisms, which are in many ways different than Khayal and Tappa, but still there are many commonalities.

The Hindi word 'Thumari' is said to be derived from – 'Thumakna' meaning an attractive gait. So, literary meaning is 'the song having attractive – rather sensuous, gait of melody and rhythm'. The content of sensuousness is the main emotive basis in Thumari,



compositions of Dadra, in fast tempo. Some able Thumari singers also sing Thumari in slow tempo Kehrawa or Dadra.

### **Difference between Thumari and Dadra:**

'Dadra' can be explained as a speedier version of Thumari, approximately. Though the name suggests about the Taal Dadra, the compositions are set in other taals than dadra such as Kehrwa and Chachar. An example of a dadra is Savare Aijaiyo by Dr Vasant Rao Deshpande

Though Thumari and Dadra are mentioned always as a twin-term, there is some distinction –

- Thumari is usually sung in slow tempo and Dadra is bit faster.
- Thumari has more elaborate, lengthy structure of improvisation than crisp, compact Dadra.
- The lyric of Thumari generally possesses only two parts – Sthayi and Antara. On the contrary, Dadra is decorated by and large with more than one Ataras.
- Thumari mostly says purely about the human love relationship. But the songs in the category of Dadra such as Kajri, Jhoola, Hori, Chaiti, etc. mostly depict the nature, seasonal variation and the human sentiments in that reference.

### **Forms under the umbrella of Dadra:**

- Kajri: 'Kajri' word means 'Black – rainy clouds'. Kajri mainly explains the pathos of a separated lover during rainy season. But the typical of Mirzapuri Kajri also narrates the joy in rains. Eg. Shobha Gurtu – Tarsat Jiyara Hamar Naihar Me
- Sawan: Sawan also is a rainy season song, but rather than explaining the human sentiments, it gives emphasis on the seasonal beauty. Eg. Shobha Gurtu – Sakhi Sawan Aayo Jhoola: This is the song while playing the swings, sung by women in north India during the rainy season, with the depiction of romantic mood of Lord Krishna and Radha. Eg. Shobha Gurtu – Jhoola Dheerese Jhulao
- Chaiti: Song to be sung in the summer month, Chaitra, which has depiction of girl asking for new bridal dress to her husband, mostly. In Chaiti, there is usage of words 'Ho Raam'. Eg. Shobha Gurtu – Chaitar Chunariya Rang De



- Barahmasa: That has description of all the three seasons in the twelve months in Indian scenario.

#### **Thumari and Bhajan:**

'Bhajan' means devotional song, specifically written by the saint-poets such as Meerabai, Kabir, Surdas, etc. Many Thumari singers sing the Bhajans in the format of Thumari-Dadra. So, it adds the repertoire of the Thumari's subject matter, not restricting it only to the sensual realm. The compositions such as 'Saiyya Nikas Gaye' or 'Barse Badariya Sawan Ki' are fine examples of Thumari-Ang Bhajans.

#### **Thumari and Ghazal:**

It is a form of Urdu-Farsi poetry with its unique stylistic construction and subject matter is mainly related to love relationship. Till the first half of 20th century, Ghazal was also often sung in Thumari-Dadra format. But later, as there was development of distinct style of Ghazal rendering, this form was separated from Thumari's influence. On the other hand, there are some Dadras in which Sher's (couplet) in Urdu poetry are rendered between two Antaras. For example – 'Chha Rahi kali Ghata' (Dadra in Desh).

#### **Gharanas in Thumari:**

Thumari is said to be originated in the Purab, i.e. eastern region of Ganga-Yamuna rivers in north India, so it is called as 'Purabi' or 'Banarasi' Thumari, which is sung mostly in slow tempo. Later new style emerge called 'Lucknowi Thumari' which gives more importance to fast tempo compositions, also called as 'Pachhahi Thumari'. Another school in Thumari came out, that was 'Punjabi Thumari' which has lighter rendering, but fanciful and startlingly attractive phrases.

Benaras Gharana – Girija Dev – Pilu – Preeti Kiye

Punjab Gharana – Ustad Bade Ghulam Ali Khan – Pilu – Kate Na Birha Ki Raat

#### **Conclusion :**

I want to highlight the literature of various Saints and Poets of Urdu, Parsi poetry with its unique stylistic construction. The composition such as, "Saiyya Nikas Gaye, Naina Ladi", or "Barse Badriya Sawan Ki", and "Yad Piyaki aaye" are the fine examples of Thumari Ang.

#### **Bibliography :**





though there are many compositions of Thumari depicting the devotional aspect. An example of thumri is Bari Umar Larkaiya Na Chhedo Saiyyan by Shobha Gurtu.

#### **Origin:**

Some musicologists speculate about the traces of Thumari in ancient form – ‘Charchari Prabandh’ or ‘Hallisak Geeti’. But the available documentation on Thumari mentions its origin in around 16-17th century A.D. Thumari is said to be originated from the songs of Northern Indian folks, specifically from the region between Ganga – Yamuna Rivers.

#### **Subject matter:**

Thumari portrays mainly various moods in love – unison, separation and such ups and downs in the journey of relationship. The main character in the lyric of Thumari is mostly a woman in love, and the illustration differs in the stages of the disposition such as age, social status, etc.

#### **Musical characteristics:**

In Thumari, the lyrics i.e. ‘Bol-ang’, is very important. So, the musical elaboration of the words with different shades is focused in the rendering, which is called as ‘Bol Banaao’. This involves Alap, sometimes with mixtures of Raags for highlighting the sentiments. After singing the Sthayi and Antara in slow tempo, usually there is rendition of words in fast progression on Tabla called ‘Laggi’ when the singer twists the words with melodic variations called as ‘Bol-Baant’.

#### **Raags:**

Thumari is sung mostly in the so called ‘lighter’ Raags such as Khamaj, Kafi, Tilang, Desh, Tilak Kamod, Sorath, Piloo, Mand, Manjh-Khamaj, Jogia, Kalingda, Shiv-ranjani, Bhairavi, etc. in which there is wider scope for emotive improvisation with the subtleties in Alankaars, mixtures of Raags. So, many times, Thumari is found in combination of some Dhuns, so called as ‘Jhilla’ and ‘Jangulaa’. But the tradition has gifted us some Thumaris in raags typical of Khayal, such as Bihag, Shahana, Sarang, Poorvi, Kalyan, Sohni, etc.

#### **Taals:**

The Taals for Thumari are Deepachandi, Addha, Ikwaai, Sitarkhani, and some ‘Bandhi Thumari’ is to be sung in Jhaptal, Ektaal also. There taals of smaller cycle, derived from folk music, that are Kehrawa, Dadra, Khemta, Chachar which are used for the

though there are many compositions of Thumari depicting the devotion of thumri is Bari Umar Larkaiya Na Chhedo Saiyyan by Shobha Gurtu.

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Thumari portrays mainly various moods in love – union, separation and such ups and downs in the journey of relationship. The main character in the lyric of Thumari is mostly a woman in love, and the illustration differs in the stages of the disposition such as age, social status, etc.

#### **Musical characteristics:**

In Thumari, the lyrics i.e. 'Bol-ang', is very important. So, the musical elaboration of the words with different shades is focused in the rendering, which is called as 'Bol Banaao'. This involves Alap, sometimes with mixtures of Raags for highlighting the sentiments. After singing the Sthayi and Antara in slow tempo, usually there is rendition of words in fast progression on Tabla called 'Laggi' when the singer twists the words with melodic variations called as 'Bol-Baant'.

#### **Raags:**

Thumari is sung mostly in the so called 'lighter' Raags such as Khamaj, Kafi, Tilang, Desh, Tilak Kamod, Sorath, Piloo, Mand, Manjh-Khamaj, Jogia, Kalingda, Shiv-ranjani, Bhairavi, etc. in which there is wider scope for emotive improvisation with the subtleties in Alankaars, mixtures of Raags. So, many times, Thumari is found in combination of some Dhuns, so called as 'Jhilla' and 'Jangulaa'. But the tradition has gifted us some Thumaris in raags typical of Khayal, such as Bihag, Shahana, Sarang, Poorvi, Kalyan, Sohni, etc.

#### **Taals:**

The Taals for Thumari are Deepachandi, Addha, Ikwaai, Sitarkhani, and some 'Bandhi Thumari' is to be sung in Jhaptal, Ektaal also. There taals of smaller cycle, derived from folk music, that are Kehrwaa, Dadra, Khemta, Chachar which are used for the