SECTION A

SCIENCE

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Antioxidant and in-vitro activities of chara sp. aqueous extract from waghur river at sakegaon, maharashtra

Authors & affiliations:
Uzma Naaz Shaikh Iqbal, S. N. Thange, Y. R. Sonawane and K. J. Patil*
Department of Biochemistry, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Antioxidants prevent or slow damage to cells caused by free radicals, unstable molecules that the body produces as a reaction to environmental and other pressures. Seaweeds are rich in different bioactive compounds with potential uses in drugs, cosmetics, and the food industry. In this study, The antioxidant potential of the crude extract of *Chara* sp. fresh water seaweed collected from Waghur river Sakegaon, Maharashtra determined using total phenolic content, total antioxidant capacity, hydrogen peroxide scavenging activity and hydroxyl radical-scavenging activity. Supplementary, the crude extract of *Chara sp.* applied in-vitro medicinal activities like anti-diabetic, anti-inflammatory, heat induced hemolytic, protein inhibitory action and hemolytic activity. The aqueous extract of *Chara sp.* shows moderate antioxidant activity as compared to L-ascorbic acid and show potential agent for medicinal activities. Further, for phytochemical research is needed to identify the active principles responsible for the antioxidant activity.
Keywords: *Chara sp.*, Antioxidant, in-vitro, anti-diabetic, anti-inflammatory

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Introduction:

Chara sp. Algae are photosynthetic organisms and are commercially cultivated for pharmaceuticals, neutraceutical, cosmetics, and aquaculture purpose. Sea nutrients such as algae and sea kelp are loaded with essential minerals and vitamins. Moreover, offer rejuvenating properties to prevent the aging process and help in activating the cell renewal. Dichloromethane, ethanol, and boiling water extracts of the brown seaweeds *Sargassum fulvellum and Sargassum thunbergii* were examined for antipyretic, analgesic, and anti-inflammatory activities in mice by Kang *et al.*,(2008). Other activities of algae reported include wound healing, anti-inflammatory, antidiabetic, antioxidative, anticancer, anticoagulants, antibiotics, anti-hypertensives, dilatory agents, blood cholesterol reducers, insecticides, anti-tumorigenic agents, nutritional supplements, and pharmaceutical applications.(Wikipedia 2019 Algae retrieved on 1 Jan 2019). *Chara globular* is though appears to be a plant but is actually a multicellular macroalga. *Chara* grows attached to the bottoms of ponds, lakes, rivers, and ditches and can form submersed beds of vegetation. Individual plants can range in size, from a few inches in length to several feet in length. Chara has whorls of 6 to 8 branchlets that arise from nodes along the stem. Monoecious and dioecious species exist, but vegetative plants persist year-round. During times of reproduction, dark, ball-like sporangia appear seed-like along the branchlets. *Chara* is also known to have a strong garlic odor. This macroalga has no true "leaves," only branches and branchlets. The thallus of *Chara* is branched, multicellular, and macroscopic, which is mainly differentiated into rhizoids and main axis. A string alga *(Cladophora)* is caused by a filamentous species of algae, which grows in long strands. These algae eventually tangle together and forming thick mats that can double their weight within 24 hours. Blanket weed or string algae tend to adhere to rocks and waterfalls, which can be unsightly. (Wikipedia 2019 *Chara* algae retrieved on 1 Jan 2019).

Antioxidants The antioxidant activity of putative antioxidant has been attributed to various mechanisms, among which are reduction of free radicals, prevention of chain initiation, binding of transition metal ion catalysts, chelating, decomposition of peroxides, prevention of continued hydrogen abstraction, and as oxygen scavengers and thus can be utilized to scavenge the excessive free radicals generated from human body. There are several methods available to assess antioxidant activity of compounds. The easy, rapid, and sensitive methods for screening-free radical scavenging

activity are hydrogen peroxide, nitric oxide, alkaline dimethyl sulfoxide, and reducing power assay methods.

Besed on above point of view and conclusion of review of litreture presnt work is focused on antioxidant activity of fresh water algae *chara sp.* and its possible application in medicinal field based on antioxidant capacity.

Materials and Methology:

Collection of sample: The sample of algae collected from Waghur River at Sakegaon, Maharashtra India. The sample collected in bags and transported to lab for further process.

Antioxidant Activity:

Determination of total phenoliccontents: Total phenolics contents assayed using the Folin-Ciocalteu reagent and gallic acid as a standard following Singleton's method slightly modified by Gulcin et al.,(2002)

Total Antioxidant capacity (TAC): The reagent solution (0.6 M Sulphuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). The absorbance measured at 695 nm. The antioxidant activity expressed as the number of equivalents of ascorbic acid. Prieto *et al.*,(1999).

Hydroxyl radical scavenging assay: Scavenging of the hydroxyl free radical measured by the method of Yu et al.,(2004).

$\% \, inhibition = [(A_0 - A_1)/A0] \, X \, 100$

A0-absorbance of blank, A_1- absorbance of extract

Hydrogen peroxide scavenging assay: Hydrogen peroxide activity of the extract estimated by replacement titration method Zhang (2000).

$\% \, inhibition = [(V_0 - V_1)/V0] \, X \, 100$

V_0-Volume of thiosulphate used to titrate blank

V_1- Volume of thiosulphate used to titrate against the extract.

IN VITRO ACTIVITY: The potency of aqueous extract of chara estimated by following methods:

In vitro α-amylase inhibition activity by Spectrophotometric method:In vitro α-amylase inhibition activity of the extract estimated by the method of Prabhakar (2013).

$Percentage \, inhibition = (Abs_{control} - Abs_{sample}) \, X \, 100/ Abscontrol$

In-vitro anti-inflammatoryactivity: Inhibition of albumin denaturation Method of Mizushima et al., (1968) followed with minor modifications.

$Percentage \, inhibition = (Abs_{control} - Abs_{sample}) \, X \, 100/ Abs_{control}$

Heat induced hemolysis: Heat induced hemolysis measured by the method of Shinde et al.,1999 and Saket et al.,2010. Percent membrane stabilization activity calculated by the formula mentioned below:

$Percentage \, inhibition = (Abs_{control} - Abs_{sample}) \, X \, 100/ Abs_{control}$

Proteinase inhibitory action: The test performed according to the modified method of Govindappa et al.,(2011).

$Percentage \, inhibition = (Abs_{control} - Abs_{sample}) \, X \, 100/ Abs_{control}$

Haemolytic activity: In vitro haemolytic activity performed by spectrophotometer method (Yang et al.,2005). The level of percentage hemolysis by the extracts calculated according to the following formula:

$Percentage \, inhibition = (Abs_{control} - Abs_{sample}) \, X \, 100/ Abs_{control}$

Result and Disscussion:

Antioxidant Activity:

Determination of total phenolic contents: The total phenolic activity of extract was 9.78, 20.96, 27.41, 37.2, 48.38 at different concentration of extract ranges from 50, 100, 150, 200 and 250 µg/ml respectively was shown in figure no. 1 and table no. 1. By comparing activity with ascorbic acid varies from 0.081 to 0.838 for 0.1 ml to 1.0 ml of acid hence the extract showed moderate activity as compared to ascorbic acid. However TFC calculated by Whankatte V. R. et al.,(2016) for Cladophora

glomerata Linn. up to 14.35 + 0.030 for 10 mg/ml by using benzene as solvent. It was varied from 31.61 + 0.046, 70.02 + 0.055 to 73.49 + 0.023 with ethanol, ethyl acetate and methanol as a solvent for same concentration.

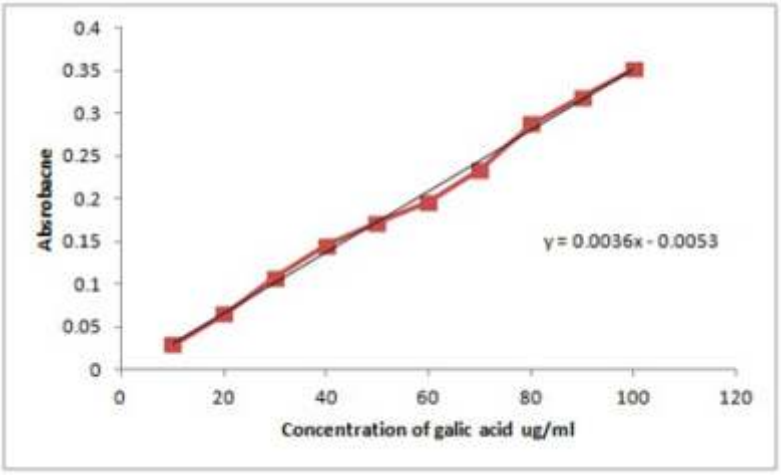

Figure 1: Standard graph of Gallic acid

Table 1: Total phenoic content of aqueous extract of Chara sp.

Concentration of substrate		Total Phenolic content
T1	50	9.78
T2	100	20.96
T3	150	27.41
T4	200	37.2
T5	250	48.38

Total Antioxidant capacity (TAC): The total antioxidant activity of extract found to be 153.69 , 219.25 , 301.47 , 338.13 and 436.47 at different concentration of extract ranges from 50 ,100 ,150 , 200 ,150 µg/ml concentration respectively was shown in figure no. 2. by compairing its activity with ascorbic acid,varies from 0.013 to 0.256 for 0.1 to 1.0 ml of acid sample hence test extract shows modrate activity as compare to ascorbic acid however TAC calculated by Massoumeh et al .; 2013 found to be only up to 81.36 for methonoic content of *C.linum* for 2 mg /ml of sample where as in work of Whankatte et al.; 2016 TAC was varies from 11.6 to 781.1 for different solvent like methanol and chloroform respectively by taking 10mg/ml concentration of sample.

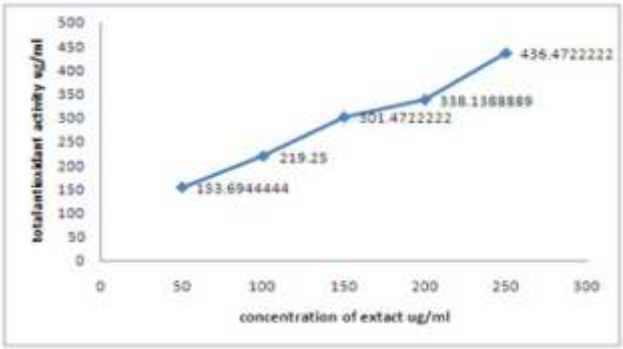

Figure 2: Total antioxidant activity of Chara sp. aqueous extract

Hydroxyl radical scavenging assay: The hydroxyl radical scavenging activity of extract was 22.03, 30.50, 45.76, 55.93, 83.05, 96.61 at different concentration of extract ranges from 50,100,150,200, 250 µg/ml concentration respectively given in table no. 2.However work done by Murugesan et al.,(2016) the HRSA for S.fusiformis exhibited the lower inhibition up about 20.05+0.02 for 392 µg/ml and C.hornemanii shows 12.62 +0.03 for 355µg/ml of sample against hydroxylradical.

Table 2: Hydroxyl radical scavenging activity of Chara sp. aqueous extract

Test extract	Concentration(µg)	HRSA
T1	50	22.033
T1	100	30.508
T2	150	45.764
T2	200	55.938
T3	250	83.050
T3	300	96.610

Hydrogen peroxide scavenging assay: The hydrogen peroxide scavenging activity of extract was 1.58, 4.76, 7.93, 11.11, 15.87 at different concentration of extract ranges from 50, 100, 150, 200, 150 µg/ml concentrations respectively mentioned in figure no. 3. However, HPSA calculated by Indu et al.,(2013) for Sargassum sp. exhibited more prominent effect was approximately up to 90% at 2 mg/ml whereas in work of Mukund et al.,(2013) HPSA was up to 60% for Oscillatoria terebriformis by taking 250 µg/ml of sample in methanolic solvent.

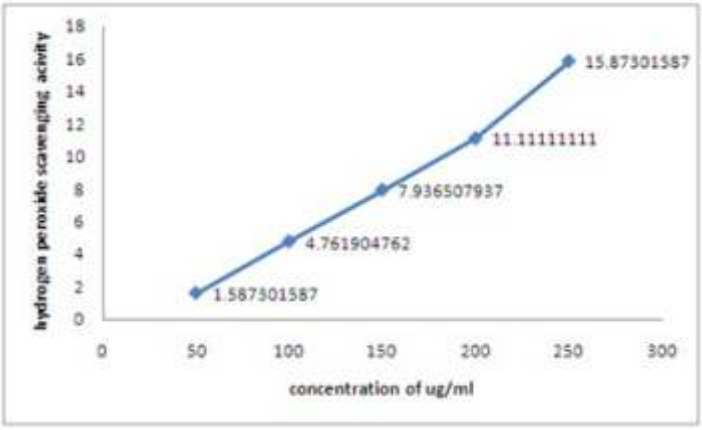

Figure 3: Hydrogen peroxide scavenging activity of extract

In Vitro Activities:

In vitro α- amylase inhibition activity by Spectrophotometric method: In the present study aqueous extract of *Chara sp.* algae assessed for in vitro α- amylase inhibition activity. The concentration 100 µg/ml exhibited 62.20% of inhibition given in table no. 3. Sangeetha et al.,(2017) reported that methanolic extract of *Nannochloropsis occulate* exhibited 66.87 % of inhibition at 1000µg/ml concentration. Won-Min Pak et.al.,(2015) investigated 13% of α-amylase inhibition activity at a concentration of 5.0 mg/ml of methanol extract of *Myagropsis myagroides* and its aqueous extract showed less than 5% of inhibition,Whereas Fruit pulp of M. charantia var. charantia (MCC) and *M. charantia var. Muricata* showed 66.5 % and 67 % of inhibition at a concentration 2.5 mg/ml of its protien extract, investigated by Poovitha and Parani (2016). As aqueous extract of *Chara* algae showed significant α- amylase inhibition activity, so with the further research it can be used as natural antidibetic.

Assessment of in vitro anti-inflammatory activity: Anti-inflammatory activity studied with the help of inhibition of albumin denaturation. Aqueous extract of Chara algae showed inhibition of denaturation of albumin in concentration dependent manner. Aqueous extract of *Chara* algae showed 94.36% of inhibition at a 100 µg/ml concentration shown in table no. 3, which was more than that showed by *Nannachloropsis occulate* studied by Sangeetha *et.al.*,(2017) & founded 32.25% of inhibition at 100 µg/ml of methanolic extract. Methanol extract of *Chlorella vulgaris* has 72.67% of inhibition at a 500 µg/ml concentration studied by Bhuvana *et.al.*,(2018). Leelaprakash *et.al.*,(2011) reported that methanol extract of *Enicostemma axillare* showed 71% inhibition of albumin denaturation at a 500 µg/ml concentration. Protein denaturation is cause of inflammation therefore investigation of anti-inflammatory studied by the albumin denaturation.

Heat induced hemolysis: Heat induced hemolysis carried out for the study of stabilization of RBC's membrane. Aqueous extract of Chara algae showed 110.12 % of inhibition at a concentration 100µg/ml given in table no. 3, which was higher than inhibition showed by the methanol extract *Chlorella* vulgaris i.e. 40.94 % of inhibition at a 100µg/ml concentration reported by Bhavana et al.,(2018). 73.45% of inhibition showed by the methanol extract of *Trametes ochracea* studied by Govindappa et. al.,(2015). G. Leelaprakash et.al.,(2011) reported that methanol extract of *Enicostemma axillare* are showed 51 % inhibition at 500 µg/ml concentration. Hence, aqueous extract of Chara algae showed significant heat induced haemolysis and thus this extract may possibly inhibit tissue inflammation caused by the neutrophil release at the site of inflammation.

Proteinase inhibitory action: Proteinase inhibitory action studied at different concentration in which standard solution showed 13.46% of inhibition at 10µg/ml concentration mentioned in table no. 3. The aqueous extract of *Chara* algae showed 4.80% and 20.19% of inhibition at 50 µg/ml and 100µg concentration. Govindappa *et al.*,(2015) reported 84.82% of inhibition in methanol extract of *Trametes ochracea.* Leelaprakash *et.al.,*(2011) reported that methanol extract of *Enicostemma axillare* showed 53% of inhibition at 500 µg/ml concentration.

Haemolytic activity: Aqueous extract of Chara algae showed dose dependent haemolytic activity showed 13.38% and 69.62% of haemolytic activity at 50 µg/ml & 100 µg/ml concentrations given in table no. 3. Some other plants also showed haemolytic activity in different extracts. Ghosh (*et al.*, 2018) reported that ethyl acetate extract of Croton *bonplandianum* showed 81.25%of haemolytic activity at a 75 µg/ml concentration. *'Tamarixa phylla and Daphne gnidium* given 6.568 % and 7.060% hemolysis at 500µg/ml. *Daphne gnidium* and *Tamarixa phylla* provoked toxicity but less than *Morettiacanescens* while a great hemolytic effect on erythrocyte cell membrane was caused by this species (14.80% hemolysis) Zohra et al.,(2014).

Table 3: In-Vitro activity of Chara sp. aqueous extract

		Percentage of Enzyme Activity	Percentage of α- amylase inhibition	InVitro Anti Inflammatory Activity	Heat Induce Haemolysis	Protease Inhibition	Hemolytic activity
Untreated		100	0	42.25	150.60	0	100
Control		76.73	23.26	26.76	105.46	13.46	13.38
T1	50	41.59	58.40	53.52	110.12	4.80	69.62
T2	100	37.79	62.20	42.25	150.60	20.19	100

Conclusion:

The aqueous extract shows potential in-vitro medicinal activities like anti-diabetic, anti-inflammatory, heat induced hemolytic, protein inhibitory action and hemolytic activity.The antioxidant activity of *Chara* fresh water algal aqueous extract was analyzed by total phenolic activity, total antioxidant capacity, hydrogen peroxide scavenging activity and hydroxyl radical-scavenging activity.

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Biological activities of alkaloidal leaves extract of carica papaya L.

Authors & affiliations:
V. V. Bhamare, N. R. Sonar and B. R. Kavimandan*
Department of Biochemistry
M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Carica papaya L. is one of the most important traditional medicinal plant consisting of various food and nutritional values. Considerable progress is observed in awareness to such essential medicinal plants. Fruits, leaves, latex, seed, stem and bark of this plant posses appreciable pharmaceutical properties. *Carica papaya* L. also contains a broad spectrum of phytochemicals such as poly-saccharides, proteins, lipids, vitamins, minerals, alkaloids, flavonoids , saponines , sterols, glycosides etc. *Carica papaya*L.leaves have great potential to fight against various diseases and infections. So alkaloids were isolated from the leaves of *Carica papaya* L. using water as a solvent and various biological activities like Anti-oxidant, Anti-microbial, Anti-diabetic, Anti-helminthic activities were carried out and obtained encouraging results.

Keywords: *Carica papaya* L., alkaloids , phytochemicals, biological activities.

Introduction:

Indian culture is blessed with Ayurveda and*Carica papaya* L. has been used as medicinal plant from traditional system of medicine in our country. Ayurvedic medicine is a system of healing that rely heavily on herbs and other the plants including oil. Currently, more than 600 herbal formulas and 250 single plant drugs are included in the pharmacy of Ayurvedic treatments. The importance of plants in search of new drugs is increasing with the advancements of medical Science(Anitha B.*et.al*., 2018).
Now a days, people are very aware about uses and importance of medicinal plants due to their effective, cheap, non-allergic, safe and nutritive nature. The alternatives for Anti-biotics, chemicals and drugs are made available by these medicinal plants (Ahmad.N.*et.al*., 2011).
Carica papaya L. plant is enriched with various food, nutritive values as well as phytochemicals and thus have acquired centre of attention in field of Pharmacy, Nutrition, Drugs and many more. Each and every part of papaya plant i.e. Leaves, Fruits, Latex, Bark, Seed, Root, Flower are useful for medicinal purposes. Out of them, leaves are store house of many nutrients which are beneficial as medicine and contains broad spectrum of phytochemicals such as Polysaccharides, Proteins, Lipids, Vitamins, Minerals, Alkaloids, Flavonoids, Saponins, Sterols, Glycosides etc. (Verma.S.*et.al.*, 2017).
Modern pharmacy now deals with medicinal plants and getting excellent biological activity of those plants. Papaya leaf extract is derived from the leaves of papaya tree. Papaya tree is commonly found everywhere. Papaya tree can said medicinal plant as it has proven several biological activity and active against several diseases. There are many health benefits of papaya leaf extract which including micronutrient provider, Red Blood Cell production, Immune booster, Anti-bacterial activity, Anti-helminthic activity, Anti-diabetic activity, Platelets enhancer and aids in digestion(Baskaran C.*et.al.*, 2012). Hence the plant enriched with all those medicinal and pharmaceutical properties selected for study.

Objective

So we have undertaken this project with following objectives:

- Extraction of leaf using suitable solvent.
- Phytochemical Qualitative Analysis of the extract.
- Total alkaloid estimation.

- Partial purification of alkaloid.
- FTIR analysis.
- Study of Anti-bacterial, Anti-helminthic and Anti-amylase activity of different extracts.

Methods:

1.Collection, shed drying and extract preparation of *Carica papaya* L. leaves:
Fresh, young and healthy leaves were collected for the research work. Firstly, they were washed properly with tap water 3-4 times and shed-dried for 2-3 days. Totally dried leaves then finely powered using blender and stored in air-tight container for future use. 30g powder was then added to 300ml D.W. and kept on stirrer continuously for 12hrs. The sample formed then filtered using filter paper thrice to get watery extract.

2.Qualitative tests for phytochemical analysis: Phytochemical screening was performed by using standard procedure given by Mahmauda Begum.*et.al.(*2014)
3.Total alkaloid estimation: Total alkaloid content of the sample was measured using 1, 10-phenanthroline method described by Irondi.*et.al*.(2013) and Shodagandha with modifications.

4. Partial purification of alkaloid: Alkaloid and its derivatives were partially purified by the method given by Faizal M.*et.al.*(2013).

5. FTIR Analysis:FTIR analysis was carried out for functional group detection using AR affinity -1, Shimadzu by William D.*et.al*.(2010), Svehla G.*et.al*.(1979)

6. Anti-bacterial activity:Anti-bacterial activity was studied against *Streptomyces gracius, E.coli, Pseudo putida, Staphylococcus aureus*and*Xanthomonascitri* by Bore-well method. Streptomycin (0.5mg/ml) was taken as standard while D.W. as control were observed against test sample (0.1mg/ml) by Lohidas J.*et.al.*(2015)

7. Anti-helminthic activity: Indian red earthworms (*Eiseniafoetida*) were selected for anti-helminthic activity. Six earthworms were kept in different sample conc. 100%,75%,50% & 25% resp. and Zentel suspension(40mg/ml)was taken as standard as well as D.W. as control. Time for paralysis(P) and death(D) were observed and noted down by Kanthal. L.*et.al.*(2012)

8. Anti-Amylase activity: Anti-diabetic activity was studied by using DNSA method at 540nm and percent of enzyme inhibition was observed by Bernfeld P.*et al*.(1955)

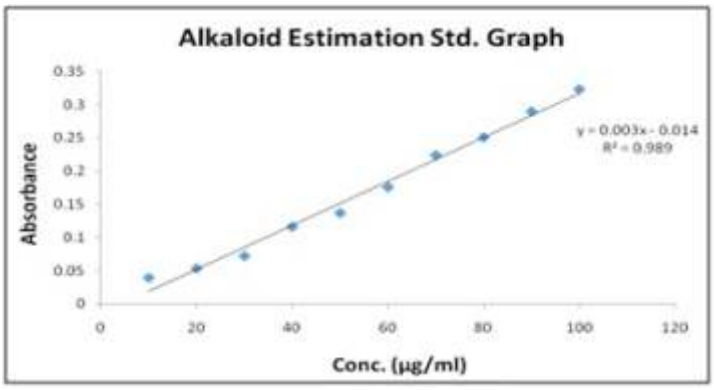
Results and Discussions:

1.Phyto-chemical screenings: Phytochemical screening showed following results;

Test	Water
1] Alkaloids	
a.Mayers Test	+
b. Wagner's Test	+
2]Carbohydrate	
a.Fehlings Test	_
b.Benedicts Test	_
3] Saponing (foam test)	+
4] Protein & Amino Acids	
a.Biuert Test	_
b.Ninhydrin Test	_
5] Fixed oils &Fats	
a. Spots Test	+
6] Phenolic Compounds & Tannins	+
a. Ferric Chloride Test	_
b. Lead Acetate Test	+
7] Flavonoids	
a. Alkaline Reagent Test	_

From the above phytochemical screening, the extract from Carica papaya L. shows presence of primary metabolites like Carbohydrates, Lipids and absence or less content of Proteins as well as Secondary Metabolites like Alkaloids, Saponins, flavonoids and absence or less content of Tannins, Phenolic compound.

2.Total alkaloid estimation: Total alkaloid concentration in sample obtained as follows;

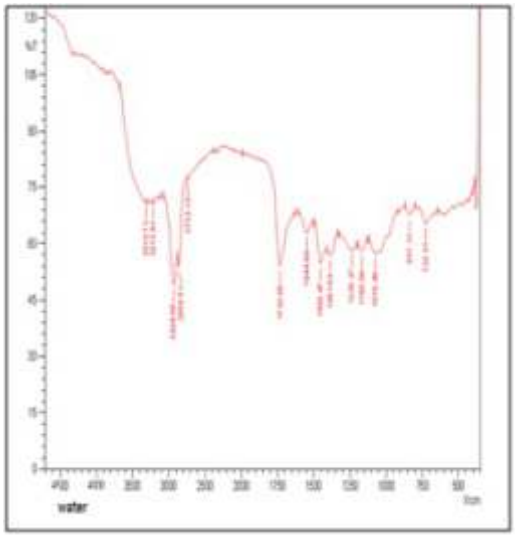


Thus, from standard graph obtained by Spectrophotometric method we found Alkaloid concentration in given extract as 28microgram/millilitre.

3.Partial purification of alkaloids:

Yield of partially purified alkaloid was found to be **5.066%** and weight **1.52g.**

4.FTIR Analysis: Functional groups were analysed as follows



Standard Frequency cm-^{1}	Observed Frequency cm-^{1}	Bond stretching
3400–3250	3313-3213	N-H Stretching in sec. amines
3000-2850	2924, 2858, 2733	C-H Stretching in methyl group
1750-1732	1732	C=O Stretching in ester
1600-1585	1544,1450	C-C Stretching in cyclic ring
1250–1020	1236	C-N Stretching in Amines
1320-1000	1165,1070	C-O Stretching in esters

The graph obtained from FTIR analysis showed presence of amines, methyl groups, esters, secondary amines and cyclic ring as functional groups.

5. Anti-bacterial activity: Bore-well method showed following results:

Streptomyces greciusE.coli

Pseudomonas putida

Staphylococcus aureusXanthomonascitri

Sr. No.	Bacteria	Diameter of zone of inhibition in Standard (cm)		Diameter of zone of inhibition in Test (cm)
		0.0001g/ml	0.0005g/ml	0.0001g/ml
1.	Streptomyces grecius	2.2	0.44	1.1
2.	E.coli	2.1	0.42	1.1
3.	Pseudmonasputida	3.2	0.64	1.6
4.	Staphylococcus aureus	2.4	0.48	1.7
5.	Xanthomonascitri	2.6	0.52	0

From above results for all five bacterias, we can conclude that the alkaloid extracted from *Carica papaya* L. is able to inhibit the growth or to kill the *Streptomyces grecius*, *E.coli*, *Pseudomonas putida* and *Staphylococcus aureus* bacterial strains and unable or less capable to inhibit growth of *Xanthomonascitri*bacterial strain. Out of all four inhibited strains, *Streptomyces grecius* showed maximum inhibitory results.

6.Anti-helminthic activity: Anti-helminthic activity of test sample was obtained as follows;**Sr.noSample conc.Time for paralysis(min)Time for death(min)**1Standard

Sr.no	Sample conc.	Time for paralysis(min)	Time for death(min)
1	Standard (0.002g/ml)	6	12
2	100% (0.002g/ml)	20	44
3	75% (0.0015g/ml)	28	56
4	50% (0.001g/ml)	34	67
5	25% (0.0005g/ml)	50	82
6	Blank	Alive	Alive

Above anti-helminthic activity observed in red earthworms showed positive results and rate of paralysis as well as death increased on increase in sample concentration. Thus, the extracted alkaloid works as a good anti-helminthic agent.

7.Anti-amylase activity: Inhibitory action of test sample on amylase enzyme has obtained by using following formula;

O.D(C) – O.D(S) / O.D(C) X 100

= 0.622 – 0.378 / 0.622 X 100 = **39.23%**

The sample showed **39.23%** inhibitory activity on amylase.

Amylase inhibitory activity shown by the extracted alkaloid is appreciable and thus the compound can be used as anti-diabetic agent as well as helpful in diabetic treatments up to certain extents.

Conclusion:

The *Carica papaya* L. plant showed positive results for Anti-bacterial , Anti-helminthic , Anti-diabetic activities and contains high amount of beneficial phytochemicals. Hence , the plant is potential therapeutic agent and useful for pharmaceutical purposes. Thus; this phytochemicallyenriched ,nutritionally valuable and pharmaceutically beneficial plant and it's alkaloidal extract can be used on medicinal as well as commercial levels.

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In-vitro screening of medicinal plants for their anti-diabetic activity

Authors & affiliations:
A. D. Wankhede, M. M. Kale, U. L. Thakkar and B. R. Kavimandan*
Department of Biochemistry
M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
In India, Diabetes Mellitus (type 2), is proving a major health problem. There are various approaches to reduce ill effects of diabetes but they come with secondary complications. So, herbal formulations are preferred due to lesser side effects and low cost. This screening project focuses on the anti-diabetic property of few natural medicinal plants by examining their amylase inhibitory activity. The medicinal plants and their parts are rich in enzymes and inhibitors. They play important role in control of endogenous amylase activity. The α-amylase inhibition activity of crude extracts of medicinal plant parts i.e. flowers of paneer phool (*Withania coagulans*), leaves of Bael (*Aegle marmelos*), Mango (*Mangifera indica*), Custard apple (*Annona reticulata*), and Guava (*Psidium guajava*) were tested. It was concluded that *Withania coagulans* shows maximum α-amylase inhibitory activity as compared to other plants taken in account.
Keywords: Diabetes Mellitus, α-amylase, inhibitors, *Withania coagulans*, *Aegle marmelos*, *Mangifera indica*, *Annona reticulata*, *Psidium guajava*.

Introduction:

Diabetes mellitus is a chronic, lifelong condition that affects the body's ability to use the energy found in food. It is a complex metabolic disorder that results in increased blood glucose level, is considered as one of the major health related problems and is growing rapidly worldwide. The International Diabetes Federation predicts that over 371 million people are struggling with this disease in 2012 with 63 million Indian contributions and 4.2 million deaths occurred (Sougata Ghosh, et. al. 2012). Basically, it is a slow killing disease which attacks the other organs further results in malfunctioning. There are three major types of diabetes; type 1diabetes (juvenile), type 2 diabetes (adult), and gestational diabetes. With diabetes mellitus, either your body doesn't make enough insulin or it can't use the insulin it produced, or a combination of both.

The trends treatment of diabetes mellitus includes many methods one is stimulation of beta-cells of islets of langerhans to secretes regulated amount of insulin. Second one includes inhibition of several enzymes known to breakdown complex food into glucose.

Alpha-amylase is an enzyme present in saliva and pancreas is carbohydrate hydrolyzing enzyme acts on complex starch convert it into glucose. Inhibition of enzyme amylase can be useful for type 2 diabetes. The inhibitors which are in clinical use were acarbose; miglitol plants use alpha amylase inhibitors as a major defensive and voglibose. (Sneha, J.A. et. al. 2011)

There exist many therapeutics for the treatment of Diabetes mellitus, but the comes with secondary complications which includes nausea, upset stomach, effect on kidney, etc. plants continue to play an important role in treatment of diabetes, particularly in developing countries where most people have limited resources and do not have an access to modern treatment. The increase in demand in industrially developed countries to use alternative approaches to treat diabetes such as plant based medicines, is also due to the side effects associated with the use of insulin and oral hypoglycaemic drugs. (Kandra L. et. al. 2004)

This is a characteristics symptom that the diabetic people face low insulin, which prevents the effective glucose metabolism in the blood. So, they need to be given α-amylase inhibitors in the order to keep their glucose under control. (Sudha, P. et. al.2011) Traditionally, some of the medicinal plants are used for the same but this lacks scientific validation. (Shukla R. et. al. 2000) The determination of

their amylase inhibitory activity can give the brief knowledge about the anti-diabetic effect of the plants.

So the current research project was undertaken to screen paneer phool (*Withania coagulans*), Bael (*Aegle marmelos*), Mango (*Mangifera indica*), Custard apple (*Annona reticulata*), Guava (*Psidium guajava*) for their in-vitro anti-diabetic activity.

Materials and Methods:

Collection of sample: Paneer phool (*Withania coagulans*), Bael (*Aegle marmelos*), Mango (*Mangifera indica*), Custard apple (*Annona reticulata*), Guava (*Psidium guajava*) were collected from local areas and were authenticated by the experts of botany department of the college.

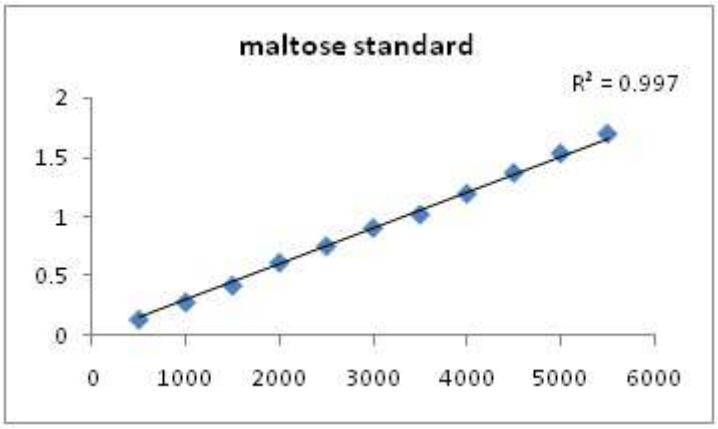
Extraction process

Flowers of paneer phool and leaves of rest of the selected plants (5gm/100ml) were extracted using water as solvent in a mortar pestle. Extracts were kept on shaker at 80rpm for 2 hours and were filtered using Whatman filter paper No. 1. Then extracts were centrifuged for 10-15 minutes and the supernatant was used as inhibitor.

In-vitro anti-diabetic activity was done by following methods:

Estimation of maltose, determination of the effect of substrate concentration on the activity of amylase and determine K_M and V_{max} of the reaction, determination of percent of inhibition activity at V_{max}, determination of effect of inhibitors on the enzyme activity of amylase and determine K_M and V_{max} of the reaction by using DNSA method given by Bernfeld et al. 1955 with slight modifications.

Standard graph of maltose: Maltose was estimated using DNSA method. (Bernfeld et al. 1955)



amylase activity using Michaelis-Menten Kinetics
Enzyme activity of amylase was estimated by DNSA method using Michaelis-Menten equation.

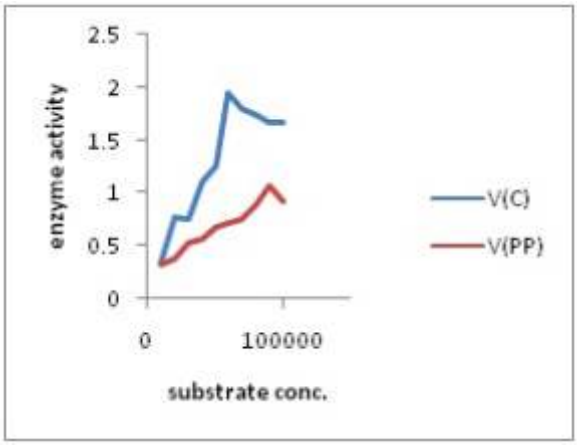
Determination of percent inhibition activity at V_{max}.
The inhibitors or the extracts were added at the V_{max} of the enzyme activity and inhibition percentage was determined.

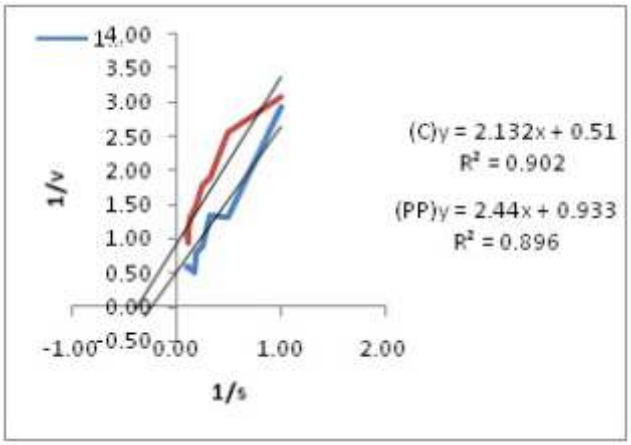
Inhibitor	Inhibition percentage
Paneer phool	56.28%
Mango	14.0%
Custard apple	19.96%
Guava	8.86%
Bael	20.45%

Amylase Inhibitory activity: The percent inhibition amylase activity by selected plant extracts was estimated and paneer phool had shown maximum inhibition and guava had shown minimum inhibition.

Effect of inhibitors on enzyme activity of amylase by using Michaelis-Menten equation
Paneer phool

Michaelis-Menten graph (V/S) Lineweaver-Burk plot

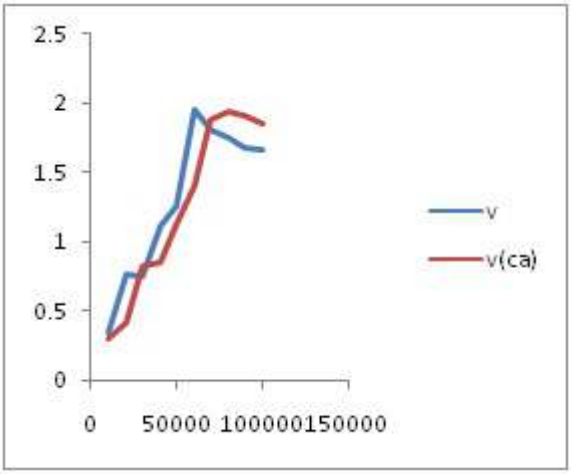




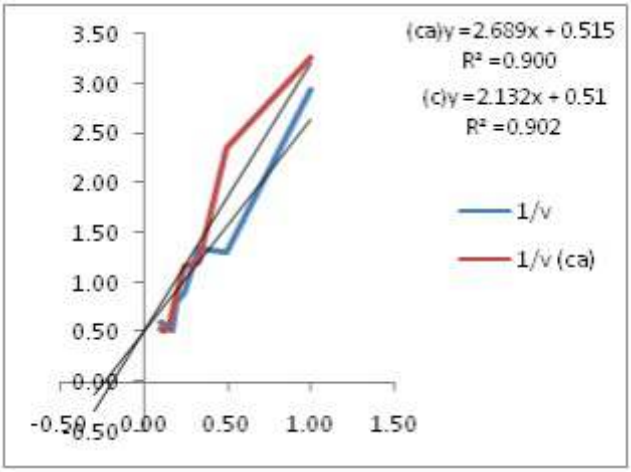
From the graph, it can be concluded that the extract of paneer phool shows uncompetitive inhibition.

Custard apple

Michaelis-Menten graph (V/S)



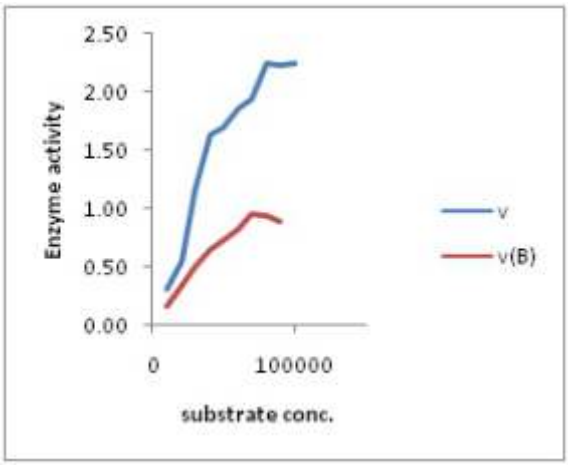
Lineweaver-Burk plot



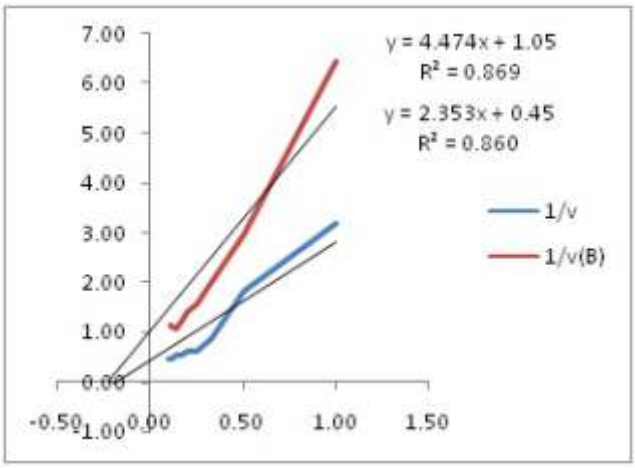
From the graph, it can be concluded that the extract of Custard apple shows competitive inhibition.

Bael

Michaelis-Menten graph (V/S)



Lineweaver-Burk plot



From the graph, it can be concluded that the extract of Bael shows uncompetitive inhibition.

Discussion

The rise of type 2 diabetes mellitus is a severe apprehension in medical world, which accounts for more than 9% death globally. α – Amylase inhibition plays an important role in the treatment of type 2 diabetes (Sneha, J.A. et. al. 2011). There are many medicinal plants used from ancient times but they lack scientific validation. The plant, which can inhibit amylase, can be an effective anti-diabetic drug. In this screening of the five medicinal plants i.e. paneer phool (*Withania coagulans*), Bael (*Aegle marmelos*), Mango (*Mangifera indica*), Custard apple (*Annona reticulata*), Guava (*Psidium guajava*) were taken in account and their α-amylase inhibitory activity was measured using Michaelis-Menten Kinetics. It was found out that *Withania coagulans* showed highest inhibition (i.e. 56.28%) followed by *Aegle marmelos* (20.45%), *Mangifera indica* (14.0%), *Annona reticulata* (19.96%) and *Psidium guajava* (8.86%). Later by comparing the data with Lineweaver-Burk plot, it was found that *Withania coagulans* and *Aegle marmelos* shows Uncompetitive inhibition where V_{max} and K_M of the inhibitor was found to be lesser than that of control enzyme activity. *Annona reticulata* shows

competitive inhibition where, $V_{max}/2$ of the inhibitor and control was found to be same but their K_M was different.

The results of the current study indicated that all of the five plant extracts, showed α-amylase inhibitory activity. The plants may effectively include bioactive compounds showing anti-amylase activity and further structural clarification and characterization methodologies have to be carried out to find out the bioactive components. The inhibition of β-Glucosidase would delay the digestion and absorption of carbohydrates and subsequently suppress postprandial hyperglycemia (Puls W. et al. 1977). This screening of medicinal plants may be a pioneer step for further study on these extracts.

Conclusion: The plant, which can inhibit amylase, can be an effective anti-diabetic drug. In this screening, five medicinal plants were taken in account and their α-amylase inhibitory activity was measured using Michaelis-Menten Kinetics. It was found out that *Withania coagulans* showed highest inhibition (i.e. 56.28%) followed by *Aegle marmelos* (20.45%), *Mangifera indica* (14.0%), *Annona reticulata* (19.96%) and *Psidium guajava* (8.86%). Later the data was compared with Lineweaver-Burk plot. *Withania coagulans* and *Aegle marmelos* showed uncompetitive inhibition where the inhibitor binds near active site after the formation of ES complex and permanently inactivates the enzyme. *Annona reticulata* showed non-competitive inhibition where the inhibitor bind to active site before the formation of ES complex and does not permanently inactivate the enzyme. The enzyme can be activated again after the inhibitor gets removed from it.

The allopathic drugs for the treatment of diabetes mellitus can have several side effects. The ayurvedic medicines can be proved as a relief to reduce side effects and increase efficiency for treatment of type 2 diabetes mellitus. The medicinal plants, which are used directly or indirectly, can reduce these troubles. These are cost effective as compared to the allopathic drugs.

All the plants that were taken in account showed α-amylase inhibitory activity at a considerable amount. Hence, they can be used as an effective substitute to allopathic drugs.

These medicinal plants can have beneficial effects in managing type 2 diabetes mellitus and could be used as effective herbal formulations and can be used in further research.

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Isolation of cellulolytic bacteria for bioethanol production from larvae bitten leaves

Authors & affiliations:
D. Suryawanshi, J. Bendale, V. Ingale and K. J. Patil*
Department of Biochemistry, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
In recent years, the quantity of agricultural waste has been intensifying rapidly all over the world. As a result, the environmental problems and negative impacts of agricultural waste are drawn more and more attention. Therefore, there is a need to adopt proper approaches to reduce and reuse agricultural waste by using eco-friendly approach. The cellulose degrading microorganisms are one of the important candidature, help to reduce agricultural waste rich in cellulose. The present study conducted to isolate cellulose degrading bacteria from leaves infected with larval bites. The cellulolytic bacteria were screened out and isolated from the collected samples by serial dilution method on modified Czapeck (CMC) agar medium and subsequent Congo red assay. Total 05 isolates selected on the basis of cellulose hydrolyzing activity. The isolated organisms are abbreviated as CD1, CD2, CD3 CD4 and CD5. Among, those on basis of ability to degraded cellulose 04 isolates CD1, CD2, CD3 and CD5 are selected for further studies, like effect of pH, temperature, on cellulose degradation. The optimum temperature for cellulose production for CD1, CD2, CD3 and CD5 was found to be 40^0C. The two types of cellulose were determined; among them one was active in acidic pH 3.0 and another in alkaline pH 9.0 for the four isolates. Among the 04 isolates the CD1 was found to be the most active cellulase producer in submerged fermentation. Supplementary, the organisms are applied ethanol production by using corn straw as raw material. The isolates are co-cultured with *Saccharomyces cerevisiae* for simultaneous saccharification and fermentation. Ethanol production was positively tested after five days of incubation with acidified potassium dichromate. The highest productions were observed in CD1.
Keywords: bio-Ethanol, cellulolytic bacteria, Agricultural waste

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Introduction:

Energy is an important factor for the economic development of a nation. One of the greatest challenges for society in the 21st century is to meet the growing demand of energy for transportation, heating and industrial process and to provide raw material for the industry in a sustainable way. Biofuel is globally considered as the sustainable and eco-friendly source of energy to enhance national energy security and to decrease dependence on imported fossil fuel (Sahoo, 2013). One of the most interesting and promising alternatives, in the short-and medium-term perspective, is the second generation bioethanol (B2G) produced from lignocellulosic by products of agricultural, forestry and industrial activities or from urban waste residues (Dantur *et al.,*2015). Biomass can be the best alternative source for the production of fuel and to meet the needs of the present and future generation. Fuels derived from biomass feed stocks popularly known as biofuel have been reported to be an attractive and excellent alternative to conventional fuel as these are neutral and renewable. Biofuel is being considered as a potential liquid fuel due to limited amount of natural resources (Masami *et al*., 2008). These are solid, liquid, or gaseous derivatives from the biomass or the biological feedstock as crops and/or crop residues. Microorganisms are important in conversion of lignocelluloses wastes into valuable products like biofuels produced by fermentation. These microbes produces extracellular cellulose and hence known as cellulolytic microorganis. Although , much effort and resources have been directed to develop industrial scale bioethanol production from lignocellulose.There is still no economically viable industrial production system available for any type of biomass (Limayem and Ricke 2012; Morone and Pandey 2014). The present work concentrates on the isolation of

cellulose-degrading bacteria from larvae bitten leaves. The cellulose-degrading bacteria and yeast applied in consortia for simultaneous saccharification and fermentation of cellulose into ethanol.

Material and Methodology

Collection of Sample Larval bitted leaves were collected from different agricultural crop from different zones of Jalgaon and brought to laboratory.

Isolation of cellulose degrading bacteria from leaves infected with larval bites The larval bitten leaves collected and brought to laboratory and a suspension was prepared and inoculated in a basal salt media ($NaNO_3$ 2.5 g; KH2PO4 2 g; $MgSO_4$ 0.2 g; NaCl 0.2 g; $CaCl_2 \cdot 6H_2O$ 0.1 g in a liter) containing 1% cellulose or CMC for the isolation of cellulolytic bacteria (Joseph et al., 2016). These cultures were incubated for 7 days in a shaker incubator at 37°C at 100 rpm. Bacterial colonies capable of utilizing cellulose as sole source of carbon were isolated on cellulose agar media composed of KH_2PO_4 0.5 g $MgSO_4$ 0.25 g cellulose 2.0 g agar 15 g gelatin 2 g and distilled water lL and at pH 6.8–7.2.

Confirmation of cellulose degrading bacteria Confirmation of cellulose-degrading ability of bacterial isolates performed by (Bashir *et al., 2013)* streaking on the cellulose Congo-Red agar media with the following composition: KH_2PO_4 0.5 g, $MgSO_4$ 0.25 g, cellulose 2 g, agar 15 g, Congo-Red 0.2 g, and gelatin 2 g; distilled water 1 L and at pH 6.8–7.2. The use of Congo-Red as an indicator for cellulose degradation in an agar medium provides the basis for a rapid and sensitive screening test for cellulolytic bacteria. Colonies showing discoloration of Congo-Red were taken as positive cellulose-degrading bacterial colonies.

Enzymatic Assay Endoglucanase activity measured at 60°C in a reaction mixture composed of 0.9 mL of 100 mM l^{-1} sodium acetate buffer pH 5.0 containing 4% of carboxymethyl cellulose (CMC) and 0.1 mL of crude enzyme solution. The reducing groups, expressed as glucose, released were assayed by using the DNS (3,5-dinitrosalicylic acid) method. One unit of endoglucanase activity was defined as the amount of enzyme able to release 1 μmol of reducing sugars per min under the assay conditions (Miller, 1959) (Nyi and Wiwiek 2014).

Effect of pH and temperature on cellulase activity of isolates The effect of pH on the enzyme activity was evaluated by DNSA method, in the pH range from 2.0 to 11.0 using BR buffer. The effect of temperature was assayed incubating the reaction mixture in temperature range from 4 to 80°C in the optimal pH (Sharma et al., 2015) (Gautam et al., 2011).

Bio-ethanol production from corn straw by using cellulolytic microorganism All the isolates were grown in mixed culture using basal salt medium in two different sets, one containing corn straw powder and the other containing 4% cellulose powder as substrate for production of cellulolytic enzyme and to initiate saccharification process. Culture incubated at 37°C with mixing at 100 rpm for 3 days. After completion of 5 days of incubation, the above culture broth was conditioned for co-culturing of Saccharomyces cerevisae by addition of filter-sterilized salt solution (KH_2PO_4 0.4 g, $MgSO_4$ 0.02 g, $CaCO_3$ 0.05 g, and NaCl 0.01 g to 1 L culture broth). The simultaneous saccharification and fermentation was carried out at 27°C for 5 days in stationary condition (Dey 2018). At the end of incubation, the culture broth was qualitatively tested for alcohol production by potassium dichromate assay (Poznanski 1928).

Result and Discussion

Isolation of cellulose degrading bacteria from leaves infected with larval bites: Leaves are washed in sterile distilled water and suspention inoculated on minimal salt medium containing cellulose after 7 days of incubation 4 microorganisms are isolated and auxinic cultures are obtained by strike plate technique. Total five isolates are obtained and checked for cellulose degradation by Congo red method (figure 1).

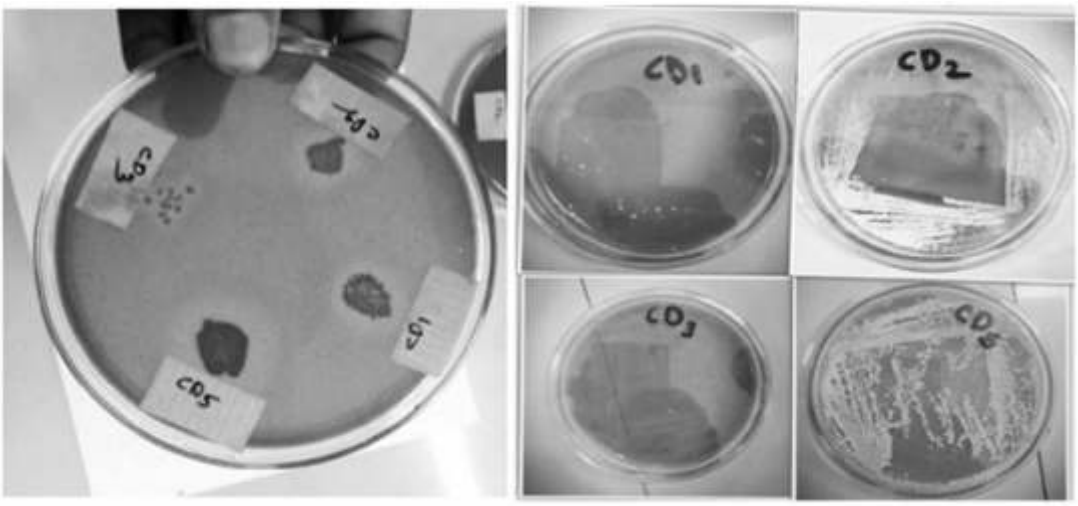

Figure 1 Zone of clearance on cellulose Congo Red agar plates for all isolates after 48 hrs of incubation. The formation of clearing zone around the colonies confirms the secretion of extracellular cellulase.

Among five four are selected for further studies on basis of zone of hydrolysis. The section done based on time taken for appearance of and colony morphology. These results indicate the bacteria in the larval gut may fall with fesses on leaves. Cellulose in the media was hydrolysed due to a cellulolytic enzymes produced by the bacteria. The hydrolysis process produced clear zone in CMC media due to the reaction between cango red with B-1,4glycocidic contained in cellulose polymer.

Abbreviations given to the isolates The isolated strains therefore these bacterial strains putatively abbreviated as CD1, CD2, CD3, and CD5.

Effect of pH on cellulase activity of isolates The effect of cellulose activity of all the isolates shows optimum activity in acidic and alkaline range. Which conclude that there may be the presence of isoenzymes having optimum activity at pH 3.0 and pH 9.0 revealed in figure 2. Amongst the isolates CD1 has highest enzyme activity and lowest shown by CD3. The results confirmed that all the tested strains produce isoenzymes among them one is active at pH 3.0 and another at pH 9.0.

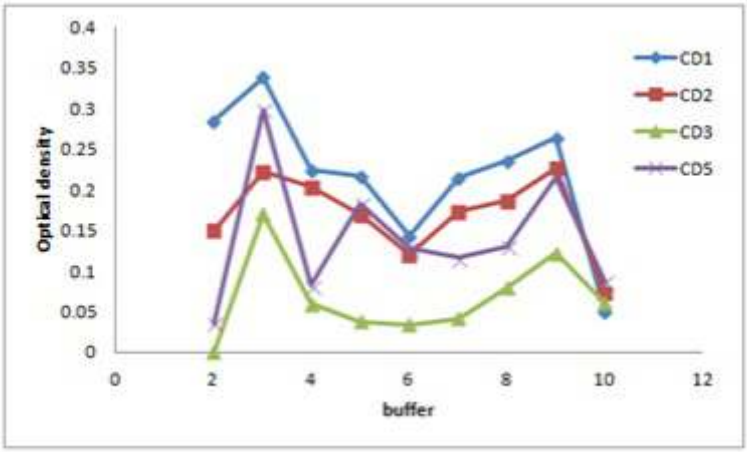

Figure 2 Effect of pH on cellulase activity of crude extract of isolates

Similar results are Optimum endoglucanase activity was observed in the acidic range at pH 4.8 and second peak was observed at 6.8 which is pH of insect gut. After pH 6.0 enzyme activities

was declined abruptly (Rehman, 2009). However, optimum pH values of 4.5-8.0 have been reported for different microbial cellulase (Bakare *et al.,* 2005; Immanuel *et al*., 2007; Dutta *et al*., 2008).

Effect of Temperature on cellolase activity of isolates

The effect of cellulase activity of all the isolates shows optimum activity 37^{0}C. Amongst the isolates CD1 has highest enzyme activity and lowest shown by CD5 shown in figure 3. Somewhat similar results were obtained by Joseph et.al. 2016 isolated *Bacillus cereus* and *Bacillus subtilis* has maximum temperature is 30 and 40°C respectively.

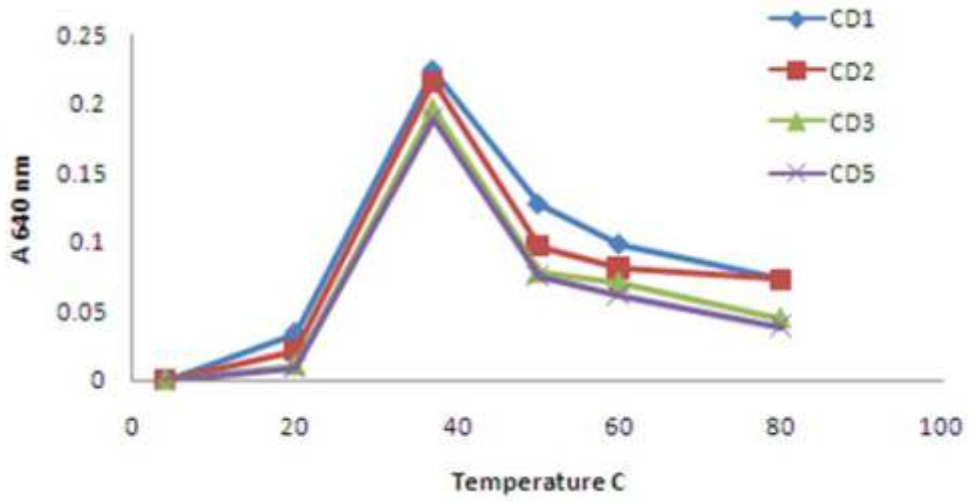

Figure 3 Effect of Temperature on cellulase activity of crude extract of isolates

Bio-ethanol production from corn straw by using cellulolytic microorganism

The isolates are inoculated in minimal salt medium and corn straw or pure cellulose as the cellulose substrate to release residual sugar. After 5 days of incubation *Saccharomycescerevisiae* added for ethanol production. The results conclude in case of corn straw the highest ethanol production detected in CD1 and lowest was found in CD5 publicized in figure 4. Whereas, in pure cellulose highest yield observed in CD2 and lowest in CD5. The variation in results may be due to the other chemical constituents of corn straw.

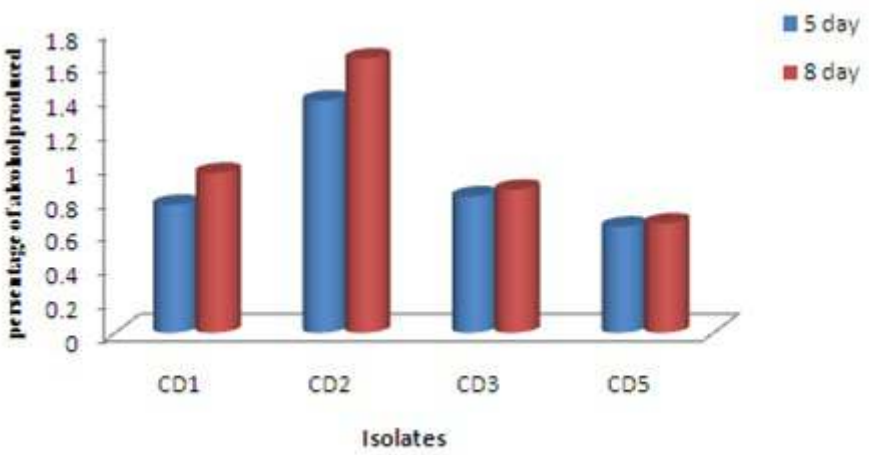

Figure 4 Ethanol production by isolates from cellulose

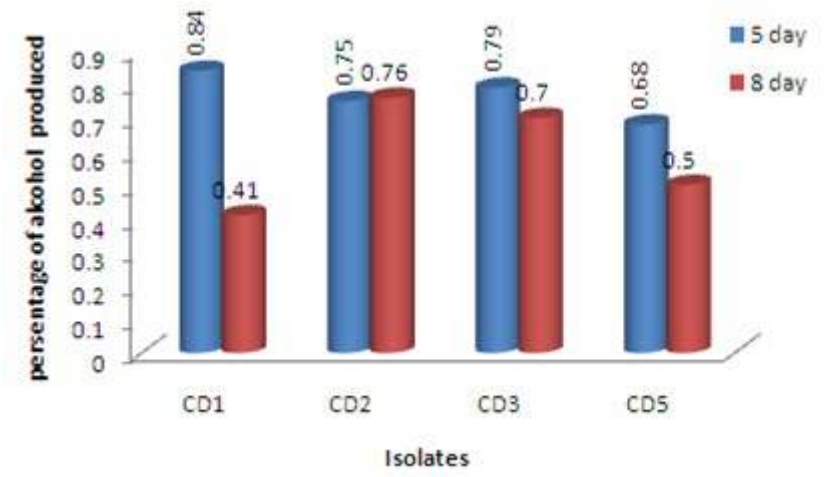

Figure 5 Ethanol production by isolates from corn straw

Conclusion:

Total five Cellulose degrading bacteria are isolated form larval beaten leaves and abbreviated as CD1; CD2; CD3, CD4 and CD5. Amongst, five four are selected for cellulose degradation capabilities. The pH range for isolates are ranges from 2.0 to 10.0 pH, were highest activity was shown in two different pH one in acidic 3.0 and another in 9.0 in alkaline pH. Results imply that the isolated organisms contain isoenzyme. Whereas, the optimum temperatures for all the isolates maximum at 37^0C. Further, the isolated organisms are applied for bioethanol production in consortia by using *Saccharomy cescerevisiae* by using purified cellulose and corn straw. In case of corn straw the highest ethanol produced by CD1 and lowest was found in CD5. Whereas, in pure cellulose highest yield observed in CD2 and lowest in CD5.

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Isolation of rhizobium and azatobacter from jalgaon region and its application as biofertilizer

Authors & affiliations:
K. Patil, K. Pawar, B. Patil, C. Kadam , K. J. Patil*
Department of Biochemistry, M. J. College, Jalgaon, (M.S.) India

ABSTRACT:
Soil depletion is one of the alarming situation occurs due to the extensive exploration of chemical fertilizer to increase fertility of soil. In action to that this leads to poor crop yields and causes soil depletion. Since, to deal with such situation it's necessary to work on soil management. To cope with this problem, bio-fertilizers are efficient candidature. The microorganism (plant growth regulators) are play the vital role in fixing nitrogen, solubilising of phosphorus and to reduce chemical fertilizer and reducing the macro nutrition in soil The main aim of this project is isolation of Azatobacter and Rhizobium from Jalgaon region and check the efficacy of isolated organism with phosphate and potassium solubilizing microorganisms. In present work, Azatobacter and Rhizobium are isolated from Jalgaon region and compared with previously isolated Phosphate and potassium solubilizing microorganisms. After 18 days, the different growth parameters of the individual plants were measured which showed a 61.058 to 313.46 % increase in relative index of individual bio-fertilizer treatment and 93.269 to 319.52 % increase in relative index combined treatment of plants compared to the control ones. Among the isolate the Rhizobium shows maximum results as compared to isolated stains individual and in M2BC2, M3C and Rhizobium in consortia. The results have successfully confirmed the efficacy of azatobacter and isolated rhizoidal strains with multiple beneficial characteristics on vigor of maize seedlings under controlled condition.
Keywords: bio-fertilizer, plant growth regulator, Rhizobium and Azatobacter

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Introduction:
The nitrogen phosphorus, potassium and other elements are play essential role of nutrients for increasing the productivity of land. Bio-fertilizers is an important association with other soil microbes and help in nutrient supply. They improve soil properties and sustaining soil fertility lead to soil enrichment as compared to the chemical fertilizer, bio-fertilizers are safer with reduced environmental damage, has more targeted activity in smaller quantities. In India, the role of photosynthetic bacteria on tropical soils was shown to be highly significant, especially in association with other asymbiotic nitrogen fixers like *Azotobacter*. Some of these organisms are known to synthesize growth-promoting substances and antibiotics and as such are important in enhancing plant growth and in combating plant diseases, though the amount of nitrogen fixed may not be large. Recent researches have brought us to an understanding of some of the difficult problems of biological nitrogen fixation through the use of powerful tools of microbial genetics. Plasmids offer a tremendous potentiality as vehicles for 'nif gene transfer to higher plants. There is still a long way to go in this exercise with an ultimate aim to obtain plants which could grow independently of increasingly expensive nitrogen fertilizers. The present work, focused on isolation of plant growth stimulating organisms including Azotobacter and Rhizobium and preparation of biofertilizer in consortia with phosphate and potassium solubilizing microorganisms.

Materials and Methodology:
Collection of Sample Soil samples were collected from different region of different zones of Jalgaon area such as “Mehrun Pimprala and Paldhi”. The sample were collected in bags and transported to lab for further process.
Adaptation and Enrichment Soil samples collected from different regions of Jalgaon were kept at room temperature for one week. Withdraw 1 gm of soil sample and inoculate in 90 ml liquid media containing 0.95% glucose, 0.045% yeast extract and 0.45% Mica make final volume to 100ml and

incubated at room temperature at 120 rpm for 7 days.

Isolation of Potassium Solubilizing Bacteria The enriched soil samples were serial diluted up to 10^{-12} and inoculated on modified Aleksandrov agar medium, plates were incubated at 27±20°C for 7 days. Potassium solubilizing colonies were selected from 10^{-8}, 10^{-9} and 10^{-10} dilution containing plates. Isolation of colonies was done on the basis of zone formed around the colonies only those isolates were selected whose shows zone (Verma et al., 2016 and Shanware *et al., 2014)*

Phosphorus solubilizing bacteria The enriched soil samples were serial diluted up to 10 and inoculated on modified Pikovskayas Agar Medium; plates were incubated at 27±2°C for 7 days. Isolation of colonies was done on the basis of zone formed around the colonies only those isolates were selected whose shows zone (Haile et al., 2016).

Isolation of Rhizobium Isolation of Rhizobium was done using yeast extract mannitol agar (YEMA) as described by Hamza and Alebejo (2017). In this, healthy, unbroken, firm and pink nodules were selected for the isolation. They were washed under tap water to remove adhering mud and soil particles, after which they were treated carefully with 5% hydrogen peroxide for surface sterilization. A suspension was made of the crushed nodules, plated on YEMA medium containing 1% Congo-red dye and incubated at 28±10C for 24 hours. Growth on YEMA plate was observed after the said incubation period.

Isolation of Azatobacter The azatobacter are isolated by spreading soli suspension on Ashby's medium and the plates for even spreading of inoculums and incubate at 28°C for 3-4 days (Bhaduri et al., 2016).

Results and Discussions

In the present study, Samples are collected from different sites of Jalgaon city eight bacterial strains having characteristic of nitrogen fixation, phosphate solubilization and potassium solubilization. The details of their abbreviation and peculiarities are given in table 1

Table 1 List of isolates and their abbreviation with peculiarities

Sample	Medium	Location	Peculiarities
M2BC1	Pikovskayas Agar	Meharun	Phosphate Solubilizing
M2BC2	Pikovskayas Agar	Meharun	Phosphate Solubilizing
M1D	Jensen's Medium	Paldhi	Nitrogen Fixing
M3C	Aleksandrow Agar	Pimprala	Potassium Solubilizing
M3B	Aleksandrow Agar	Meharun	Potassium Solubilizing
M2C	Pikovskayas Agar	Pimprala	Phosphate Solubilizing
Rhizobium	YEMA	Yamuna Nagar	Nitrogen Fixing
Azatobacter	Ashby's Agar	M.J.College	Nitrogen Fixing

Azatobacter and rhizobium are isolated from jalgaon region and check the efficacy of isolated orgnism with phosphate and potassium solubilizing microorganism.Compared the growing ability of azatobacter and rhizobium with previously isolated Phosphate and potassium solubilizing microorganisms. After 18days,the different growth parameters of the indiviual plants were measured which showed a 61.058 to 313.46% increasing relative index of individual biofertilizer treatment and 93.269 to 319.52%increasing relative index of combined treatments. Among the isolate the Azatobacter shows maximum results as compared to isolated stains indiviual and in M2BC2,M3C And Rhizobium in consortia.

Figure 1 Plant growth in pot assay of selected microorganism

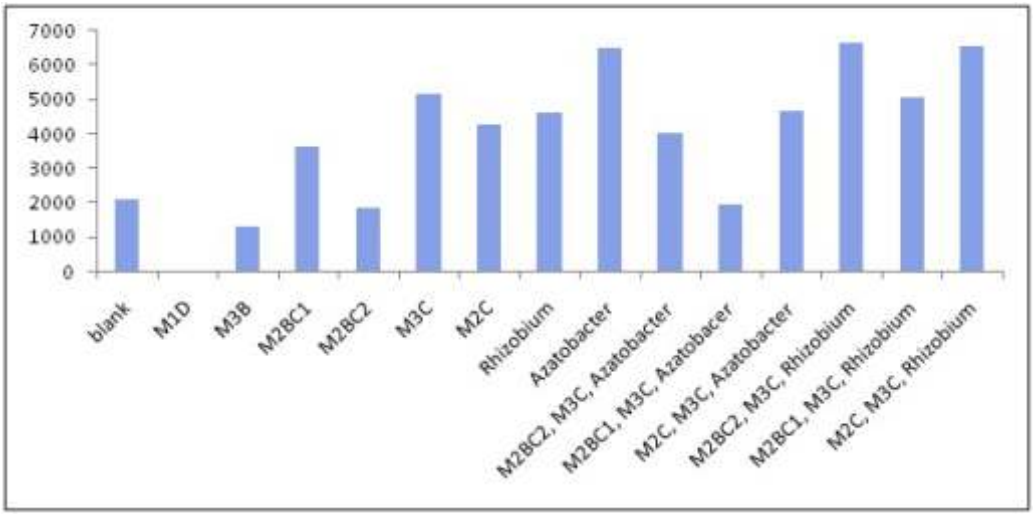

Figure 2 Vigor index of isoaled microorganisms

Table 2 The plant growth stimulating ability of tested organims

	Total					Average						
	plant length	shoot length	root length	plant weight	seeds germinated	plant length	shoot length	root length	plant weight	% of seed germination	vigor index	relative index
Blank	104	83.5	20.5	1.86	4	26	20.875	5.125	0.465	80	2080	100
M3B	63.5	40.5	23	2.27	1	63.5	40.5	23	2.27	20	1270	61.05769
M2BC1	182	69.5	112.5	6.65	4	45.5	17.375	28.125	1.6625	80	3640	175
M2BC2	91.2	37	54.2	2.76	4	22.8	9.25	13.55	0.69	80	1824	87.69231
M3C	257.7	129.2	128.5	8.36	4	64.425	32.3	32.125	2.09	80	5154	247.7885
M2C	213.7	120.5	94.2	8.16	4	53.425	30.125	23.55	2.04	80	4274	205.4808
Rhizobium	231	143.5	88.3	9.25	5	46.2	28.7	17.66	1.85	100	4620	222.1154
Azatobacter	326	165	131	11.01	4	81.5	41.25	32.75	2.7525	80	6520	313.4615
M2BC2, M3C, Azatobacter	201.9	117	119.5	8.66	4	50.475	29.25	29.875	2.165	80	4038	194.1346
M2BC1, M3C, Azotobacter	97	61.5	35.5	3.33	2	48.5	30.75	17.75	1.665	40	1940	93.26923
M2C, M3C, Azatobacter	233.1	127	109.1	7.08	5	46.62	25.4	21.82	1.416	100	4662	224.1346
M2BC2, M3C, Rhizobium	332.3	160.1	172.2	15.59	5	66.46	32.02	34.44	3.118	100	6646	319.5192
M2BC1, M3C, Rhizobium	252.25	127.25	125	11.15	4	63.0625	31.8125	31.25	2.7875	80	5045	242.5481
M2C, M3C, Rhizobium	326.8	151.4	175.4	13.52	5	65.36	30.28	35.08	2.704	100	6536	314.2308

Conclusion

The Rhizobium and Azotobacter was isolated from root nodules of leguminous plants from Jalgaon area. The individual Azatobacter found as potential biofertilizer amongst the tested isolates. Whereas the consortia of M2BC2, M3C, and Rhizobium shows maximum plant growth stimulating ability as compared to other combinations. From current research it was found that the consortia of phosphate, potassium solubilizing and nitrogen fixing microorganisms are efficient as compared to individual microorganisms.

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Development of Eco-friendly bio-fertilizer by using feather degrading microorganism

Authors & affiliations:
U. M. Patil, V. S. Chavhan, P. D.Umbarkar and P.S. Patil*
Department of Biotechnology, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
The aim of this study was isolate and screen feather degrading organism from dumping site and make eco-friendly bio-fertilizer by using thus strain. The soil sample was collected and dilute, plated on specific agar media and different colonies of bacteria was isolated. After that secondary screening was perform by using casein agar media and zone of clearance around the colonies was observed. The bacterial strain No-04 was give higher zone of clearance around the colonies, thus strain was selected for further studies. The strain was enriched by Nutrient agar medium and this culture was added in soil with feather and wheat husk this culture was incubated for some days after that compost was prepared. The field experiment was conducted with up to some days comparatively different compost (cow-dunk, urea, soil use as a control) for growth of plant. The result indicates that the impact of feather treated plant showed maximum growth than control, cow-dunk and urea. This is result using Eco-friendly bio-fertilizer feather compost. The proposed ecologically safe method and economically viable, the application on agriculture field.

Keywords: Feather degradation, keratinase enzymes, bacillus species, feather compost, eco-friendly bio-fertilizer.

Introduction:

Feathers are produce in large amounts as a waste by-product of poultry processing plant (Sangali and Brandeli,2000).A current value added use for feathers is the conversion to feather meal, a digestible dietary protein for animal feed, using physical and chemical treat-ments(Mortiz and Latshaw,2001;Anbu et al,2005) In poultry birds, keratin form a major component of the epidermis and its appendages viz. hair, feathers, nails, horns, hoofs, scales and wool. Structural confirmation of keratinase have been classified into (alpha helix of hair and wool, beta helix of feather). Feather is pure keratin protein and is insoluble and hard to degrade due to highly rigid structure rendered by extensive disulphide bond and cross-linkages. The keratin chain is insoluble, high stable structure tightly packed in the alpha helix and beta-sheets into super coiled polypeptide chain (Onifade et al 1998).

Worldwide 24 billion chickens are killed annually and around 8.5 billion tonnes of poultry feather are produced. According to a recent report in leading news paper, India's contribution alone is 350 million tonnes. The poultry feathers are dumped, used for land filling, incinerated or buried, which involves problems in storage, handling, emissions and control and ash disposal. Discarded feather also causes. Various human ailments including; chlorosis, mycop-lasmosis(Williams et al 1991).Preparation of bio-fertilizer using chicken feather wastes is attracting the focus of many research scientists. The plant growth promoting activity of protein hydrolysates could also be effectively applied in agriculture. Thus, microbial degradation of feather represents and alter-native for development of slow release nitrogen fertilizers. The degraded product of chicken feathers could generate appropriate amount of tryptophan which can serve as the key source of IAA synthesis. The composting method of producing a stabilized organic material that can be used as a source of nutrient and soil conditioner.(Raja PrabuMasilamani, 2017).

The aim of this study is to isolate and identify karatinolytic bacteria showing high feather de-gradation ability. Preparation of feather compost by using karatinolytic isolates and its appli-cation in plant growth stimulation..

Materials and Methodology:

Materials:

All Media ingredients and chemical reagents used for experiment were of analytical grade, purchased from Himedia Ltd, Mumbai.

Collection of sample:

The feather sample was collect from 4 different dumping sites of the Jalgaon, Maharashtra. And wash with distilled water later wash with 70% ethanol to removed unwanted microor-ganisms of feather sample. Specific salt Media components were use for growth of only keratinase producing microorganism.

Methods:

Primary screening

1. The feathers and soil sample was collected from 4 different dumping sites.
2. Feathers and soil was incubated in a specific salt medium for 4 days on a rotary shaker.
3. Another feathers was wash with tap water and then with 70% ethanol and allow to air dry.
4. After autoclaved the media was poured in a petri plates and allow to solidify.
5. The suspension from previously incubated flask was taken and serially diluted by different Dilutions.
6. Each plate was then spread with 0.1ml of suspension and incubated for 48hrs.
7. After incubation colonies was developed around the crushed feathers, the developed co-lonies was then isolated and sub cultured for secondary screening.

Secondary screening

1. Casein media was prepared and autoclaved it, after solidify the media poured in Petri plates.
2. The bacterial strain No-4 shows higher zone of clearance ,then subculture it and stored for ferther studies

Preparation of Bio-fertilizer

1. The nutrient broth was prepared and autoclaved. After the autoclaved isolated bacteria was inoculated on the rotary shaker for the 4-5 days at room temperature.
2. The soil sample and wheat husk was taken and mix properly. Add feathers in the propor-tion of wheat husk1;1 ratio to make compost.
3. The previously incubated nutrient broth was added in the soil compost.
4. The water was sprinkled on the compost to maintain their moisture content.
5. The pH and temperature was calculated after one day interval till up to 15 days.

Method of plant sawing

1. After preparation of compost black gram was saw in the compost, soil, urea, cow-dunk (used as control) for comparatively study.
2. After the 8 days the height of the plant, root, shoot was measured.
3. The black gram shows well result in feather compost as compared to soil, urea, cow dunk.

Results and Discussions:

In the present study, Samples was collected from different sites of Jalgaon city and a potential keratinolytic bacteria was isolated and partially identified by studying colony characteristics and biochemical test. The details of their colony characteristic and biochemical test are given in table 1 and 2.

Figure 1 Primary and secondary screening of isolated bacteria

Table 1 Colony character of isolated bacteria

Shape	Circular
Elevation	Raised
Margin	Entire
Opacity of colony	Transparent
Chromogenesis	Cream Color
Consistency or texture	Smooth Buttery
Size	24mm
Gram character	Gram positive
Shape	Cocci

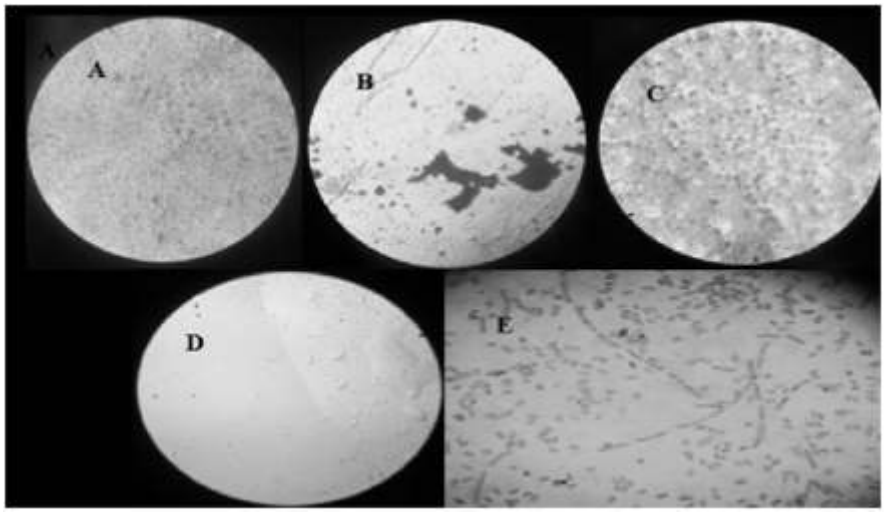

Figure 2 A-Gram stating, B-Capsule staning, C-Flagella staining ,D-Motility testing , E-Endospore staining

Table 2 Biochemical characterization of isolates

Test	Result	Test	Result
MR	Negative	Catalase test	Positive
VP	Negative	Oxidase test	Negative
Indol	Negative	Gelatin test	Negative
Capsule staining	Negative	Glucose	Positive
Endospore staining	Positive	Sucrose	Positive
Fagellastainng	Positive	Lactose	Positive
Starch hydrolysis	Negative	Maltose	Positive
Casein hydrolysis test	Positive	Mobility	Positive
TSI	Negative		

Further the feather compost is prepared by degrading feathers in optimum conditions and applied for plant growth efficacy. The effect of feather compost is compared with standard control containing only soil, cow dung, and urea. The results are discussed in as follows. The height of the plant, length of root, shoots and weight of the plant was recorded in table 4.

Figure 3 Plant generated after 7 days.

Table 4 Effect of fertilizer and isolated bio-fertilizers on plant growth

Fertilizer	Number of seed ger-of minated	Height of plant cm	No. of leaf	Weight of plant gm	Length of root cm
Control	4	31.5	3	0.68	8.7
only soil		25.2	2	0.55	7.8
		26.1	2	0.65	8.2
		31.3	2	0.59	10.3
Cow dung	4	25.4	3	0.63	5.3
		23.7	2	0.51	4.9
		24.6	2	o.53	5.6
		23.2	3	0.54	6.1
Urea and soil	4	29.5	3	o.55	7.8
		28.6	2	o.44	2.0
		24.6	2	0.36	2.3
		29	2	0.95	2.9
Biofertilizer	4	33	3	0.73	12.5
		21.5	2	0.55	7.8
		32.2	3	0.69	11.5
		31.3	2	o.59	10.3

Table 5Vigour index and relative index of fertilizers on plant growth

	Seed germinated	Percentage of germination	Average plant height cm	Vigour index	Relative index
Blank	4	100	22.82	2282	109.71
Cow dung	4	100	19.38	1938	93.17
Urea	4	100	22.34	2234	107.40
Feathercompost	4	100	23.6	2360	113.46

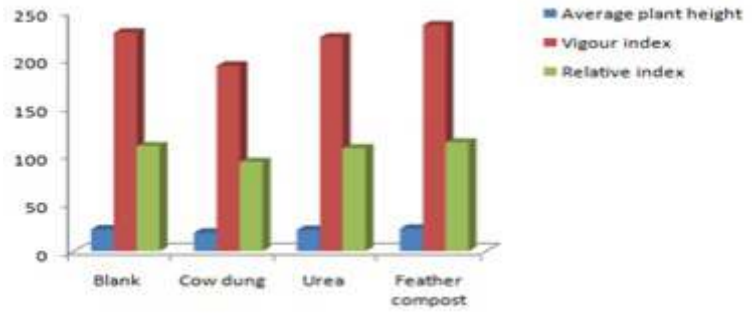

Fig:Residual plot for growth of plant in cow dung,urea,feather compost After 7 days.

Conclusion:

A potential Feather degrading bacteria was isolated from Jalgaon area. The feather are degraded by isolated bacteria and compost was prepared. The feather compost was applied for plant growth stimulation activity and compared with cow dung and urea. The vigour index and relative index of feather compost is 2360 and 113.46 respectively is greater than urea and cow dung.

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Isolation and optimization of polythene degrading microorganism from dumping soil

Authors & affiliations:
S. J. Katole, M. G. Thakare and L. D. Dhake*
Department of Biotechnology, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Plastics are organic polymers of high molecular mass. In last few decades the uses of plastic increases tremendously for various purposes such as packaging, transportation, industry and agriculture in rural as well as urban areas and at present the threat of plastic pollution is on the boom. Jalgaon is also knows for many renowned industries and having a population more than 460,228 lack as per 2011 data. The domestic and industrial waste may create tons of plastic pollution.
Objectives: This study aims to isolate and identify the bacteria and optimize the growth rate by varies site present in Jalgaon Dumpsite that have the potential to degrade plastic components such as Polyethylene Glycol (PEG) and Polyethylene films.
Methodology: Partially degraded samples were obtained from Jalgaon dumpsite and cultured microorganisms were morphologically and biochemically studied and evaluated for their plastic degrading capabilities.
Results: Seven PEG degrading microorganismwere isolated from dumpsitesamples. Thesemicroorganisms' plastic degrading capabilities were confirmed through weight loss of polythene films after incubation. The microorganism are named as M1 to M7, among them
Conclusion: The microorganisms isolated from Jalgaon Dumpsite have the potential to degrade plastics, particularly PEG and polyethylene films. Now optimize the media for plastic degradation by pH adjusting, temperature distribution etc work has done.
Key words: plastic degrading bacteria, biodegradation, identify name of bacteria, optimize growth, Jalgaon

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Introduction:

Plastics are organic polymers of high molecular mass. The name plastic is derived from the Greek word *'plastikos'* which means "capable of being shaped or molded" (Joel 1995).The plastic is made up of carbon, hydrogen, silicon, oxygen, chloride and nitrogen. For extraction of the basic materials of plastics oil, coal and natural gas are used (Seymour 1989). In last few decades the uses of plastic increases tremendouslyfor various purposes such as packaging, transportation, industry and agriculture in rural as well as urban areas and at present the threat of plastic pollution is on the boom. The extensive utilization of plastics is due to their stability, durability, favorable mechanical and thermal properties. The vast usage and incredible application of plastic in daily life immensely, since it is quit impossible to ban on plastic usage. Although, many biodegradable plastic are developed to full fill the demand of plastic. Excluding, due to expensive and unbearable nature, they are not enough to complete the demand of society. Hence, plastic recalcitrant and remain inert to degradation and deterioration leading to their accumulation in the environment, and therefore creating serious environmental problems. In this context, it is necessary to develop environment friendly approach to reduce plastic pollution. Microorganisms are seams to play a significant role in environment maintenance. The various microorganisms are reported for plastic degradation. The microorganism degraded the plastic by mineralization or biotransformation by synthesizing extracellular or intercellular enzymes. But efficient polythene degrading microbe is still need to be screened from all the sources. On basis of previous research microbial species are associated with the plastic or polythene degradation are Bacillus subtilis, A. niger, Aspergillus nidulance, Aspergillus flavus, Aspregillus glaucus, Penicillium, Pseudomonas, Staphylococcus aureus, Streptococcus lactis, Proteus vulgaris, Micrococcus were found to degrade polythene and plastic efficiently (Abrusci et al.

2011; Aswale and Ade 2008; Kathiresan 2003; Nanda et al. 2010; Reddy 2008), Aspergillus sp., and Lysinibacillus sp., (Esmaeili et al. 2013), Aspergillus clavatus (Anudurga Gajendiran et.al., 2016), A. oryzae (Indumathil and Gayathri, 2016), *Kocuria kristinae, Dermacoccus nishinomiyaensis, Pseudomonas stutzeri,* and *Acinetobacter haemolyticus (*Nicole R. Bolo, 2015). In-vitro biodegradation of plastic waste through microbial strains could offer a solution to this problem (Das, M.P., 2013). The present study aims to isolate microorganism from dumped soil area of Jalgaon region. Further, the isolation of the potential plastic degrading microorganisms

Material and Methods:

Sample Collection Partially degraded plastic bags were collected from three different garbage dumping regions in and around Jalgaon (Maharashtra),

Enrichment Of Microorganisms The partially degraded plastic bags are collected and inoculated in Minimal salt medium (MSM). MSM contains the following constitutions in 1000ml distilled water (K2HPO4, 1g; KH2PO4, 0.2g; NaCl, 1g; CaCl2.2H2O, 0.002g; (NH4)2SO, 1g; MgSO4.7H2O, 0.5g; CuSO4.5H2O, 0.001g; ZnSO4.7H2O, 0.001g; MnSO4.H2O, 0.001g and FeSO4.7H2O, 0.01g). 100mg of PEG (poly ethylene glycol) powder was added were incubated at room temperature for 1 week.

Isolation And Screening Of Microorganism Polyethylene Degrading Microorganisms The enriched microbial culture was serially diluted and spread on minimal salt medium contains 0.1% PEG and incubated at 37^0C for 48-96 hours. Further the pure organism are isolated by streak plate technique and incubated at 37^0C for 48-96 hours. The isolated microorganisms were screened based on their ability to utilize polyethylene as sole carbon source. They were microscopic examined and biochemical analyzed.

Microbial Degradation of Polyethylene Waste under Laboratory Conditions Lamination Polyethylene sheets were cut and initially weighed and washed with sterile distilled water. They were then soaked in crude black phenol for 30 min and dried in laminar air flow for 15 min. This polyethylene film was inserted into 50ml of mineral salt medium aseptically 2 loops of each bacterial isolates were inoculated into separate flasks containing mineral salt medium and polyethylene film. The flasks were incubated at 37°C in an incubator shaker for a month. After incubation the polyethylene films were washed with sterile distilled water and then sprayed with alcohol, air dried and weighed (final weight). Percentage degradation of polyethylene was determined by taking initial weight and final weight.

pH, temperature distribution: The PEG media is prepared and the set its different pH as pH 3,pH4, pH5, pH6, pH7, pH8.and incubate the isolated bacteria in them and incubated it at 37^0 c for 24 hrs and observed high growth in which culture.

Media cultivation (Hi-media used):

NH4NO3 0.1, MgSO4 0.2, K2HpO4 1 gm, cacl2 0.1 gm, kcl 0.15, yeast extract 0.1, feso4 1 mg, znso4 1 mg, mnso4 1 mg, with using plastic component PEG having ability of plastic degradation. The plastic is inoculated in hi-media before adding the plastic piece gating the weight as initial and incubates it for 25 days.

Growth factor increases (nitrogenous compound containing additives):

The MSM media compound additives with nitrogen adding source replace by ammonium sulphate to peptone, gelatin, casin, and urea and observed the proper and sustain growth in media and shows higher turbidity.

Different type of plastic degradative level checks by bacteria:

The ordinary plastics are present in surrounding so it chose by hardness and color (white, ordinar black, pink, hard black plastic). That plastic incubate in MSM media with taking the plastic initial weight and incubate for 30 days and observed the weight loss. Because of this experiment it should

recognize that different type of thickness plastic is also can be degraded.

Result and Discussion:

Polyethylene degrading bacterial strains were isolated from dumping sites of Jalgaon city. These isolates were capable of growing on a carbon free synthetic medium containing polyethylene films as sole carbon source which was seen by succeeding sub-culturing followed by plating on medium containingpolyethylene glycol. Eight bacterial strains designated as M1 to M8. All the isolated bacterial strains were studiedfor colony morphology, and Gram staining character is summarized in Table 1 and 2.

Table 1 Colony characteristic of plastic degrading isolates

Test/ of isolates	Shape	Configuration	Margin	Elevation	Surface	Pigment	Opacity
M1	Coccus	Circular	Filamentous	Convex	Smooth	buttery white	Shiny
M2	Coccus	Circular	Punciform	Convex	Smooth	buttery	Translucent
M3	Coccus	Circular	Punciform	Convex	Smooth	pink	Opaque
M4	Coccus	Circular	Filamentous	Convex	Smooth	buttery	Opaque
M5	Coccus	Circular	Undulate	Flat	Smooth	buttery	Opaque
M6	Coccus	Circular	Entire	Convex	Smooth	buttery	Opaque
M7	Coccus	Circular	Punciform	Convex	Smooth	buttery white	opaque
M8	Coccus	Circular	Entire	Convex	Smooth	blackish	Opaque

Table 2Biochemical characteristic of plastic degrading isolates

Test/ No of isolates		M1	M2	M3	M4	M5	M6	M7	M8
Sugar fermentation									
Glucose	Acid	-	+	+	-	-	-	-	+
	Gas	-	-	-	-	-	-	-	-
Maltose	Acid	-	-	+	-	-	-	-	-
	Gas	-	-	-	-	-	-	-	-
Sucrose	Acid	-	-	-	-	-	-	-	+
	Gas	-	-	-	-	-	-	-	-
Cellulose	Acid	-	-	-	-	-	-	-	-
	Gas	-	-	-	-	-	-	-	-
Starch	Acid	+	+	+	+	+	+	+	+
	Gas	-	-	-	-	-	-	-	-
Indol test		-	-	-	-	-	-	-	-
MR test		+	+	+	+	+	+	+	+
VP test		+	+	+	+	+	+	+	+
citrate utilization		+	+	+	+	+	+	+	+
H2S gas production		-	-	-	-	-	-	-	-
amylase production		+	+	+	+	+	+	+	+
Gelatin hydrolysis									
ureaase production		+	+	+	+	+	+	+	+
Catalase production		+	+	+	+	+	+	+	+
peroxidase production		+	+	+	+	+	+	+	+
Grams character		-	-	-	-	-	-	-	-
Endospore character		+	-	-	-	-	-	-	-

3.2 Determination of dry weight of residual polyethylene by MSM media and HI-media:

After 20 days of incubation period, the percentage of weight reduction was estimated and it is shown in figure 2. Polyethylene films incubated with bacterial isolates. Highest degradation shown by M1 and lowest by M7 showed weight loss of 0.0429 -0.0426 % in S1 strain respectively. While similarly all the isolates are incubated with polythene in HI-media. The results are different than percentage of degradation, where highest degradation shown by S1 and lowest by S8 showed weight loss of 0.5858 to 0.5855 respectively. The variation in results may be due to the varied chemical composition of media. Nevertheless, in both the medium the degradation found to be greater than the weight loss

obtained in control. Therefore, the observed percentage weight loss of polyethylene strips incubated on bacterial isolates was not as result of chemicals in the medium but because of a biological process.

pH and temperature distribution:
The PEG media is prepared and set it at pH at like pH 3, pH4, pH 5, pH6, pH7, pH8 respectively and inoculated the bacteria in each strain and incubate for 24 Hrs. and observed the growth in different strain. After incubation the greatest growth observed in pH 6 and pH7 and high optical density 0.014 - 0.029 respectively. By this experiment it concluded the bacteria stable at pH 6 to pH 7.

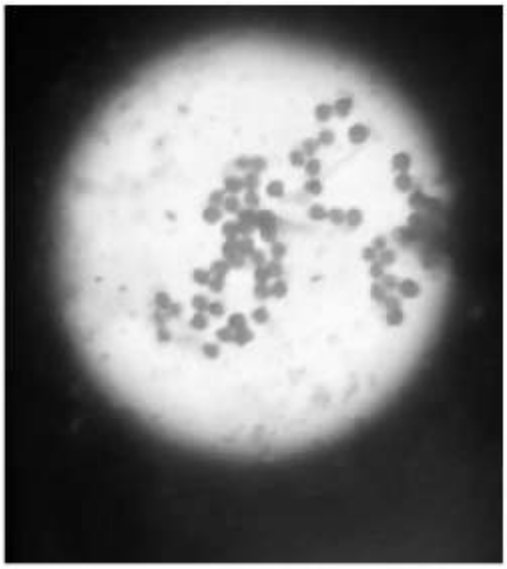
Temperature distribution:
Same as the PEG media prepared and inoculated bacteria and put at as per respective temperature such as 5^0c, room temperature, 37^0c, and 45^0c. Incubate it for 24 Hrs. observed result. The high growth observed in room temperature and 37^0c as 0.041-0.048 respectively.

Check bacterial activity on soil and crop:
The serial suspension is inoculated in soil which contains wheat grains and observed the proper growth after 4-5 days. After germination it indicted as the growth is normal and no effect on soil and its pH. By this experiment we concluded as there is no effect of suspension on living organism as well as crops

Identification of bacteria name:

Gram character	Gram negative
Shape	Coccus
Color	Pink
Motility	Non motile
Surface	Smooth
pH	6.5-7.5
Temperature	27-45

According to following table the isolate bacteria strain is gram negative and from Group X1 member and the name is *Neisseria* it is concluded and it belongs to *Neisseria* genus.



Result and bacterial image

Research conclusion:

From total 8 isolates, were screened based on their ability to utilize polyethylene as carbon source. The isolated organisms, showed weight loss in plastic ranging from 0**.0429% to 0.0426%** in minimal salt medium in one month. The weight loss is the evidence for the polyethylene degradation. The results of plastic degradation show that, the isolates posses' potential to degrade polythene. Microbes have potential to degrade polythene although it requires media standardization and the optimum factors for plastic degradation to achieve maximum degradation in less duration. Hence this method can be used widely for biodegradation and serve as a promising tool for the elimination of polythene from the environment.by changing the media from MSM to Hi-media that also shows the degradation as **0.5858% to 0.5855%** one month respectively.

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Isolation of chitosan from chitin and its applications

Authors & affiliations:
S. D. Mahajan, S. C. Rade, H. R. Patil and P. R. Avasthi*
Department of Biotechnology, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
The aim of this study was isolation and application of chitosan. Chitin is biopolymer and chitosan is prepared from chitin by demineralization, deactivations, filtration process. Antifungal application is studied for hair, nail, paint and wood. Anti-termites application is also studied. Chitosan is biocompatible, biodegradable and non-toxic which have made wide applicability in the pharmaceutical field. Chitosan having both anti-fungal and anti-termite application so we conclude that it is used to cure fungal infection on hair, nail, paint and wood.
Keywords: Chitin, Chitosan, Shrimp Shell, Anti-termite activity, Anti-fungal activity

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Introduction:

Chitin is found within the skeletal structures of numerous organisms, including Algae (i. e., in diatoms), Fungi (i. e.,in yeast), Porifera (marine and fresh water sponges), Anthozoa (corals), Nematoda, Annelida and Mollusca, as well as a broad variety of arthropods (Insecta, Crustacea and Arachnida).

Chitin is a linear polysaccharide consisting of (1-4)-linked 2-acetamido-2-deoxy-b-D-glucopyranose (Tomihata and Ikada, 1997; Roberts, 1992). Chitin exists abundantly in nature as cellulose and has wide spread applications in agriculture, medicine, biotechnology, cosmetics and bio-inspired materials science (Synowiecki. J. and N.A. Al. Khateeb et al, 2003). Is the second most abundant natural polymer and forms the exoskeleton of many living organisms, commercial chitin is mainly derived from a few organisms, including crabs, shrimps and krill.

Materials and Methodology:

1. Extraction of Chitin and Chitosan
   - 4% NaOH is used for chitin preparations.
   - Boiled samples containing shrimp shell samples was removed from the hot plate and cooled at 30min at room temperature and chrushed to pieces.
2. Deacetylation
   - Adding 50% NaOH to sample and boil at 100˚C for 2hrs on hot plate.
   - Sample cooled for 30min.
   - Samples was washed continuously with 1N NaOH and filtered with chitosan.
   - Samples was left uncovered and oven dried at 110˚C for 6hrs.
   - Creamy white form of chitosan was obatained and is used for further characterization and applications.
3. Purification of Chitosan
   - Removal of insoluble with filtration using NaOH.
   - Reprecipitation of chitosan with 1N NaOH.
4. Demineralization using acetic acid
   - 0.1% standard chitosan powder was added in 0.1N acetic acid and kept on magnetic stirrer.
5. Anti-fungal activity studied
   - Sterile Potato Dextrose Agar plates (PDA) was prepared.
   - The fungal suspension was spreaded on sterile PDA plates and chitosan dipped nail, hairs, paint and wood were placed.
   - Incubate at 37˚C for 11days.

• Anti-fungal activity of chitosan was observed.

6. **Anti-termite activity studied**
   • Termites was placed in empty petri dish.
   • In control petri dish termites was placed.
   • In test petri dish, chitosan sample was added on termites.
   • Anti-termite activity of chitosan was observed.

Result and Discussion:

• Extraction of chitosan:

The fine powder of Chitin can be treated with concentric NaOH in a process called deacetylation to yield Chitosan. After the deacetylation of Chitin with 50% NaOH, The better result were obtain after the treatment with 50% NaOH. So we used 50% NaOH for the extraction of Chitosan by the deacetylation of Chitin and then dried at 110°C for 6hrs.

• Anti-fungal activity:-

Potato Dextrose Agar media was used, sterilized and solidified. The fungal strainAspergillusnigaruse. On control plates, the nail, hairs, paint and wood was found to be fungal infected and on the test plates in which nail, hairs, paint and wood was dipped with chitosan sample was found to be not infected by fungi. The test fungi were found to be resistant for the nail, hairs, paint, wood and termites. Hence, chitosan have strong action against the fungi.

• Anti-termite activity:-

In a control plate, in which the termite was live and in a test plate in which the termite was dipped with chitosan sample was found to be killed with 2min 30sec.

Conclusion: Chitin is natural, bio-degradable, biopolymer with antifungal activity and has a wide range of application in different fields. The second most abounded natural biopolymer. The source made from chitin i.e. chitosan show an excellent anti-fungal activity against the infection of Aspergillusnigeron nail, wood, paint, hairs and also on termites. Chitosan shows anti-fungal and anti-termite activity. So, chitosan might be good candidate for removing infection from the nail, wood, paint and hairs.

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Isolation of lipase from microbes [bacteria and fungi] using oil contaminated soil

Authors & affiliations:
K. V. Koli , H. N. Koli , S. D. Koli and R. V. Gavit*
Department of Biotechnology, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Lipases, triacylglycerol hydrolases, are an important group of biotechnologically relevant enzymes and they find immense applications in food, dairy,detergent and pharmaceutical industries. Lipases are by and large produced from microbes and specifically bacterial and fungal lipases play a vital role in commercial ventures.Some important lipase-producing microbial genera include *Bacillus sps. Pseudomonas* and *Aspergillusniger, Penicilliumsps*. Lipases are generally produced on lipidic carbon, such as oils, fatty acids, glycerol or tweens in the presence of an organic nitrogen source. Bacterial lipases are mostly extracellular and are produced by submerged fermentation.

Introduction :

Lipases are the enzymes capable of catalysing the hydrolysis and synthesis of esters formed from glycerol and long-chain fatty acids (Sharma et al., 2001; Svendsen et al., 2000). Commercially useful lipases are usually obtained from microorganisms that produce a wide variety of extracellular lipases. Only about 2% of world's microorganisms have been tested as enzyme sources. Microbial lipases are produced mostly by submerged culture (Ito et al., 2001), but solid-state fermentation methods can be also used (Chisti, 1999). In general, solid-state fermentation is a well-adapted and cheaper process than submerged fermentation for the production of a wide spectrum of bioproducts (animal feed, enzymes, organic acids, biopulp, aroma compounds, antibiotics, compost, biopesticide, biofertilizeretc). Solid state fermentation is a high recovery method for the production of industrial enzymes (Pandey et al., 1999). Solid state fermentation (SSF) is known to offer many advantages over submerged fermentation leading to overall reduction in the cost of production (Losane et al., 1985; Ghildyal et al., 1985) and the products obtained by SSF are many folds higher than those obtained in submerged cultivation. In addition, the products obtained have slightly different properties (e.g. more thermotolerance) when produced in solid-state fermentation and submerged fermentation. Therefore, if solid-state fermentation variables are well controlled and the purity of the product is defined, this technology may be a more competitive process than is commonly thought. Solid-state fermentation offers many advantages over submerged fermentation for production of the enzyme lipase. Coconut cake: a potent substrate for production of lipase by Candida rugosa in solid-state fermentation (Benjamin et al., 1997). High lipase productions were obtained by cultivation of Rhizopus sp. (Christen et al., 1995; Ul- Haq et al., 2002), Aspergillus sp. (Kamini et al., 1998; Mahadik et al., 2002). Recently cheap agricultural by products like gingelly oil cake (Kamini et al., 1998) and olive oil cake (Cordova et al., 1998; Kademi et al., 2003) have been gaining a great interest as suitable substrates in solid state fermentation for fungi. (Viniegra-González 1998) state that selection of mold strains geared to produce enzymes on solid substrates.(Mahler et.al 2000) stated that Gum Arabic used to emulsify lipid substrates can enhance enzymes production by improving the availability of the substrates.

(Ramachandran et al., 2004) stated that coconut oil cake – a potential raw material for the production of amylase. (Rathi et.al 2002) observed that olive oil is the most used lipid substrate to induce lipase production by bacteria.(Elibol and Ozer 2001) stated that lipase production is influenced by the type and concentration of carbon and nitrogen sources, the culture pH, the growth temperature and the dissolved oxygen concentration. Used palm oil as substrate in mill effluent treatment by tropical marine yeast (Oswal et al., 2002).Mixed solid substrate fermentation (Benjamin et al., 1998) a novel process for enhanced lipase production by Candida rugosa. Therefore, some important problems associated to solid state fermentation:

Methodology:

Materials and Methods

Sample collection:
For isolation of lipase producing organism, soil sample was collected from 4-5 cm depth with help of sterile spatula in a sterile plastic bag from the petrol pump area in Jalgaon.

Screening of microorganism:
After collection, sample was brought to the laboratory and 1 g of sample was suspended in 100 ml of sterile distilled water, agitated for 30 min on a shaker at 500 rpm and 0.2 ml was spread on nutrient agar plates and incubated at 30-37 C for 24-48 hours. Enriched sample was used for plating to get only lipolytic isolates thus enriched samples was plated containing (gm/lit): beef extract 3.0, peptone 5.0, sodium chloride 5.0, agar 15.0, calcium chloride 0.05, and glycerol tributyrate 0.2ml, after incubation for 24 h a total of 10 colonies showing clear zone were picked. One isolate OCR-02 which showed maximum activity was selected and maintained on differential agar medium slantat 40C. The culture was examined for various morphological and biochemical characteristics as per Bergey's Manual of determinative Bacteriology.

Morphological characteristics of Bacteria:

Characters	Shape	Size	Margin	Colour	Elevation	Motility	Opacity	Gram nature
Observation	Rounded	4mm	Entire	Milky white	Convex	Motile	Opaque	Gram positive

Substrate:
Among all the substrate, the maximum lipase activity was observed with Coconut oil cake (Table 1). These results were in accordance with observed lipase production from different literature. Different substrate occupied surface area according to their sizes was an important parameter in solid state fermentation.10 gram of substrate yields maximum production of lipase. Due to its easy penetration, the microbial mass of the bacterial culture showed high growth rate with Coconut t oil cake as a substrate due to which more lipase production was observed. The less lipase production at higher level was due to low mass transfer rate and difficulty in penetration of the organism (Rao et al., 2003). Coconut oil cake was more focused due to its good activity and availability. Coconut oil cake, neem oil cake, mustard oil cake, linseed oil cake were used assubstrates. Different oil cakes used as substrate and their biotechnologicalapplications (Ramachandran et al., 2006). They were procured from a local market ofVellore, India and were dried at room temperature to reduce the moisture content andground to the desired size.

Effect of different substrates on lipase activity.

Substrate	**Lipase activity (U g ds-1)**
Linseed oil cake	1.08
Mustard oil cake	1.53
Neem oil cake	1.23
Coconut oil cake	3.09

Inoculum Preparation:

In order to prepare the inoculum, a loopful of cells from a freshly grown slant was transferred into a 250 ml conical flask containing 50 ml of minimal media (without agar) KH2PO4 3.0 g, Na2HPO4 6.0 g, NaCl 5.0 g, NH4Cl 2.0 g, MgSO4 0.1 g in 1 lit of distilled water and incubated at 30oC in a shaking incubator at 180 rpm for 24 h (Oswal et al., 2002).

Media Preparation:

10 gm of desired oil cake was suspended in 90 ml of minimal media in a 250 ml flask. It was then autoclaved at 15 lbs pressure, 120oC for 20 minutes. It was cooled before Using.

Media preparation in 1000ml distill water:

CONTENTS	AMOUNT
Yeast extract	0.20gm
Mannitol	5.0gm
Monohydogen ammonium phosphate	1.09gm
KCL	2.0gm
MgSO4	0.20gm
Bromoserol purple	0.0075ml
Agar	15.40gm
PH	7.2

Solid State Fermentation:

The above prepared medium was inoculated with 5 ml of inoculum. After thorough588 ChaturvediM etalmixing, all the flasks were incubated at desired temperature in a shaking incubator for48 hours. After a stipulated period samples were drawn. The fermented matter washomogenized and a small amount of sample was taken from each flask for extractionand subsequent analysis

Enzyme Extraction:

The crude enzyme from the fermented material was extracted by simple extractionmethod. The fermented substrate was mixed thoroughly with 90 ml of 0.05 M ofSorenson phosphate buffer (pH 8.0) and then shaking the mixture in a rotary shaker (180 rpm) at 30oC for 48 hrs. The crude enzyme obtained from centrifugation and wasused to determine enzyme activity.

Lipase Assay:

The crude enzyme obtained from centrifugation was assayed for lipase activity. The activity of lipase was determined as described in literature (Winkler and Stuckman) with the following modification, 10 ml of isopropanol containing 30 mg of p – nitro phenyl acetate was mixed with 90 ml of 0.05 M of Sorenson phosphate buffer (pH8.0), containing 207.0 mg of sodium deoxycholate and 100 mg of Gum acacia. According to this method a 2.4 ml of freshly prepared p-nitro phenyl acetate substrate solution was mixed with 0.1 ml of crude enzyme. After 15 minutes of incubation at 15oC, optical density was measured at 410 nm against an enzyme free control. One unit of lipase activity is defined as the amount of enzyme releasing 1 mole p – nitrophenol per minute under assay conditions.

Optimization of Medium Parameters:

The different parameters selected and optimized (Pau & Omar, 2004) were substrate selection, pH of the medium, incubation time and effect of moisture content of substrate.

Results:

Isolation of bacteria from soil sample

Microbiological analysis of soil samples revealed that contaminated sites with organic waste contain high bacterial count. For Total Viable Count, the soil samples were diluted up to 10-7 in Ringer's Solution and plated in Nutrient agar. After incubation the colonies were counted and the results were tabulated. The bacterial count varied from 1.0 x 108CFU/g to 5.2 x 108CFU/g of the soil sample.

Isolation of lipolytic strains

The serially diluted soil samples were plated on Differential agar medium and lipolytic count varied from 1.1 x 108CFU/g to 4.9 x 108CFU/g of the soil sample. It was found that the soil samples collected from oil refineries waste contaminated sites showed highbacterial count.

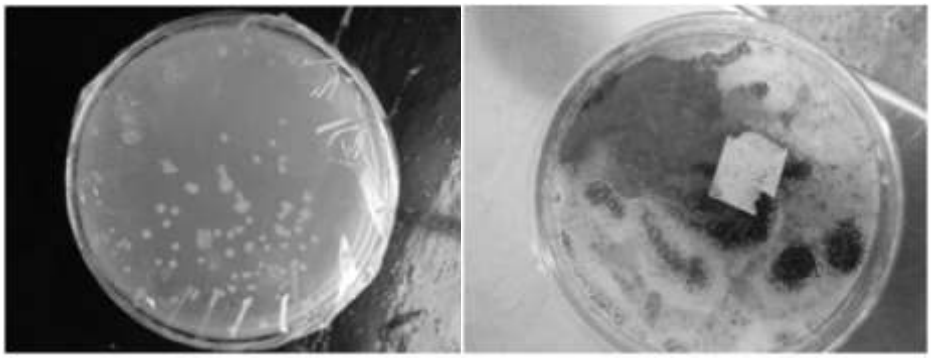

Fig.[A] Bacillus subtillis *Fig.[B] Aspergillusniger*

Grams staining:

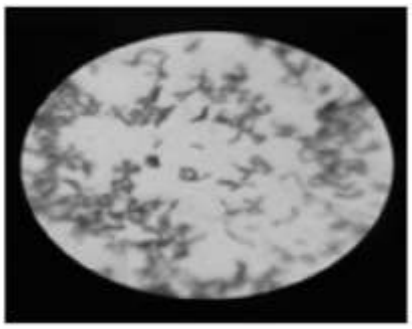

Fig. Bacillus subtillis[gram positive]

Screening of potential bacterial strain for lipase production

On the basis of larger clear zone formation onDifferential agar medium , 08 potential isolateswere selected in the study. The isolates were grown in Differential broth at pH 8 andthe supernatant were assayed for lipase activity after incubating for 48 hours. It was found that the isolate OCR – 02 was found to have highest enzyme production as compared to other isolates. The isolate OCR - 02 was taken for further studies.

Effect of incubation time

The amount of lipase produced was observed after every 12 hours till 60 hours. The maximum lipase activity was observed after 48 h of fermentation listed in Table 2. After that, although the bacterial growth rate went on increasing but the specific growth rate (slope of the curve in Figure 3) decreased. After 48 h, the growth showed divergence from the exponential because in place of homogeneous growth, bacterial pellets began to form in which nutrients and oxygen supply became the growth limiting. After that lipase yield got reduced due to the consumption of nutrient materials.

Table 2: Effect of incubation time on lipase activity.

Incubation time (hours)	Lipase activity (U g ds-1)
12	0.35
24	1.73
36	2.10
48	4.65
60	2.62

Effect of pH on enzyme production

As pH is the important parameter required for the growth of bacterial culture in respective media so lipase activity got affected with basic pH, this indicates that suitable pH is responsible for bacterial growth in the media. The data obtained clearly indicates that there is a strong influence of pH on lipase enzyme production. Thus the maximum activity was reported at pH 8, mentioned in Table 4 (Fig 5).

pH of medium	Lipase activity (U g ds-1)
6	2.09
7	3.23
8	4.71
9	3.52
10	3.12

Conclusion:

The various composition influenced enzyme production by the bacteria, it appears that the nature of the substrate had significantly influenced the impact of initial moisture content and incubation period of overall enzyme yield. The physical nature and water holding capacity are important criteria for a solid substrate for its use in SSF process and the moisture content of themedium is a critical factor that determines the microbial growth and product yield inSSF. Fermentation in shake flask improved the lipase yield with an activity 4.65 U gds-1 with in 48 h, 4.62 U g ds-1 with initial moisture content of 70% and 4.71 U g ds-1at pH 8 using Coconut oil cake as substrate by Bacillus subtilis. Thus this study hasproved that the optimization of growth parameters in a suitable solid state medium hassignificant effect on improved production. Solid state fermentation for production ofhighertitres of thermostable enzyme with two peaks achieved by optimizing the pH592 Chaturvedi M et al(Ramesh &Lonsane, 1989). This is one of the prime objectives of industrialmicrobiology for large scale production of valuable metabolites, which can beachieved with balanced nutrient supply.

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Plant oil as corrosion inhibitor

Authors & affiliations:
G. K. Patil, V. S. Dandge, K. R. Chavan and A. S. Rajhans*
Department of Biotechnology, M. J. College, Jalgaon. (M.S.) India.

ABSTRACT:
The purpose of this study is to make the people aware of organic corrosion inhibitors. The oils of various plants used as vapour phase corrosion inhibitors because of their volatile nature. Oils derived from plants are also used to prepare coating which are used to prevent metals from corrosion. This is also an efficient method of preventing metals from corrosion by forming a surface film.
Keywords: inhibition of corrosive bacteria,bacillus and pseudomonas sp.,plant oils ,zone of inhibition.

Introduction:
Steel is universally applied to the manufacture of reactors, gathering pipelines, storage vessels, drilling equipment, and other apparatuses, due to its excellent mechanical properties and thermal conductivity. Corrosion of steel occurs readily in acidic solution during the procedures of pickling, scale-removal and rust-cleaning, which results in metal failure and huge financial losses. A range of corrosion inhibitors have been synthesized and exhibit high inhibition efficiency, their applications in industry are limited due to their toxicities and complex manufacturing processes. In comparison with conventional synthetic inhibitors, plant extracts display the advantages of low cost, abundant resources, simple production procedures, and being environmentally friendly. According to the relevant literature, plant extracts contain a variety of phytochemical constituents. Such as tannins, alkaloids, flavonoids, polyphenols, saponins, glycosides, anthraquinones, amino acids, proteins, and other heterocyclic compounds. These phytochemicals have been considered to be potential corrosion inhibitors. To the best of our knowledge, the introduction of a low toxicity plant as a corrosion inhibitor was attributed to Elhosary et al. (1972). The oils obtained from various plants are also used as corrosion inhibitors because they are cheap, renewable and environmentally safe (Bouyanzer and Hammouti, 2004a, b; Grassino et al., 2009; Halambek et al., 2010; Okafor et al., 2008; Benabdellah et al., 2006; Eddy, 2009) When an inhibitor is chosen for a process, the following points must be taken into consideration:
· #They should be cheap.
· #The inhibitor chosen must be non-toxic.
· The inhibitor chosen should be easily available if it availability is low the inhibitor, often becomes expensive.

Objectives: This study aim was to inhibit the bacteria causing corrosion of metals using plant oils.

Material and Methodology:
Material :
Neem oil, castor oils ,and ginger oils are buy from market in already prepared form.

Isolation of bacteria: Pseudomonas Sp. & Bacillus Sp.Isolated from Corrosion of Diselpipeline & fresh water

Methods : Antibacterial activity-By Disc And Paper Diffusion Method.
Bacillus sp. , pseudomonas sp. was isolated from corrosion of diselpipelines and fresh water. Oils are purchesd from market in already prepared form and by paper disc method and by well method anti-

bacterial activity was studied.
Spread the bacterial culture on the sterile nutrient agar plate. Place the disc in the centre of plate which contain the oil. Incubate the plate at 37°c for 24hrs.Observe the Zone of inhibition.

Results: The antibacterial activities of the oils of different medicinal plants were studied by disc and paper diffusion method.LB and Nutrient agar media was used,sterilized and solidified . Two bacterial strains Pseudomonas sp.and gram positive Bacterial Bacillus sp.were used. wells made and oil with different volume were deeped & kept for incubation at 37°C for 24 hrs zone of inhibition for control and oils were measured. The microorganisms found to be resistant for oils but results indicated that oils have strong action against microorganisms.

Zone of inhibition (mm) "Castor oil"

Bacteria	6 µl	8 µl	10 µl
Bacillus Sp.	17	18	18
Pseudomonas Sp.	20	24	20

Zone of inhibition (mm) (Neem oil)

Bacteria	6 µl	8 µl	10 µl
Bacillus Sp.	22	25	28
Pseudomonas Sp.	22	20	28

Bacteria zone of inhibition (mm) "Ginger oil"

Bacteria	6 µl	8 µl	10 µl
Bacillus SP.	21	19	22
Pseudomonas Sp.	20	28	24

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Hazardous fungi associated with indian currency: A case study in jalgaon city, maharashtra.

Authors & affiliations:
S. B. Gavhare, N. P. Purohit, and G. M. Rane*
Department of Botany, M. J. College. Jalgaon, (M.S.) India.
email : gaurirane64@gmail.com

ABSTRACT:
Currency is widely used and handled several times by several people and it could be one of the potential media to transmit various diseases among people who handled them. Contaminated currency not only reduces the life of it but can also cause serious diseases like mycosis, gastric disturbances, respiratory infections etc. In the present study we tried to isolate and identify the fungal flora associated with Indian currency notes and coins. For these isolations notes and coins were collected from different food sellers like tea, panipuri, vadapav, chiness food, fruit, vegetables, sugar cane juice, anda bhurji, chanajor and from bank. Conventional method of swabbing was used to isolate fungal flora from notes wile coins were washed in sterile distilled water and these washings were inoculated on culture media. Great diversity of fungi was observed during studies. Overall 393 colonies were isolated from notes and 186 colonies were isolated from coins. Total 38 species of 11 genera were isolated during these studies. From notes total of 24 species of 7 genera and from coins total of 34 species of 10 genera were isolated. *Aspergillus* was the dominant genus with 16 species. Species of *Alternaria, Cladosporium, Curvularia, Mortiriella, Mucor, Penicillium, Pseudotorula, Rhizopus* and *Trichoderma* were isolated. Overall maximum numbers of fungi were isolated from notes of Rs. 10 and coins of Rs.1 and minimum numbers of fungi were isolated from notes of Rs. 100 and coins of Rs. 2.Many pathogenic fungi found associated with Indian currency.
Key words: Hazardous Fungi, Food Sellers, Indian currency, Coins, Notes.

Introduction:

A currency refers to diverse denomination made up of paper issued by the Reserve Bank of India or the nationalized bank and coins made of metals. Paper currency and coins are broadly used in most day to day cash transactions. Money in form of notes and coins pass through the hands of many people as a form of payment for goods and services settlement of debits and for deferred payment in economic activities (Beg and fisher, 1997). These paper currency variously contaminated by different microorganism like bacteria, fungi, viruses, etc. These are the one of the main source of pathogenic or transfer of disease from one person to another person or diseased person to healthy person. Generally modern bank notes are made up of special blend of 75% cottons and 25% of linen with small segment of fiber so paper money is something of isomer (Badvi, Jawed M. 2017). This paper money also have cellulosic material in the potential source for growth of the fungal spores. Paper currency provides large surface area as a breeding ground for pathogens. The use of saliva and water pads for counting notes moistens the notes and encourage the growth and establishment of a micro habitat of microbes. Money is contaminated as it circulates from hand to hand, besides reducing the life span of the notes some of these fungi have the potential to cause skin, eye and gastrointestinal infections. Infections of internal organs as well as serious diseases of the respiratory tract in man, Since money is not screened for microbes. There had been fewer studies on the microbial contamination of currency worldwide and especially in India. There is a need to study the fungal contamination of currency either in form of notes and coins. It has become necessary for thorough investigations to be carried out to determine safety of the Indian currency.

Materials and Methods:

Study of fungal contamination on Indian currency was carried out during the period of August to December in the year 2017.

Sample Collection: Total 243 samples of Indian currency including notes and coins were collected from different sellers of Jalgaon city. Notes of denominations of Rupees 10, 20, 50,100,200 and coins of Rupees 1, 2, 5 and 10 were randomly collected from six different venders as, Tea seller**,** Panipuri seller**,** Vadapav seller, Chinese seller, Fruit seller**,** Vegetable seller sugar cane juice seller, anda bhurji seller, chanajor seller and bank. Two notes of each denomination and coins two of each were aseptically collected into sterilized polythene bags by wearing sterilized gloves and brought to the laboratory for isolating the fungi and diversity studies.

Inoculation: Paper Currency

For inoculation the conventional method of swabbing was used. The surface of the currency was wiped under aseptic condition on both sides by sterile wet cotton swab. Separate cotton swabs was used for each denomination and diluted separately in test tube containing 10 ml sterile distilled water. 1 ml of dilution was taken and inoculated on two different culture media viz. Czapek Dox Agar and Lactose Yeast Extract Agar separately. After 5 days of inoculation fungal colonies were observed.

· **Coins**

Different method was used isolating fungi from coins. Coins were directly washed in sterile distilled water. Each coin was separately washed in petriplate containing 10 ml sterile distilled water. 1 ml of that water was inoculated on two different culture media Czapek Dox Agar and Lactose Yeast Extract Agar separately. After 5 days of inoculation fungal colonies were observed.The diameter of the colony was measured, surface and reverse colours of colony were noted. The semi-permanent slides were prepared using cotton blue and lactophenol. Photomicrographs of these fungi were taken. Identification of fungal forms was done using relevant literature, viz Ellis (1971, 1976), Subramanian (1971), Thom and Raper (1945) etc.

Results and Discussion and Conclusion:

Total 38 species of 11 genera were isolated during these studies. From notes total of 24 species of 7 genera and from coins total of 34 species of 10 genera were isolated. *Aspergillus* was the dominant genus with 16 species. Species of *Alternaria, Cladosporium, Curvularia, Mortiriella, Mucor, Penicillium, Pseudotorula, Rhizopus* and *Trichoderma* were isolated. (Table)

Table: List of fungi associated with currency

Sr. No.	Name of fungus	Notes					Coins			
		10	20	50	100	200	01	02	05	10
1.	*Alternaria amaranthi* (Peck)Van	-	-	-	+	+	-	-	+	+
2.	*A. tenuissima* (Fries) Wiltshire	+	-	-	+	-	-	-	-	-
3.	*Aspergillus candidus* Link	+	+	-	-	-	+	+	-	+
4.	*A. effuses* Tiraboschi	+	+	-	+	+	-	+	+	-
5.	*A.flavipes* Bainier & Sartory	+	-	-	-	-	-	-	-	-
6.	*A. fumigatus* Fresenius	+	+	+	-	-	+	+	+	+
7.	*A. janus* Raper and Thom	+	-	-	-	-	-	-	+	-
8.	*A. luchuensis* Inui	+	+	+	+	+	-	-	-	+
9.	*A. micro-virido-citrinus* Costantin	-	+	-	-	-	-	-	-	+
10.	*A. miyakoensis* Nakazawa	-	-	-	-	-	+	-	-	-
11.	*A. niger* var-*cinnamomeus*	+	+	+	+	+	+	+	+	+
12.	*A. niveus* Blochwitz	-	-	-	-	+	-	-	-	+
13.	*A. sulphureus* Thom and Church	+	+	+	+	+	-	+	+	+
14.	*A. sydowi* (Bain and Sart.)Thom and	+	+	+	+	+	-	-	+	+
15.	*A. ustus* (Bainier) Thom and Church	+	-	+	-	-	+	+	-	-
16.	*Bipolaris specifera* Subram	-	-	-	-	-	+	-	-	-

17.	*Clodosporium cladosporioides* (Fres.)DeVries	+	+	+	-		-	+	-	-
18.	*C. oxysporum* Berk and Curt	+	+	+	-	- -	-	-	-	+
19.	*Curvularia affinis* Boedijn	-	-	-	-	-	-	-	+	-
20.	*C. andropogonis* (Zimm) Boedijn	-	-	-	-	-	-	-	-	+
21.	*C borreiae (Viegas)*	-	-	-	-	-	-	+	-	-
22.	*C. lunata* (Wakker) Boedijn	-	-		-	-	+	-	-	-
23.	*C. maculans* (Bancroft) Boedijn	+	-		-	+	-	-	-	-
24.	*C. prasadii* R.L. and B.L.Mathur	+	-		-	-	-	+	+	+
25.	*Mortierella isabellina* (oudemans) Zycha	-	-	-	-	-	+	-	-	-
26.	*Mucor mucedo* Fres.	-	-	-	-	-	-	-	+	-
27.	*Penicillium cyaneum*	+	-	-	+	+	-	-	+	-
28.	*P. frequentans* Westling	-	-	-	-	+	-	+	+	-
29.	*P. implicatum* Biourge	+	+	+	-	-	-	-	-	-
30.	*P. multicolor* Grigorieva-Manoilova and Poradielova	-	-	-	-	-	-	-	-	+
31.	*P. purpurrescens* (Sopp.)n.comb.	-	-	-	-	-	-	-	-	+
32.	*Pseudotorula herbarum* (Pers.)Link	-	-	+	-	-	-	-	-	-
33.	*Rhizopus nigricans* Ehrenb	+	-	+	+	+	-	-	-	+
34.	*Trichoderma herbarum* (pers) Link & Fri	-	-	-	-	-	-	+	-	-
35.	Brown Sterile mycelium	-	-	-	-	-	-	-	-	+
36.	Green Sterile mycelium	-	-	-	-	+	-	+	-	+
37.	White Sterile mycelium	-	-	+	-	-	+	-	+	+
38.	Yellow Sterile mycelium	-	+	-	-	-	-	+	+	+
	Total	**18**	**12**	**12**	**09**	**12**	**09**	**12**	**14**	**19**

Total 393 colonies were isolated from currency notes of rupees 10, 20, 50 and 100,200 and total 187 colonies were isolated from coins of rupees 1, 2, 5 and 10. Maximum numbers of colonies i.e. 156 were isolated from notes of Rs 10, followed by 94 from notes of Rs 20, 85 from notes of RS 50, 89 from Rs.200 and minimum i.e. 59 from notes of Rs 100. From coins maximum number of fungal colonies i.e. 58 were isolated from 1 rupees coin, followed by, 55 from coins of 5 rupees, 52 from coins of Rs.10 and minimum number i.e. 41 from coins of 2 Rs. Maximum numbers of colonies i.e. 104 were isolated from the notes collected from a anda bhurji seller followed by 89 from sugarcane juice seller,71 from tea seller, 66 from chanajor seller, 42 from the Chinese food seller, 34 from the fruit seller, 33 from the Vadapav seller, 21 from the vegetable seller , 16 from the Panipuri seller, minimum no of colonies i.e. only 07 from notes obtained from bank. From the coins maximum no of colonies i.e. 18 from tea seller, followed by 15 from vadapav seller, 13 from vegetables seller, 12 from Chinese food seller, 9 colonies each from Panipuri and fruit seller and minimum number of colonies from the bank i,e, 4.

Great diversity of fungi was observed during this study. Total 3 species of 3 genera from Zygomycotina and 31 species of 6 genera from Deuteromycotina were isolated. Overall from the notes, maximum number of fungi were isolated from Rs.10 while minimum number of fungi were isolated from notes of Rs.100 this is due to notes of Rs10 are more in circulation and widely used. In general the food cost is comparatively low so we can purchase many food items using 10 Rs notes. However Rs100 notes are not so commonly used hence less number of fungi were isolated from these notes. In case of coins very few food items are available at Rs 1 or 2 so Rs 10 coins are more widely used for purchasing these food items. Therefore more number of fungi were isolated from coins of Rs 1& 2.In this study *Aspergillus* genus was dominant genus. *A. Niger* var –*cinnamomeus* was isolated

from all notes and coins which was collected from all sellers and from bank also. To overcome the threat of disease spread through currency handling some suggestions are 1)Use E-banking for safe transactions.2) Plastic Currency should be introduced ,which can be washable.3) The coating used for currency notes should be with antimicrobial properties. 4) There should be regular disinfection of currency deposited in banks & post offices by using fumigation. 5) Should eat the food from clean places. Person / Vendor should wear gloves while serving by which we can overcome the diseases cause by fungal infections. Ex.- Dysentry, Vommeting, Various viral infections, Mucorosis , Penicilliosis, various skin and eye diseases, etc.6) Make awareness towards contaminated currency and its effect in public.

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Hydroponic using household plastic waste

Authors & affiliations:
S. A. Mahajan, S. P. Verma, V. R. Patil and M. K. Deshattiwar*
Department of Botany, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
With increasing population per capita land is decreasing day by day also the yield because of some land problem like soil fertility. Hydroponics is the technique which fulfills the future food requirement of the humankind. The main objective of this study is the use of waste plastic wares, so that layman will be able to do this at his own place. In this study stagnant hydroponic system was used to grow *Trigonella foenum-graecum* seed in the plastic bottles containing the sofa foam as a supporting material as well as nutrient solution. Nutrient solution made by using KH_2PO_4, KNO_3, $FeSO_4$and$MgSO_4$, as potassium, nitrogen phosphorus, Iron and Magnesium source respectively.
Keywords: Waste plastic ware, seeds of *Trigonella foenum-graecum* and Nutrient solution

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Introduction:

The word Hydroponics comes from Latin and means working water. Simply put, it is the art of growing plants without soil (www.Simplyhydro.com/what). Hydroponics, a method of growing plant in nutrient solution, without soil. The principle of hydroponics gardening have been used since ancient times. They were brought to popular attention in the United states by Dr. W.F. Gerick, who introduced the word 'hydroponics'. The earliest published work on growing terrestrial plants without soil was the 1627 book Sylva Sylvarum or 'A Natural History' by Francis Bacon, printed a year after his death. In 1699, John Woodward published his water culture experiments with spearmint. He found that plants in less-pure water sources grew better than plants in distilled water. By 1842, a list of nine elements believed to be essential for plant growth had been compiled, and the discoveries of German botanists Julius von Sachs and Wilhelm Knop, in the years 1859–1875, resulted in a development of the technique of soilless cultivation. In 1929, William Frederick Gericke of the University of California at Berkeley began publicly promoting that solution culture be used for agricultural crop production.

Gericke created a sensation by growing tomato vines twenty-five feet (7.6 meters) high in his back yard in mineral nutrient solutions rather than soil. He introduced the term hydroponics, water culture, in 1937, proposed to him by W. A. Setchell, a phycologist with an extensive education in the classics. While he was eventually provided greenhouse space, the University assigned Hoagland and Arnon to re-develop Gericke's formula and show it held no benefit over soil grown plant yields, a view held by Hoagland. In the 1960s, Allen Cooper of England developed the Nutrient film technique. In addition, conventional crop growing in soil (Open Field Agriculture) is somewhat difficult as it involves large space, lot of labor and large volume of water. Beibel, J.P. (1960). In the 1974s, Ellis et al, Soil is usually the most available growing medium for plants. It provides anchorage, nutrients, air, water, etc. for successful plant growth. The suspended net pot, non-circulating hydroponic growing method is a unique and powerful technique for growing leafy, semi-head and small romaine lettuce cultivators, because the entire crop can be grown with only an initial application of water and nutrients (kratky,1993,1995,1996,2004,2005).

Water use efficiencies of less than 20 liters per kg of lettuce are common, efficiency as low as 11 liters per kg of lettuce has been recorded (Kratky et al. 2008). Growers typically aim for 150 to 250 gram heads with this growing method, and the nutrient solution consumption range of 3 to 6 liters per plant (Kratky , B.A. (2009).

In 2007, Eurofresh Farms in Willcox, Arizona, sold more than 200 million pounds of hydroponically grown tomatoes. Eurofresh has 318 acres (1.3 km2) under glass and represents about a third of the commercial hydroponic greenhouse area in the U.S. Eurofresh tomatoes were pesticide-free, grown in rockwool with top irrigation. Eurofresh declared bankruptcy, and the greenhouses were acquired by NatureSweet Ltd. in 2013. As of 2017, Canada had hundreds of acres of large-scale commercial hydroponic greenhouses, producing tomatoes, peppers and cucumbers.

In recent decades, NASA has done extensive hydroponic research for its Controlled Ecological Life Support System (CELSS). Hydroponics research mimicking a Martian environment uses LED lighting to grow in a different color spectrum with much less heat. Ray Wheeler, a plant physiologist at Kennedy Space Center's Space Life Science Lab, believes that hydroponics will create advances within space travel, as a bioregenerative life support system. Due to technological advancements within the industry and numerous economic factors, the global hydroponics market is forecast to grow from $226.45 million USD in 2016 to $724.87 million USD by 2023.

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Why Hydroponics Crops In India?

In India, the demand for green vegetable and other crops is increasing on the account of diversified use

of agricultural residues. Adequate attention is not being given to production of vegetables and other crops due to increasing pressure on land for constructing building and also land used for crops production is increasing day by day.
In order to meet this increasing demand for vegetables other crops and fruit, the next alternative is Hydroponics crops. Some of the benefits of hydroponics crops production being Land preservation, Water conservation, faster growth and maturity, Minimal use of Fungicide and Pesticide, less labour and maintenance costs. Control over Growing environment. Time saving continual produce and Weed free. Present study aims to develop a less expensive and suitable method for cultivation crops plants hydroponically by ordinary people.

Materials And Methods:
Seed material collection:
Seeds of crops like Fenugreek where obtained from the local market.

Chemicals:
All the chemicals used in the present study were analytical grade and were obtained from the Himedia, Qualigens India.

Glassware:
Glassware used in all the experiments was procured from the 'Borosil' India. Conical flasks (100-1000ml capacity) , measuring (10-1000ml capacity) and pipettes (1-10ml capacity) were used during present study.

Plastic ware (Bottles):
In the present study used plastic bottles of different capacity were obtained from road side or collected thrown waste drinking water bottles.

Preparation of Media:
In the present study double distilled water was used for the preparation of hydroponic media. After addition of macro elements micro elements were added and the final volume is adjusted. Composition of Hoglands media is given in Table No.1 .

Methods for the cultivation of plants:
For the cultivation of hydroponic plants there are various method are available such are covered tank, 4 litre bottle method, Float support system methods, etc. These methods are extremely expensive, therefore in the present study we used simplified methods some of them as follow:

Method:
In this method we drilled the bottles at particular distance and placed the sponge on to the bottles and then seeds were placed on the foam or sponge.
In all this methods, we used waste plastic water bottle place vertically and drilled holes on the upper side of the bottles with help of needle or any suitable instrument .
Firstly we collected waste plastic water bottle from road side or where they were available. With the help of needle we made holes on the upper side of bottle then through these holes, we placed foam in the bottle. Stock solution of the Hoagland media was prepared. Using stock solution of the Hoagland media, working media was prepared. Prepared media were poured in the bottles the seeds were placed on the foam for germination.
After few days the seeds were germinated and in few days plants were developed. Photographs were taken in 2-7 days intervals.

Table No.1: Composition of Hoglands media

Component	Stock solution	MI Stock solution/1L
Macronutrients		
KNO3	50.5	2.5
FeSO4	3.175	1.5
MgSO4	123.25	1
Micronutrients		
H3BO3	0.715	1
MnSO4	0.4525	1
ZnSO4	0.05	1
CuSO4	0.0127	1
H2MoO4	0.02	1
Na2MoO4	0.03	1
KH2PO4	34	0.5

Results and Discussion

Commercially for the cultivation of hydroponic plants various methods are available. These available methods are very expensive and these methods are not understandable for the ordinary people. The studies undertaken deals with the development of easy methods for the cultivation of crops in hydroponic which are understandable and trouble free for common man. For this study we used crop like fenugreek.

Cultivation of Fenugreek:

For the cultivation of fenugreek plants hydroponically, waste bottles were used. Waste plastic bottles were drilled at specific intervals to make holes, through holes sponge were placed in the bottles. Fenugreek seeds were placed on the sponge . Growth of the plants were observed, it found that for the sprouting of the seeds it took 3-5 days Photoplate No. 1. Plants took 8-9 days Photoplate No. 2 for the development of roots and shoots. Plants grow luxuriantly when kept in the morning sun rays and then shifted to the shade in 15-20 days Photoplate No. 3

Photoplate No. 1 After 3-5 days of sowing

Photo-plate No. 2: After 8-9 days of sowing

Photo-plate No. 3: After 15-20 days of sowing

Conclusion:
Cultivation of plants hydroponically is the need of future generation because of the available land for cultivation of crop is decreasing day by day. Finally we would like to conclude that method is the best and easy for the cultivation of hydroponic plants.

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Preliminary studies on rhizosphere and non rhizosphere fungi of some local medicinal plants

Authors & affiliations:
M. V. Patil, J. J. Wani, D. Wani, A. Bhokare* and G. M. Rane*
Department of Botany, M. J. College. Jalgaon, (M.S.) India.
email: gaurirane64@gmail.com

ABSTRACT:
Soil is rich in microflora. Among them fungi are present in large numbers. Many fungi are associated with root region called as rhizosphere fungi. They have interaction with roots. Here the efforts have been made to study the rhizosphere fungi of some locally available plants used in treatment of diabetes. Diabetes is an important human ailment affecting many people from various walks of life in different countries. In India it is proving to be a major health problem, especially in the urban areas. For the present study different locally available medicinal plants are selected viz. Curry leaf (*Murraya koenigii* L.), Jamun (*Syzygium cumini* L.) and Neem (*Azadirachta indica* A. Juss.). From rhizosphere and non rhizosphere of *Murraya koenigii* total 23 species of 7 genera were isolated. From rhizosphere and non rhizosphere of *Syzygium cumini* total 18 species of 6 genera were isolated. From rhizosphere and non rhizosphere of *Azadirachta indica* total 32 species of 9 genera were isolated. In all the three plants more numbers of fungi were isolated from rhizosphere soil than from non rhizosphere soil.
Key words: Rhizosphere, Non rhizosphere, Fungi,Medicinal plants

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Introduction:

Soil is considered to be the most biodiverse and dynamic habitat on Earth. It is made up of different components such as sand, slit, clay and organic matter (Zain et al. 2014) which provides habitat for a wide variety of organisms including bacteria, fungi, protozoan, nematodes and earthworms where they interact with each other and with their physical environment (Shahnaz et al. 2014). Along with micro-organisms soil support growth of different plants. Many of them are medicinal used in treatment of diseases like diabetes. Involvement of soil micro organisms is in a wide variety of metabolic and physiological activities that influence the microhabitat. Rhizosphere as the area around a plant root that is inhabited by a unique population of microorganisms influenced by the chemicals released from plant roots. During literature survey it has been observed that no much work has been done on the rhizosphere mycoflora of Neem (*Azadirachta indica*), Jamun (*Syzygium cumini*) and Curry tree (*Murraya koenigii*) so the present investigation was undertaken.

Materials and Methods:

3 samples each from Rhizospheric and Non- Rhizospheric soil of 3 different medicinal plants *Azadirachta* indica A.Juss., Syzygium *cumini* L and *Murraya koenigii* L. were collected from the campus of M .J. College, Jalgaon. Samples were collected by gentle uprooting the plants using sterile shovel. The soil particles adhered to the roots were transferred to sterile polythene bags. Also the soil adjacent few centimeters away from the roots were isolated and were considered as non-rhizospheric soil. The samples were carried aseptically to the laboratory and were processed within 24 hours. The 2 media used for isolation of fungi were Czapek's Dox Agar (CzDA) and Lactose Yeast Extract Agar (LYEA).The samples were inoculated using Direct plate method (Waksman, 1916).,Serial dilution method (Waksman, 1916). For identification, morphological characters of the colony were noted .Semi permanent slides of these fungi were prepared using cotton blue & Lacto phenol. Camera lucida drawings and photomicrographs were also done. The identification of different soil fungi was done with the help of relevant literature such as Thom, and Raper (1945), Raper & Thom (1949), Tandon (1968), Seth (1970), Ellis (1971&1976), Subramanian (1971) etc.

Results, Discussion and Conclusion

Sr.No.	Name of fungus	Name of medicinal plants					
		Murraya koenigii		***Syzygium cumini***		***Azadirachta indica***	
		R	**NR**	**R**	**NR**	**R**	**NR**
1.	*Aspergillus atropurpureus* Zimm	-	+	-	-	-	+
2.	*Aspergillus awamori* Nakazawa	-	-	-	+	-	+
3.	*A. caespitasus* Raper and Thom	-	-	-	-	+	-
4.	*A. candidus* Link	-	+	+	+	+	+
5.	*A. conicus* Blochwitz	-	-	-	-	+	-
6.	*A. effusus* Tiraboschi	-	+	-	+	-	+
7.	*A. flavipes* (Bainier and Sartory)	+	+	+	-	+	+
8.	*A. flavus* Link	+	+	-	-	+	-
9.	*A. fonsecaeus* h.sp.	-	-	+	-	-	-
10.	*A. fumaricus* Wehmer	-	+	-	-	-	-
11.	*A. fumigatus* Fresenius	-	-	-	-	+	-
12.	*A. itaconicus Kinoshita*	+	+	+	-	+	-
13.	*A. janus* Raper and Thom	+	-	-	-	-	-
14.	*A. japonicus* Saito	-	-	-	-	+	-
15.	*A. luchuensis* Inui	-	-	-	-	+	-
16.	*A. nidulans* (Eidam)Wint	-	+	-	-	-	-
17.	*A. niger* mut *schiemanni* (schiem) n. Comb	+	+	+	+	-	-
18.	*A. ochraceus* Wilhelm	-	-	-	-	-	+
19.	*A. oryzae* (Ahlburg) Cohn.	-	-	-	-	+	-
20.	*A.* parasiticus Speare	+	-	-	-		
21.	*A. sulphureus* Thom and Church	-	+	-	-	-	+
22.	*A. sydowi* (Bain and Sart.)Thom and Church	-	+	-	-	-	-
23.	*A. terreus* Thom	+	-	-	-	-	-
24.	*A.* terricola Marchal	+	-	-	-	-	-

25.	*A. ustus* (Bainier) Thom and Church	-	-	-	-	+	-
26.	*A. versicolor* (Vuill) Tiraboschi	-	-	+	-	-	-
27.	*Cladosporium cladosporioides* (Fres.) de Vries	+	-	-	-	-	-
28.	*C. herbarum* Link ex Fries	-	+	-	-	-	-
29.	*Fusarium dimerum* Penzig	-	-	-	-	+	-
30.	*Fusarium moniliformae* J.Sheldon	+	-	+	-	-	+
31.	*F. oxysporum* Schlecht ex.Fries	-	-	-	-	-	+
32.	*F. sporotrichioid* Sherb	-	-	-	-	+	+
33.	*F. poae (Peck)*	-	+	+	-	-	+
34.	*Mortirella isabellina* (oudemans) Zycha	-	-	-	-	+	-
35.	*Mucor hiemalis* Silvaticus	-	-	-	+	+	-
36.	*M. mucedo* Linne ex Fres	-	-	-	-	-	+
37.	*Mucor racemosus* Fresen.	+	-	-	+	+	-
38.	*Penicilium citrium* Sopp, Thom, Series	-	-	+	-	-	-
39.	*P. corylophilum* Dier (kx)	-	-	+	+	-	+
40.	*P. rubrum* Stoll	-	-	+	-	-	+
41.	*P. rugulosum* Thom	-	-	+	-	+	-
42.	*Phoma betae* (Berl.) Nevodowsky	+	-	-	-	-	-
43.	*Rizopus nigricans* Ehrenb.	+	-	+	-	-	+
44.	*Rizoctonia solani* Kuhn	+	-	-	-	+	-
45.	*Trichoderma viride* Pers.ex Fries	-	-	+	+	+	-
46.	*Verticillium luteoalbum* (Link ex Fries) Subram.	-	-	-	-	+	-
47.	White Sterile mycelium	+	+	+	+	+	+
48.	Green sterile mycelium	+	-	-	-	-	-
	Total	**16**	**14**	**15**	**09**	**21**	**16**

From rhizosphere and non rhizosphere of *Murraya koenigii* total 23 species of 7 genera were isolated, among them 2 genera and 2 species from Zygomycotina and 5 genera with 21 species of Deuteromycotina and 2 sterile mycelia. From rhizosphere soil total 16 species of 7 genera were isolated among them 2 genera and 2 species from Zygomycotina and 5 genera with 12 species of Deuteromycotina and 2 sterile mycelia were isolated, while from non rhizosphere soil total 7 genera of 14 species were isolated, among them 2 genera with 2 species from Zygomycotina, 5 genera with

10 species of Deuteromycotina and 2 sterile mycelia. Among these fungi 15 species of Aspergillus, 2 species each of Cladosporium and Fusarium and single species each of *Mucor, Phoma, Rhizopus* and *Rhizoctonia* were isolated. 4 species of *Aspergillus* were present both in rhizosphere and non rhizosphere while 7 species of *Aspergillus, Cladosporium herbarum* and *Fusarium poae* were isolated only from non-rhizosphere, while 3 species of *Aspergillus, Cladosporium cladosporioides, Fusarium moniliformae, Mucor racemosus, Phoma betae, Rizopus nigricans, Rizoctonia solani* were isolated only from rhizosphere soil (Table no. 1).

From rhizosphere and non rhizosphere of *Syzygium cumini* total 18 species of 6 genera were isolated, among them 2 genera of 3 species from Zygomycotina and 4 genera with 14 species of Deuteromycotina and 1sterile mycelia. From rhizosphere soil total 15 species of 5 genera were isolated among them 4 genera with 13 species from Zygomycotina and 1 genera with 1 species of Deuteromycotina and 1 sterile mycelia were isolated while from non rhizosphere soil total 4 genera of 9 species were isolated, among them 1 genera with 2 species from Zygomycotina, 3 genera with 6 species of Deuteromycotina and 1 sterile mycelia. Among the total isolated fungi 8 species of *Aspergillus,* 4 species of *Penicilium* 2 species each of *Fusarium* and *Mucor,* single species each of *Rhizopus* and *Trichoderma* were isolated. *Aspergillus candidus, Aspergillus niger, Penicilium corylophilum, Trichoderma viride* and white sterile mycelium were isolated from both rhizosphere and non rhizosphere, while *Aspergillus flavipes, A. fonsecaeus, A. itaconicus, A. versicolor, Fusarium moniliformae, F. poae, Penicilium citrium, P. rubrum, P. rugulosum, Rizopus nigricans* were isolated only from rhizosphere, while *Aspergillus awamori, A. effuses, Mucor hiemalis, M. racemosus* were isolated from non rhizosphere soil (Table no. 2).

From rhizosphere and non rhizosphere of *Azadirachta indica* total 32 species of 9 genera were isolated, among them 3 genera with 5 species from Zygomycotina and 6 genera with 27 species of Deuteromycotina and 1sterile mycelia. From rhizosphere soil total 21 species of 9 genera were isolated among them 3 genera with 3 species from Zygomycotina and 6 genera with 17 species of Deuteromycotina and 1 sterile mycelia were isolated while from non rhizosphere soil total 5 genera of 15 species were isolated, among them 1 genera with 1 species from Zygomycotina, 3 genera with 13 species of Deuteromycotina and 1 sterile mycelia. Among the total isolated fungi 16 species of *Aspergillus,* 5 species of *Fusarium* 3 species each of *Mucor* and *Penicilium and* single species each of *Rhizopus, Rhizoctonia, Trichoderma* and *Verticillium* were isolated. *Aspergillus candidus, Aspergillus flavipes, Fusarium sporotrichioid* and white sterile mycelia were isolated from both rhizosphere and non rhizosphere soil, while *Aspergillus atropurpureus, A. awamori, A. effusus, A. ochraceus, A. sulphureus, Fusarium moniliformae, F. oxysporum, F. poae, Mucor mucedo*, *Penicillum corylophilum*, *P. rubrum*, *Rizopus nigricans* were isolated only from non rhizosphere.
In all the three plants more numbers of fungi were isolated from rhizosphere soil than from nonrhizosphere soil. There is variation in rhizosphere fungi of all the 3 plants. Fungal colonization in the rhizosphere of medicinal plants has been widely studied. The distribution in the rhizosphere may vary depending upon host species, growing season soil properties, local climate and environmental factors (Rani et al.2017). This is almost first report on rhizosphere of *Murraya koenigii, Syzygium cumini* there are no earlier reports on this, while very fragmentary data is available on rhizosphere of *Azadirachta indica.* So this work is important and helpful to the researchers who would like to study the diversity of rhizosphere mycoflora of these medicinal plants. In all these isolations, members of Deuteromycotina and species of *Aspergillus* were dominant. From above results it can be concluded that Diversity of fungi is observed.

Total 23 species of 7 genera from rhizosphere and non rhizosphere of *Murraya koenigii.* Total 18 species of 6 genera from rhizosphere and non rhizosphere of *Syzygium cumini.* Total 32 species of 9 genera from rhizosphere and non rhizosphere of *Azadirachta indica.*More number of fungi was isolated from rhizosphere than non rhizosphere. Deuteromycotina and *Aspergillus* dominant throughout the studies.

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Water purification through Moringa Oleifera Lam.

Authors & affiliations:
D. J. Patel, J. Bhoi. and P. O. Pandey*
Department of Botany, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Water is used for variety of purposes drinking, washing, bathing etc. Reports of WHO (World Health Organization) says that about one million people lack safe drinking water and many people die every year from water borne diseases. To deal with this situation water purification is needed. There are various methods of water purfication, however biological methods are most promising and eco - friendly. The present study deals with use of Moringa oleifera to purify water. Moringa oleifera is grown in many countries as a multi -purpose tree as it possesses medicinal and nutritional values. Dry Moringa seeds and seed powder were used for water samples collected from different localities like water from Tube well and Well of four localities. These samples were tested for Total Hardness, Calcium Hardness, Magnesium Hardness, Acidity, Alkalanity, Acidity, TDS, TS, TSS, Chloride, Sulphate, Phosphate, pH, Temperature, etc. parameters before and after treatment. It is observed that Moringa oleifera Lam. Seed is an effective purifier and coagulant to treat variety of water samples. Use of seed powder is more promising as compared to entire seeds of Moringa.
Keywords: Moringa oleifera, Water purification, seed.

Introduction:

Water supply is basic need required for living creature and human. In this world available resources are limited for living creatures. Groundwater is one of the major sources of drinking water. But it's direct use for drinking is not suitable, for several reasons (Anekar N.R. 2017). Hence to make it suitable for drinking water treatment is essential. This study deals with water treatment using natural coagulant *Moringa oleifera*.

Importance – water is used for several purposes by humans such as, irrigation, cooking, washing, bathing, cleaning, drinking, also provides home to millions of creature, enables transportation. Regulates the temperature of Earth. Our body uses water in all its cells, organs and tissues to help regulate its bodily functions. Because our body loses water through breathing, sweating, digestion. It is important to rehydrate by drinking fluids and food containing water. Water is very important substance on earth. If there is no water there would be no life possible on earth.
These are several causes of water pollution such as sewage and wastage water, dumping garbage into river and accidental oil leakage, burning of fossil fuels, chemicals, fertilizers and pesticides, global warming, radioactive waste, urban development, animal wastage, underground storage leakage. According to United Nations estimates, the amount of waste water produced annually is about 1500 Km^3,6 times more water than that exist in all the rivers of world.

Purification –Though water pollution is a major problem but it can be overcome by various ways boiling, granular activated carbon adsorption, distillation, reverse osmosis, desalination, direct contact membrane distillation (DCMD), in situ chemical oxidation, bioremediation. Of those bioremediation is cheapest and healthier method mostly followed. For this variety of biological things can be used such as cilantro, jackfruit, java plum, peel of fruits, banana peels, along with these *Moringa Oleifera* Lam. is easily available in each corner of world.

MORINGA OLEIFERA LAM.

It is a tree of polypetalae most widely cultivated species in genus *Moringa*, the only genus in plant family Moringaceae common name include, *Moringa*, Drumstic tree , Horse Radish, Ben oil Or

Benzoil tree (oil derieved from seeds).*Moringa oleifera* is fast growing,drought resistant tree. Native to 'tropical and sub tropical' region of South Asia. Widely cultivated for its young seeds and pods and leaves used as vegetables. Also used for water purification. *M. oleifera* is considered to be an aggressive invasive species**.**

Water is used for several purposes by humans, but the level of purity of the water being consumed is very important since it has a direct effect on health. More than half of all illness caused by germs, which get into the mouth via water and food.Hence, purification of water is great need of time. This can be achieved through various ways. Chemical methods leads to purification by synthetic means however, use of biological methods are most convenient and eco-friendly. Hence these are more recommended.

There are various ways by which waters is purified biologically. Such as use of Jack fruit seeds : Prapat Pentamwa, Wanwisa Tanta, Pornwarin Milamai (2011) have studied effect of Jackfruit seeds on water in their work entitled "Water Treatment by Using Lychee, Jackfruit, and Rambutan seed coagulants".Muhammad Raziq Rahimi Kooh, Muhammad Khairud Dahri,Linda B.L. Lim (2016) also found effect of Jackfruit seed as sustainable adsorbent for the removal of Rhodamine B dye".Use of Coriander (cilantro) : Research done by undergraduate students at a community college, Douglas Ph. D. Said that Cilantro, also known as coriander shows activity as a new "Biosorbent" for removing lead and other toxic heavy metals from contaminated water.(Phys.org, 2013)Use of *Jatropa curcus* seeds : "Preliminary study on *Jatropa curcas* as coagulant in waste water treatment" had been studied by Zurina Zainal Abidin, N. Ismail, Robiah Yunus, I.S. Ahamad (2011) Robert Natumanya have worked on "Evaluating coagulant activity of locally available *Syzygium cumini, Artocarpus heterophyllus* and *Moringa oleifera* for treatment of community drinking water, Uganda."

Materials and Methods:
The study was conducted under a controlled environment in order to eliminate interference from human activities, rainfall and solar intensity.
Sample collection -The water samples were collected from Jalgaon district, Maharashtra. The two samples of water are taken. One of well and other tubewell. The geographical condition of Jalgaon i.e. latitude is $20^0$99"N and longitude is 75.5626^{0}E

Materials –Dry *Moringa oleifera* seeds are used in this study. These seeds were harvested from the trees of residential area of Jalgaon, Maharashtra, India. Seeds of *Moringa oleifera* act as natural coagulants and biocoagulants. Wings and coat from seeds were removed, fine powder was prepared and sieved. Test-tube, measuring jar, cylinder, beakers, glass rod, lids, test-tube holder, durham tubes, morter & pastle are required.

Procedure - Extraction of *Moringa* seed powder: mature seeds of *Moringa oleifera* were chosen from dry dehiscent pod. The outer covering of seed kernels were removed using a sharp knife, and using a laboratory mortar and pastle. The containers were cleaned thoroughly before taking the sample to avoid contamination. The initial volume of each sample was recorded at the collection point. The following parameters were determined: Temperature , PH , alkalinity , TDS , conductivity , oil grease , total hardness , calcium hardness , chloride hardness , phosphate , acidity.

In this procedure crushed seed powder 7.5 grams were mixed with 250 ml water sample for 7 days. And also 15 grams seed powder were soaked in 250 ml water for 14 days. Stir quickly for 30 seconds, then slowly and regularly for Five minutes. Water is covered without disturbing it for at least an hour. After 7 days and 14 days the extraction were filtered using filter paper. And water samples are collected. Again the water is treated and all parameters are measured such as acidity, TDS, oil grease, temperature, total hardness, calcium hardness, pH , chloride hardness , phosphate , conductivity, etc. The results of treated water samples were compared with raw water sample along with WHO standards.

Sr. No.	Parameters	Units	S1	S2	S3	S4 Limits	BIS
1	Total Hardness	Mg/l	320	290	330	460	300
2	Acidity	Mg/l	110	180	120	160	120
3	Alkalinity	Mg/l	90	230	390	390	200
4	Chloride	Mg/l	290	420	370	410	250
5	Calcium Hardness	Mg/l	100	180	150	200	200
6	PH		7.68	7.63	7.94	7.48	6.5 to 8.5
7	Temperature	0 c	33.3	33.8	33.5	33.4	
8	Sulphate	Mg/l	37.0	93.6	64.0	85.6	200
9	Phosphate	Mg/l	9.6	35.4	15.3	35.6	5
10	MPN	MPN/100ml	130	50	8	4	0/100ml
11	Magnesium Hardness	Mg/l	220	110	180	260	100
12	Total Dissolved Solids	Mg/l	460.4	415.1	317	410.2	500
13	Total Suspended Solids	Mg/l	189.2	121.65	160.85	204.05	No Standard
14	Total Solids	Mg/l	649.6	536.75	477.85	614.25	500
15	Turbidity	NTU	7	6.8	10.2	15.4	5
16	Ca ion Mg/l	60.9	77.7		48.3	73.5	75
17	Mg ion Mg/l	31.60	21.88		14.59	19.54	50

Table no. 2 Results obtained after Physio-chemical Study of Treated water with Seed Powder

	7 Days				14 Days			
Parameters	**S1**	**S2**	**S3**	**S4**	**S1**	**S2**	**S3**	**S4**
Total Hardness	280.00	250.00	277.00	247.00	270.00	309.00	351.00	305.00
Acidity	10.00	30.00	10.00	20.00	40.00	30.00	10.00	20.00
Alkalanity	310.00	230.00	90.00	290.00	620.00	230.00	210.00	260.00
Chloride	180.00	360.00	150.00	230.00	250.00	410.00	320.00	460.00
Calcium Hardness	144.00	139.00	139.00	103.00	150.00	148.00	168.00	117.00
PH	7.87	8.55	7.42	7.70	6.80	6.96	6.83	6.30
Temperature	23.80	24.20	23.60	23.30	24.00	23.90	24.10	23.50
Sulphate	25.80	80.30	54.10	75.30	25.10	81.40	55.20	77.40
Phosphate	7.20	24.20	7.55	24.60	7.70	23.10	7.20	24.20
MPN	130.00	50.00	8.00	4.00	130.00	50.00	8.00	4.00
Magnesium Hardness	136.00	111.00	138.00	144.00	120.00	161.00	183.00	188.00
Total Dissolved Solids	420.40	399.10	295.00	405.20	421.20	293.04	368.02	400.36
Total Suspended Solids	160.20	100.65	120.85	196.05	165.20	125.03	155.04	164.00
Total Solids	580.60	499.75	415.85	601.25	586.40	418.07	523.06	564.36
Turbidity	9.70	8.90	9.50	17.30	10.30	10.10	10.90	18.40
Ca ion	63.20	56.30	37.10	70.10	52.20	64.02	33.30	69.70
Mg ion	21.64	15.60	7.60	8.43	26.02	9.10	10.80	11.50

Table no. 3 Results obtained after Physio-chemical study of Treated water with Seeds

	7 Days				14 Days			
Parameters	**S1**	**S2**	**S3**	**S4**	**S1**	**S2**	**S3**	**S4**
Total Hardness	290	257	284	251	246	320	360	312
Acidity	30	20	40	20	30	40	20	30
Alkalanity	180	410	190	410	120	250	430	610
Chloride	310	450	850	350	360	280	140	320
Calcium Hardness	140	147	152	110	110	163	178	138
PH	8.28	8.30	8.05	7.96	6.89	8.36	6.82	7.62
Temperature	22.8	22.9	23.1	22.7	23.8	23.9	24.1	24.2
Sulphate	24.1	79.8	53.1	75.9	25.9	80.20	50.4	70.5
Phosphate	7.9	23.8	7.64	25.7	7.5	23.6	7.15	22.8
MPN	130	50	8	4	130	50	8	4
Magnesium Hardness	150	110	132	141	136	157	182	174
Total Dissolved Solids	428.69	392.5	283.20	428.21	400.36	421.02	256.08	274.32
Total Suspended Solids	156.08	104.96	126.78	209.34	164	165.02	155.01	144.02
Total Solids	584.77	497.46	409.98	637.55	564.36	586.02	411.09	518.34
Turbidity	7.9	8.6	9.2	16.7	9.6	8.8	9.4	17.2
Ca ion	56.3	70.1	38.1	63.2	53.4	71.02	32.4	61.1
Mg ion	20.80	11.44	10.2	9.12	19.40	10.12	9.8	9.2

Results and Discussion:

The results of the treated water samples were compared with raw water samples along with World Health Organization Standard. Total Dissolved Solids (TDS), Conductivity, Temperature, and pH of raw and treated water samples were determined. Turbidity and Total suspended solids were also determined.

According to Eilert (1978), the seeds of *Moringa oleifera* contains significant quantities of low molecular-weight water soluble protein which carries positive charge when the crushed seeds are added to the raw water, the proteins produce positive charges acting like magnets and attracting the predominantly negatively charged particles.

Aho, I. M. (2014) in his study reported that the coagulative efficiency of using *Moringa oleifera* seed extract is almost 100%, when compared with alum which is commonly used in conventional water treatment. Our study is in accordance with them as in terms of availability, *Moringa oleifera* seed or seed powder extract is a better alternative as compared to alum coagulant because it's high cost and non – biodegradability.

Raw water treated with *Moringa* seeds for seven days have shown positive results as compared to water treated for fourteen days. In seven days treatment, total hardness, Calcium hardness, Ca ions, Mg ions have been reduced and these values have come to standard range of BIS (Bureau of Indian standard). While pH and Chloride have increased in all samples those treated for seven days and treated for fourteen days.

If we compare these results sample wise then it is observed that sample number 3 which is sample of Tube well collected from residential area of Jalgaon city have shown positive results after treating with *Moringa* seeds and seed powder.

The experimental study includes treatment of raw water with *Moringa* seeds and another set of treatment of raw water with *Moringa* seed powder. Both treatments had shown improvement in

quality of water. However it is faster in water treated with seed powder which can be observed through values obtained from Table number 2 and 3.
Thus from this, we can conclude that use of *Moringa* seed powder as a water purifier is more effective as compared to entire seeds.
During this study two experimental sets were laid, one of seven days and another of 14 days. It has been observed that 7 days are quiet enough to purify water using *Moringa* seeds.

Conclusion:

From present experimental study following conclusions are drawn.
Moringa oleifera Lam. seed is an effective purifier and coagulant to treat variety of water samples.Use of seed powder is more promising as compared to entire seeds of *Moringa*.The water can be purified effectively using seed powder or seed within one week only.Quantity of *Moringa* seed powder required to purify one litre of raw water is around 30 gms.*Moringa oleifera* Lam. Being medicinal plant not only work as water purifier but also add and improve quality of water as it posseses many phytochemicals that have wide scope in medicines. The present study deals with biological method of water purification thus it is eco-friendly.

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Benign synthesis of silver nanoparticles and its application towards kinetic studies on degradation of azo dyes

Authors & affiliations:
A.D. Dhumale, D. V.Patil and Y. B. More*
Department of Chemistry, M J. College, Jalgaon, (M.S.) India

ABSTRACT:
Global researchers have been engaged in developing and improving a more effective method to deal with increasingly severe organic dye pollution. The field of nanotechnology is one of the most active field of modern material chemistry. The nanostructured material is mostly focus on current research of nanoscience and nanotechnology. In the present work, focus was benign synthesis of silver nanoparticles and environmental remediation by degradation of dyes like methyl orange found in waste water of dyes, textile, and pharmaceutical and chemical industries using synthesized silver nanoparticles. These silver nanoparticles were found to act as a potential catalyst for the degradation of methyl orange in the presence of sodium borohydride. Rate constants for the catalyzed and uncatalyzed reactions were determined. The catalyzed reaction spectrum had shown a sudden fall in absorbance value confirming catalytic effect of silver nanoparticles. No significant change in the absorbance in case of uncatalyzed reaction was observed, indicating very slow reduction rate of methyl orange. Thermodynamic parameters, such as free energy change ($\triangle G0$), enthalpy changes ($\triangle H\ 0$) and entropy change ($\triangle S0$) were calculated and their values indicates degradation of methyl orange process was spontaneous at high temperature and non-spontaneous at low temperature.
Keywords: Nanoparticles,Azodyes, Degradation, AgNPs

Introduction:

Azodyes are organic compounds bearing the functional group R−N=N−R′, in which R and R′ are usually aryl. They are a commercially important family of azo compounds, i.e. compounds containing the linkage C-N=N-C. Azo dyes are pervasively used to treat textiles, leather articles, and some foods. Chemically related to azo dyes are azo pigments, which are insoluble in water and other solvents [1}. An important and active area of water chemistry is the search for cheap, simple and effective methods to destroy unmanageable pollutants. Of these, azo dyes are by far the most popular class, for example, over 80% of direct and reactive dyes being azo. In many applications, once dying is complete a large amount of dye polluted waste water must be inclined and because of their high solubility in water, they are transported over long distances when discharged into water. The major and most hazardous threat to the modern world is the pollution caused by the industries for e.g. textiles, pulp and paper mills, distilleries and tanneries, where the processing of chemicals leads to generation of large quantities of coloured wastewater, which is eventually discharged into waterways. The colour of wastewater is attributed due to the presence of toxic dyes which contains a chromophore i.e, colour causing group [2-4]. These industries however may differ in the composition of colored compound present in waste water, where even a trace amount of dye is enough to impart colour to it. After China, our country is the second largest in the world which exports the dying stuffs, where 80% of the total dyes is used in textile industries alone [5]. The waste stream from dye factories can be intensely colored as well as potentially toxic. Great interest has been shown in the degradation of azo dyes to deliver a nontoxic, colorless effluent.

Moreover the effluent of dye components from industries forms a thin layer on the surface of water bodies thereby deteriorating the aquatic flora and fauna. Since dyes are highly complex, stable toxic pollutants in the environment over a longer duration, there is a high scope for exploring and developing various methods for their degradation.

Till date, there are various techniques which are used for the removal of toxic and unnatural dyes from

water bodies so as to minimize the risk of toxicity to the ecosystem. These techniques comprise the physical method such as membrane-filtration process (Nano filtration, electro dialysis, reverse osmosis) and sorption techniques. It was reported that adsorption is an effective and best equilibrium process for the removal of decontaminants from the waste water. The chemical method involves coagulation or flocculation, often integrated with filtration, ozonation , irradiation, adsorption, photolysis, ion pair extraction.

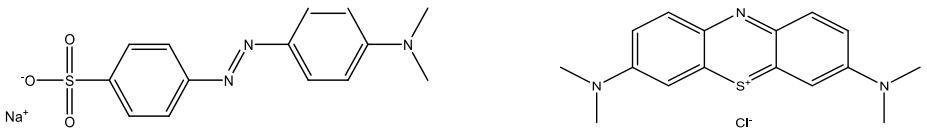


Fig 1: Structure of MO and MB

Materials and methods:

Synthesis of AgNPs (Chemical reduction method)

Preparation of solutions

1 MM Silver Nitrate (AgNo3)(The solution was prepared by dissolving 0.0085 gm of (AgNO3)in 50 ml of D/W, 2MM sodium Borohydride ($NaBH_4$)**(**The solution was prepared by dissolving 0.0037gm of $NaBH_4$ in 50 ml D/W), 0.01 M sodium chloride (NaCl)**(**The solution was prepared by dissolving 0.029 gm of NaCl in 50ml of D/W.)

Procedure:

10 ml 1mM of AgNo3 was taken in 50 ml beaker and kept on a magnetic stirrer. Freshly prepared 30 ml of 2mM $NaBH_4$ in ice cold water was added to above solution. The colour changes from black to orange and then yellow. After that 0.6 ml of 0.1 M stabilizing agent was added to above solution and stirred vigorously, till room temperature attained. The synthesized AgNPs were kept at room temperature for 24 hrs before using to let unreacted $NaBH_4$ escaped. The synthesis was achieved by using 1:3 molar ratio solution of Ag+ and $NaBH_4$.

3.2. Characterisation of Silver nanoparticles

The technological application of nano particles usually depends upon their structure. It is then difficult to manage the surface and thus the properties of individual particle. There is qualitative analysis of the surface of individual nano particles. The following methods are employed to characterize the nanoparticles.

SEM

The morphology of the product w

as examined by SEM. Fig.3 depicts the SEM image of nanoparticles. It shows that the Silver nanoparticles are flower shaped. The size of particle observed in SEM image is about 50-75 nm.

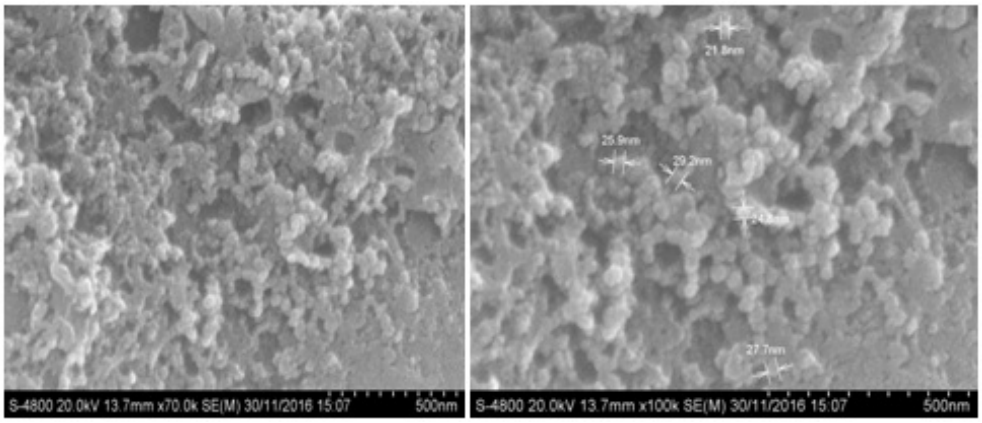


Fig 4: SEM image of the Ag nanoparticles

c) Preparation of methyl orange and methylene blue solutions.

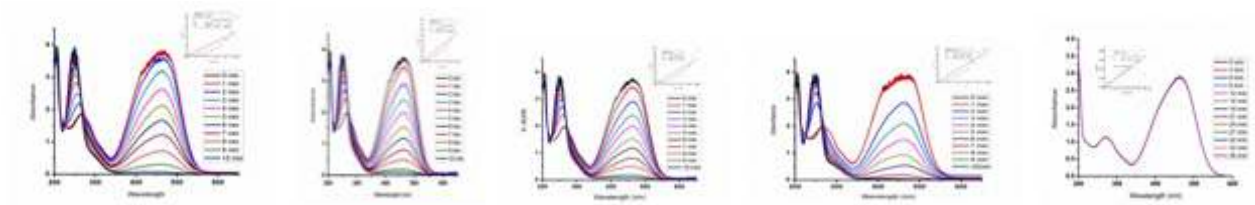
Methyl orange solution was prepared by crushing the 0.001mg methyl orange dye in a mortar and dissolved it in 50 ml distille water. Similarly methylene blue solution was prepared by dissolving 0.001 mg in 50 ml distilled water.

Results and Discussion:

a) Spectrophotometric analysis of degradation of MO and Mb dyes

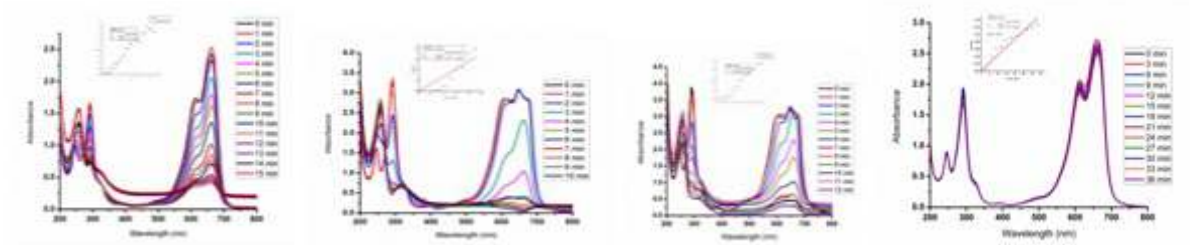
Catalytic Degradation of MO

Methyl orange solution acts as stock solution. About 0.001ml of AgNPs was mixed to 1ml of methyl orange solution along with 0.5 ml of $NaBH_4$ solution. This sample was monitored in interval of time period of 1 min, the suspension was measured against absorbance from 200-800 nm at temperatures 303K, 313K, 323K, 333K to evaluate the photo catalytic degradation of dye using UV-visible spectrophotometer.



Catalytic Degradation of Mb

Methylene blue solution acts as stock solution. About 0.001ml of AgNPs was mixed to 1ml of methylene blue solution along with 0.5 ml of $NaBH_4$ solution. This sample was monitored in interval of time period of 1 min, the suspension was measured against absorbance from 200-800 nm at temperatures 303K, 313K, 323K, 333K to evaluate the photocatalytic degradation of dye using UV-visible spectrophotometer.



b) Degradation kinetics of MO and Mb dyes

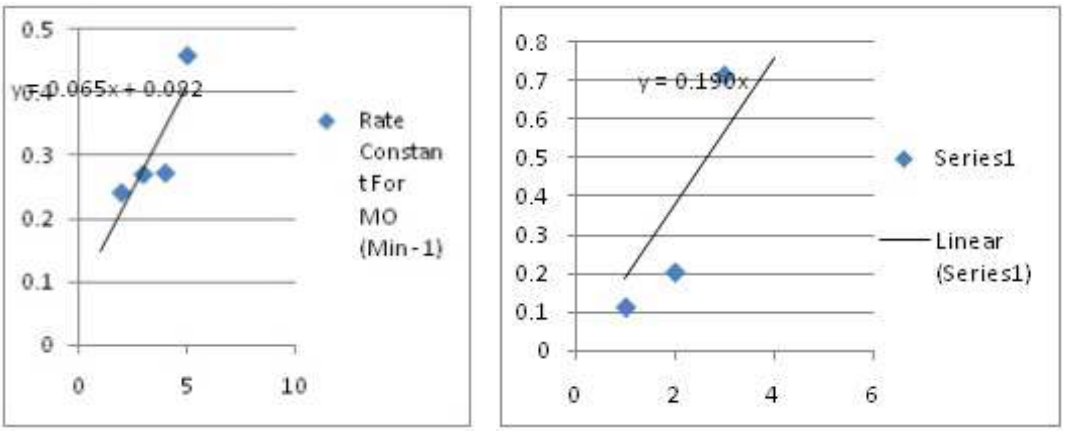


Fig 8: Temperature dependence graph for Degradation of MO and MB

Table: Rate constant for the degradation reaction of Methyl orange and Methylene blue

Temperature in Kelvin(K)	Rate Constant For MO (Min -1)	Rate Constant For Mb (Min -1)
303 K	0.242	0.113
313 K	0.271	0.2033
323 K	0.273	0.714
333 K	0.459	

Conclusions:

- The study suggests that synthesis route is low cost, environmental friendly and can be prepared in simple laboratory equipment in ambient condition.
- Silver nanoparticles stabilized by NaCl stabilizer has been successfully prepared by chemical reduction method.
- Qualitative characterization from the SEM and FTIR indicates that AgNPs have different size and may be of different shapes.
- The most active AgNPs was the one stabilized by with NaCl.
- In absence of AgNPs, the rate of degradation of Azo dyes is very slow. But on the addition of AgNPs into Azo dye, rate of degradation is enhanced significantly indicating the improved catalytic behavior of AgNPs.

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Benign synthesis of copper nanoparticles and its application towards degradation of methyl orange and methyl blue

Authors & affiliations:
M.M.Sethia, U.R.Patil, P.A.Malpure and Y. B. More.*
Department of chemistry, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
Global researchers have been engaged in developing and improving a more effective method to deal with increasingly severe organic dye pollution. The field of nanotechnology is one of the most active field of modern material chemistry. The nanostructured material is mostly focus on current research of nanoscience and nanotechnology. In the present work, focus was benign synthesis of copper nanoparticles and environmental remediation by degradation of dyes like methyl orange found in waste water of dyes, textile, and pharmaceutical and chemical industries using synthesized copper nanoparticles. These copper nanoparticles were found to act as a potential catalyst for the degradation of methyl orange in the presence of sodium borohydride. Rate constants for the catalyzed and uncatalyzed reactions were determined. The catalyzed reaction spectrum had shown a sudden fall in absorbance value confirming catalytic effect of copper nanoparticles. No significant change in the absorbance in case of uncatalyzed reaction was observed, indicating very slow reduction rate of methyl orange.
Keywords : Copper Nanoparticles, Methyl Orange, Rate constants.

Introduction:
Nanotechnology is rapidly enhancing field that making an influence on human life such as health, food, pharmaceuticals, chemical industry, energy science,cosmetics,Environmental- science and space industries. There are various way to synthesize Nano particles such as sol gel method, chemical reaction, solid state reaction and co-precipitation comparing to those methods green synthesis method is one of the best technique for the synthesis of Nano particles in the recent years. These method have diverse benefits namely low cost, simple, use of less toxic materials, most important is ecofriendly [1]. Nano technology has started leaving follow of laboratory and beat the new application to change our lives [2].Nanotechnology refer to an emerging field of science that includes synthesis and development of various Nanoparticles [3].
Biological degradation of azodyes: Micro Organisms can be used to completely degrade the azodyes because microorganism reduce the azodye by secreting enzymes such as Laccase, azoreductase, peroxidase and Hydrogenase the reduced form of azodyes are further minerlizedinto simpler compounds and are utilized as their energy source.[4]
The current research involves use of metallic nanoparticles, i.e. copper, Gold, Silver nanoparticles for the degradation of methyl orange as model dyes for assessing its photo catalytic potential. The copper, gold and silver nanoparticles were synthesized by chemical reduction approach. The synthesized CuNPs were characterized through techniques, which includes i.e. Fourier Transform Infrared Spectroscopy (FTIR) and Scanning electron microscopy (SEM). The catalytic activity in the presence of UV light of copper nanoparticles was studied at different temperatures and finally the rate and extent of decolourisation and degradation was modeled using existing equations.

Experimental Work:
Synthesis of CuNPs (Chemical reduction method)
10 ml 1mM of$CuCl_2$, was taken in 50 ml beaker and kept on a Magnetic stirrer. Freshly prepared 30 ml of 2mM $NaBH_4$ in ice cold water was add to above solution. The color changes from black to orange and then yellow. After that 0.6 ml of 0.1 M stabilizing agent was added to abovesolution and stirred vigorously, till room temperature attained.

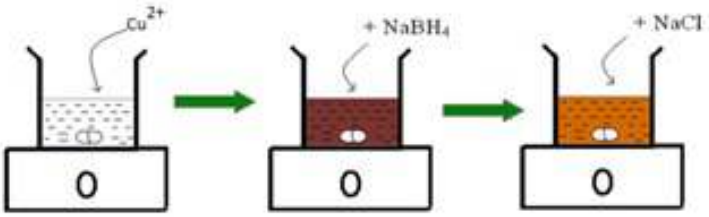
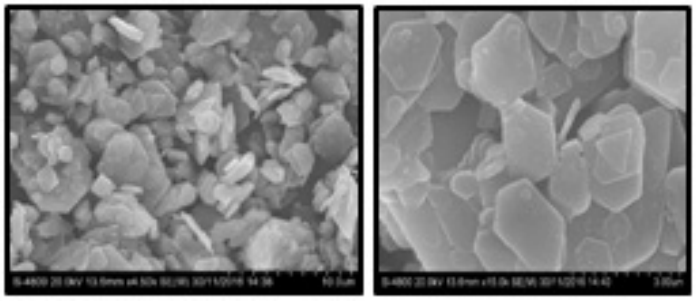


Fig 1: Step wise procedure for synthesis of Cu nanoparticles stabilized by NaCl.

Results and Discussions:

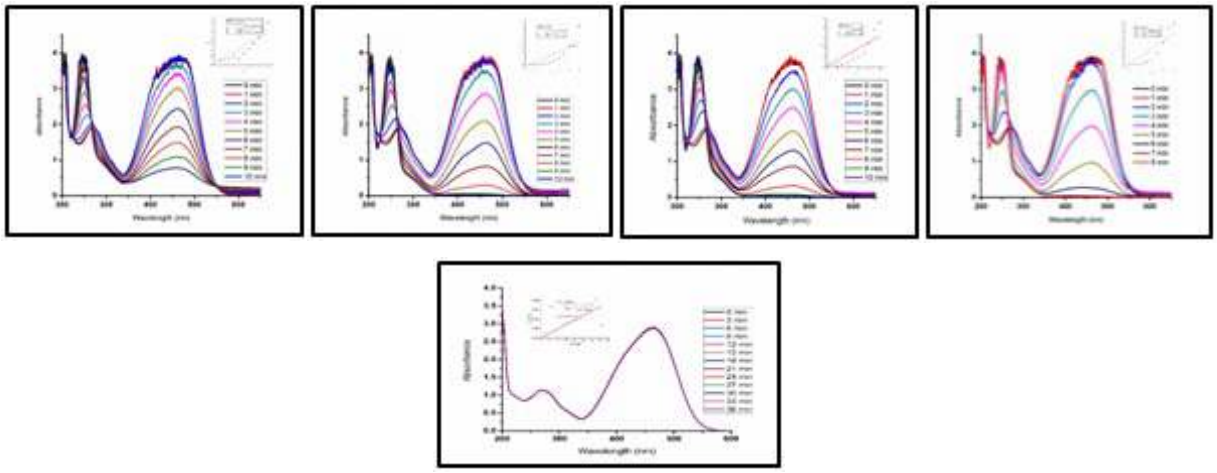
3.1. Morphology Analysis of CuNPs:

SEM is a surface imaging method, fully capable of resolving different particle sizes, size distributions, nanomaterial shapes and the surface morphology of the synthesized particles at the micro and Nano scale. Using SEM, we can probe the morphology of the particles and derive a histogram from the image by either by measuring and counting the particles manually, or by using specific software[5]. In the present work copper nitrate has been used to prepare CuO Nano particles with spherical shape via precipitation method, Anandan et al who reported synthesis CuO crystallites self-organized into spherical assemblies or "dandelions" with a puffy appearance. [3]

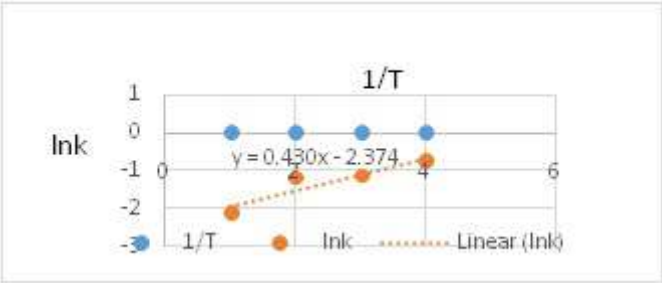


3.2 Catalytic degradation of methyl orange

The sample was monitored in interval of time period of one minute, the suspension was measured against absorbance from 200-800nm at temperature 303K, 313K, 323K, 333K, to evaluate the photocatalytic degradation of dye using UV-visible spectrophotometer.



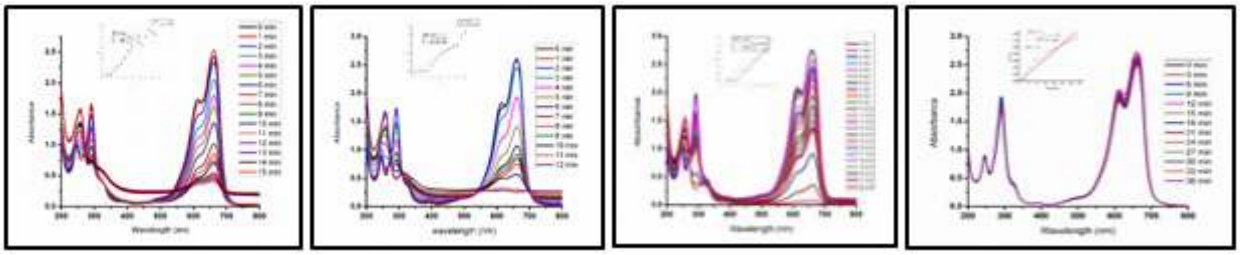
Temperature	1/T	Rate constant(k)	lnk
1. 303k	0.0033	0.117	-2.145
2. 313k	0.0031	0.306	-1.184
3. 323k	0.0030	0.319	-1.142
4. 333k	0.0030	0.484	-0.725



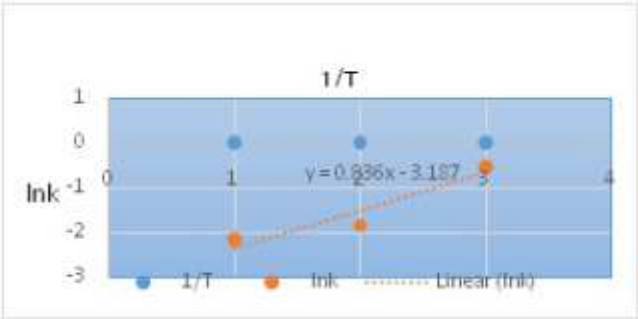
Slope= 0.4302 ; Intercept=-2.3745

3.3 Catalytic degradation of methyl blue:

The sample was monitored in interval of time period of one minute, the suspension was measured against absorbance from 200-800nm at temperature 303K, 313K, 323K, to evaluate the photocatalytic degradation of dye using UV-visible spectrophotometer.



Temperature	1/T	Rate constant(k)	lnk
1.303k	0.0033	0.113	-2.180
2.313k	0.0031	0.156	-1.857
3.323k	0.0030	0.602	-0.507



Slope= 0.8365; Intercept= -3.1877

Conclusion:

1. The study suggests that synthesis route is low cost, environmental friendly and can be prepared in simple laboratory equipment in ambient condition.
2. Copper nanoparticles stabilized by NaCl stabilizer has been successfully prepared by chemical reduction method.
3. Qualitative characterization from the SEM indicates that CuNPs have different size and may be of different shapes.
4. In absence of CuNPs, the rate of degradation of Azo dyes is very slow. But on the addition of CuNPs into Azo dye, rate of degradation is enhanced significantly indicating the improved catalytic behavior of CuNPs.
5. Rate constant for methyl orange is 0.4302 and rate constant for methyl blue is 0.8365.

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Synthesis and characterization of curcumin loaded nano-emulsion

Authors & affiliations:
G. Desale, A. More, V. Palwe and P. D. Baviskar*
Department of Chemistry, M. J. College, Jalgaon (M.S.), India.

ABSTRACT:
Curcumin is a natural polyphenolic compound with potent anticancer, anti-inflammatory, and antioxidant activities. However, the solubility and bioavailability of curcumin is less as compared with the traditional mechanical methods, ultrasound is a superior tool to obtain nanoemulsion with smaller and homogeneous globule size and physical stability. The goal of this study was to develop a curcumin nanoemulsion by ultrasonication, containing a high curcumin load, small droplet size and good physical stability. The composition and preparation method effects on entrapment efficiency, droplet size, polydispersity index, and zeta potential of the nanoemulsions were evaluated. Curcumin nanoemulsions were successfully prepared by combined thin-film hydration emulsification and ultrasonication methods, the this method 8- 10 % oleic acid was used in the form of oil phase where as 1% mixture of surfactants (Teween 20 and Span 20) was use as a aqueous phase. The stable emulsions are isolated after centrifugation and micelle size of nano-emulsion was measured by using particle size analyzer and zeta potential also measured.
Keywords: curcumin, nano-emulsions, emulsification, ultrasonication, surfactants.

Introduction

Curcumin, the bioactive component obtained by the extraction and purification of ground rhizomes of Curcuma longa has been found to exert wide range of beneficial biological and pharmacological activities including antioxidant, anti inflammatory, antimicrobial and anticancer in clinical trials [1]. But studies addressing the metabolism and uptake of curcumin had shown that either no curcumin or little amount was detected in serum or tissue after administration. The main reasons attributing to the low bioavailability are supposed to be the poor solubility of curcumin in aqueous media, rapid hydrolysis followed by molecular fragmentation at physiological pH and inactivity of its metabolic products. To improve the bioavailability of curcumin, numerous approaches have been undertaken by many researchers including, preparation of nanocurcumin, liposomal curcumin, curcumin phospholipid complex, and chelation with metals micro / nano encapsulation technology of poorly water soluble bioactives has attracted wide attention in the food and pharmaceutical industry for the past few years for various applications like protection of bioactivity and their controlled release for improving bioavailability[2]. The encapsulation of polyphenols overcome the drawbacks of their instability, alleviates unpleasant tastes or flavors, as well as improves the half-life of the compound in vivo and in vitro.

The use of surfactant mixtures is a common practice in the industry and it has been extensively studied by many researches. Combining two or more surfactants in a formulation may be beneficial, not only for the reduction of interfacial tension already mentioned (leading to smaller droplet sizes for the same energy expenditure) but for the decrease of the overall surfactant concentration required to produce stable emulsions, as opposed to single surfactant systems. [8,9] Using the a forementioned approach, a nanometric droplet size minimum is attained and the corresponding formulation is used to develop a mixing procedure using a high-shear, thin film-spinning device[10]. The thin film-spinning (TFS) apparatus has impellers that spin a film of the fluid to the walls of a cylindrical vessel at very high speed, and under cavitation-free conditions. The resulting flow field induces a very high shear and droplets of the dispersed phase are fast and efficiently reduced in size. These apparatus can handle high throughputs; the small model used in this work may produce up to 40 kg/h using a 15 mL vessel and a 25 cm × 30 cm footprint.

Therefore, this study aimed at producing nanoemulsions for encapsulating turmeric extract, and incorporating the dried encapsulated extract into minced pork to make functional canned ham. Considering that the ham would be directed to Korean consumers, Korean turmeric was used for developing the formulation as it is preferred there because of its mild color and flavor [16]. The physicochemical characteristics of the nanoemulsions were determined, and sensory evaluation of the canned ham was also performed [17]

Material And Methods

The sample of curcumin used in this study was isolated from curcumin plant by using column chromatographic separation. Oleic acid, Tween 20 and Span 20 were obtained from MooljiJaitha college Jalgaon. Water was double distilled using a distillation assembly. It is worth noting that all the components are pharmaceutically acceptable for oral administration and fall under GRAS (Generally Regarded as Safe) category.

Formulation of emulsions

This study began with the evaluation of the formulation parameters required to produce stable oil-in-water emulsions to be used as carriers of curcumin in the oil droplets. In this sense, two non-ionic surfactants were mixed in varying concentrations and proportions to yield different hydrophilic-lipophilic balances (HLB) were evaluated. The surfactants evaluated were Tween 20 and Span 20. The first is a polyoxyethylenesorbitan monooleate hydrophilic emulsifier; the second is a lipofilic emulsifier consisting of sorbitan monooleate. As mentioned above, these two surfactants were mixed using different T20/S20 w/w ratios in order to obtain variable HLB values.The total surfactant mixture concentration was also evaluated; the oil ratio in the emulsion varied in a relatively narrow range (12%-18% w/w) while the water content was constant (80% w/w). Table 1 shows the formulation parameters involved in this study. The screening procedure was as follows. First, the surfactants were homogenously mixed in the aqueous phase. Then, the organic phase was added into the aqueous solution and the mixture was stirred by hand to produce an emulsion; the resulting droplet size was measured and the rate of phase separation was monitored during one week after emulsification. This procedure led to the formation of oil-in-water emulsions, stable enough to be characterized. The formulation that allowed for the smallest droplet size and the most stable emulsion (least phase separation) was selected for the remaining tests.

Curcumin dissolution profile

Since curcumin has a poor solubility in the oil phase, ethanol was used to achieve the total dissolution of the target amount, 5 mg/mL, in the oil phase. The dissolution profile of curcumin was carried out as follows. First, powdered curcumin was added into 15 mL centrifuge tubes containing the mixture of surfactants and liquid paraffin, selected as described in the previous section. Various amounts of ethanol calculated based on surfactant plus alcohol mass (0% ethanol-100% surfactants; 2% ethanol-98% surfactants, 4% ethanol-96% surfactants, 6% ethanol-94% surfactants and 8% ethanol–92% surfactants w/w) were also added to the mix in order to facilitate curcumin dissolution. Then, the tubes were heated at 50°C for 30 min and stirred overnight in a shaker at room temperature.While the degree of curcumin dissolution could be easily determined by visual inspection of the tubes, it was decided that obtaining the curcumin partitioning between the oil and the water phases in the emulsion, was more informative. To this end, the content of 3 mL centrifuge tubes was mixed with water and stirred using a magnetic stirrer at 400 rpm, during 1 min, to produce an oil-in-water emulsion containing 65% w/w of water. Immediately, the emulsions were centrifuged at 3000 g for 5 min, to induce phase separation. The curcumin content of both the oil separated phase and the precipitated solid were measured by visible UV, as described in section. By this means, the undisclosed curcumin could be determined as well as the curcumin that could have transferred to the water phase, by computing the difference between the total curcumin added to the system and the amount that ended in the oil phase and the solid precipitate.

Nano Emulsification of Formulation

High speed homogenization and probe-sonincation.

The emulsifying particles is also expected to be increased the effectiveness of the item covering capacity or infiltration activity for instance. One of the important characteristics of an emulsion is its distribution of the droplet sizes, is usually used as a part of experimental examination due to its geometry of the valve which can impacts the droplet disturbance. A stable sub-micron emulsions or nano-emulsion in low-viscosity fluids also can be created by using high speed homogenizers. Two important Factors of the drops, size and distribution magnitude in order to attain good distribution of particle size the emulsions was kept for high speed homogenization forat 7000 rpm for 30 min before ultrasonication treatment.The nanoformulation of curcumin was achieved by ultrasonication method. In this method curcumin loaded oil phase with surfactants in appropriate proportion as mentioned above and water was kept for ultrasonication by using prob-sonnicator at 150 w for 25 min in ice bath. the nano size of emulsion was charecterise by particle size analyzer and zeta potential measurement.

Result and Discussion

Characterization

Particle size Measurement

The particle size of nanoemulsions are analysed employing photon correlation spectroscopy (PCS) using Malvern Zetasizer, which monitors the variation in light scattering because of Brownian motion of particles as function of time. PCS is based on the principle that the particles with small size travels with higher velocity as compared to particles with large size. The laser beam gets diffracted by sub-micron particles present in solution. Due to diffusion of particles, rapid fluctuations in laser scattering intensity occur around a mean value at a fixed angle and this is dependent upon particle size. The calculated photoelectron time correlation function generates a histogram of the line width distribution that can be related to the size of particle. For measuring particle size, weighed amount of formulation is dispersed in double-distilled water for obtaining homogenous dispersion and that has to be used instantly for measuring the particle size. The particle size was found to be 244 nm as shown in figure 01.

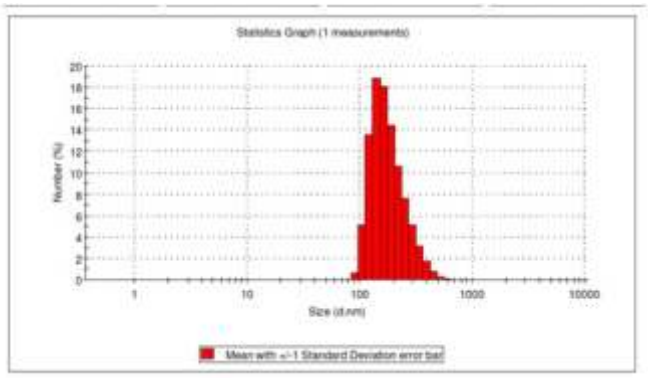


Figure 01 : The statistical data of particle size analysis by particle size analyzer

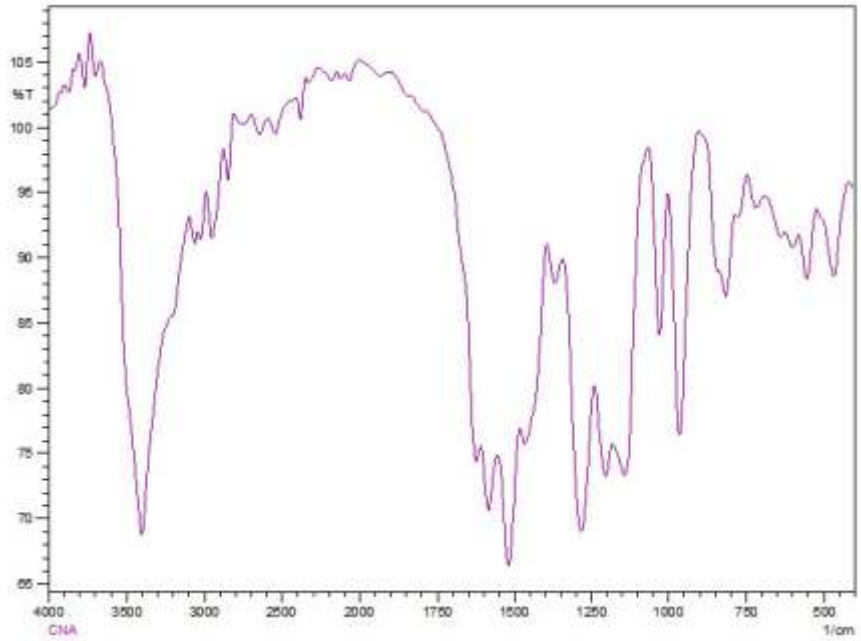
Zeta Potential Measurement

The zeta potential is a method for measuring surface charge of particles when it is placed in liquid. Zeta potential is used for predicting dispersion stability and its value depends on physicochemical property of drug, polymer, vehicle, presence of electrolytes and their adsorption. It is measured by Malvern Zetasizer instrument. For measuring zeta potential, nanoemulsion is diluted and its value is estimated from the electrophoretic mobility of oil droplets. Zeta potential of ±30 mV is believed to be sufficient for ensuring physical stability of nanoemulsion. In the present study zeta potential was recorded about 14 mV.

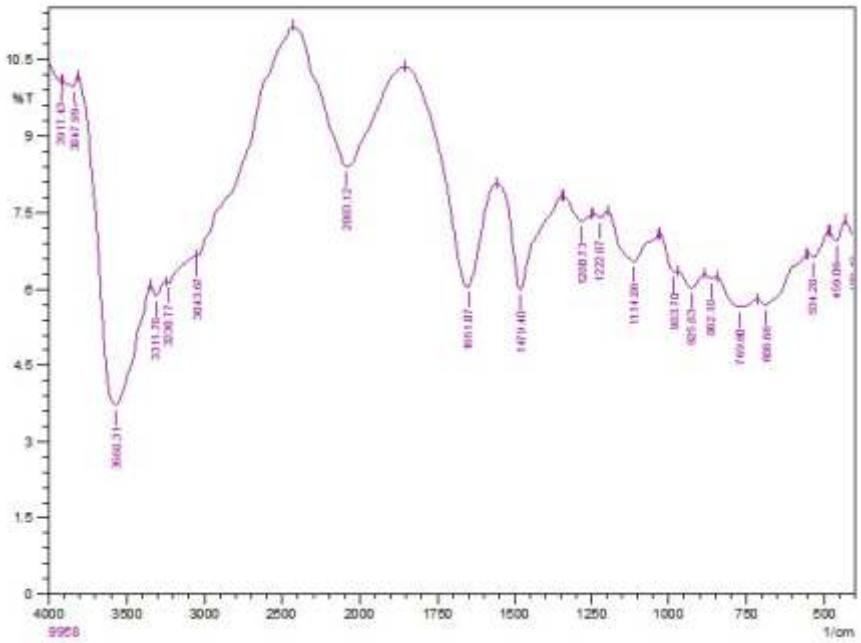
Fourier-transform infrared (FTIR) spectral analysis

The most of the spectral data of curcumin in remain as it is in its nano emulsion form.stretching on the polysaccharide skeleton. In the case of curcuminincorporatednanostarch, the broad peak around 3400 cm-1 indicatesthe stretching of hydrogen bonded -OH groups. In the FTIR spectrum of curcumin, the functional groups such as hydroxyl group, carbonyl group and the ethylene group showed peaks at 3429 cm-1, 1782.66 cm-1 and 1514.12 cm-1respectively. For the curcumin incorporated nanostarch, the peaks corresponding to these functional groupswere observed at 3439 cm-1, 1629 cm-1 and 1508 cm-1 respectively, which indicates that the major peaks of curcumin were retained in the case of curcumin incorporated nanoform also. In the spectrum of curcumin, the peaks at 752.11 cm-1, 823.25 cm-1and 966.23 cm-1 indicated the bending vibrations of -CH bond of alkene group.

FT-IR spectra of free curcumin (A)



FT-IR spectra of nanoemulsion (B)



Conclusion

Curcumin being highly unstable and hydrophobic is difficult to incorporate in aqueous food systems. For this reason the curcumin was encapsulated inside carrier oil in an emulsion form. The results in the present study showed that curcumin was easily loaded in emulsion having oil phase in nanometer size. The drug formulation involved modification in surface of drug using nanotechnology causes huge effect on action and performance of drug. These results have important implication in the design of nanoemulsions of curcumin for commercial applications. It is concluded that the nanoencapsulation of highly lipophilic and unstable compounds is an effective platform to increase the hydrophilicity, bioaccessibility and to protect them from degradation.

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Synthesis of azo disperse dyes from 2-amino thiazole and their UV study

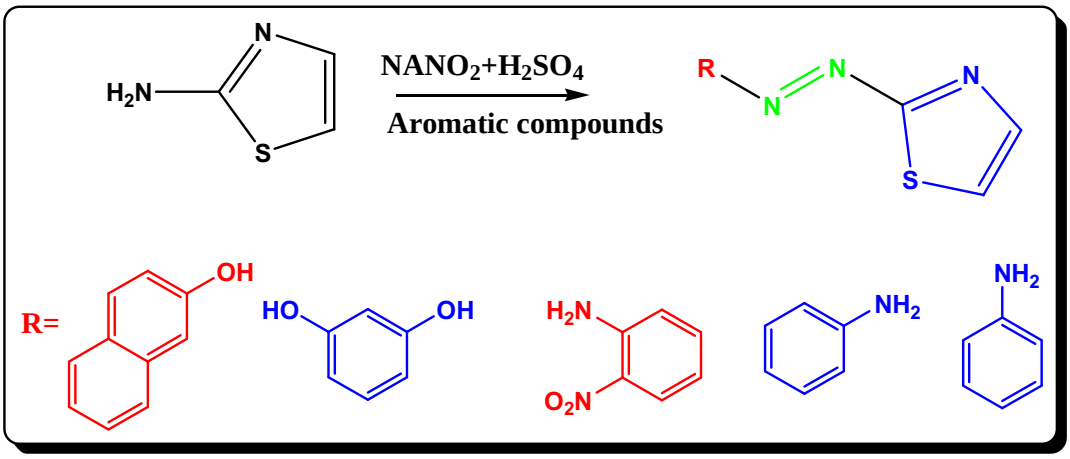
Authors & affiliations:
S. Sachdev, R. Sharma and J.D.Bhirud*
Department of Chemistry, M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
We report herein synthesis of substituted (thiazol-2-yl)diazenyl) derivatives in two steps. Initially diazotization of 2-aminothiazole was takes place in presences of sodium nitrite, then this diazotized product undergo coupling reaction with β-Napthol/ recorcinol/ m-nitro aniline/ O-amino benzoic acid/ P- amino benzoic acid, yield azo dyes in good yield.We increased the yield by adjusting the pH to 7-8 by addition of aqueous solution of KOH. The synthesized dyes were characterized by IR spectral analysis. The UV-visible study shows effects of solvent polarity on the maximum absorption wavelength. The results shows that most of the dyes showed positive solvatochromism when the solvent is changed to a more polar solvents.The synthesized dyes were found to give various shades of red.
Key words: Azo dyes, coupling reaction, diazotization, solvatochromism.

Introduction:
Azo dyes are the most essential group of disperse dyes. Disperse dye is one kind of organic substances which is free of ionizing group. Disperse dye are less soluble in water and used for dyeing synthetic textile materials. They are valuable because, universally constitute morethan 50% of disperse dyes having strong tinctorial strength, ease of synthesis and low cost of manufacture [1]. It is well known that azo compounds are the widest class of industrial synthesised organic dyes owing to their versatile application in various fields, such as dyeing textile fibre, biological-pharmacological activities, and advanced application in organic synthesis [2–5]. It was our main objective to synthesize the disperse dyes consisting thiazole and azo substituent. We synthesized dyes from diazotized aminothiazole coupled with β-Napthol/ recorcinol/ m-nitro aniline/ O-amino benzoic acid/ p- amino benzoic acid with adjusting pH for improvement in yield. The synthesized dyes were characterized by IR spectral studies and solvatochromic behavior was studies from visible absorption spectra.

Materials and methods:
All reagents used were of analytical grade. Solvents were distilled before use. Melting points are determined in open capillaries and are uncorrected. IR spectra were recorded on Shimadzu FT-IR (Affinity model) using KBr .Thin layer chromatography plates were obtained from silica gel slurry prepared in Chloroform. The TLC was performed Using N-Hexane: Ethyl acetate (80:20%).

Experimental:
A finely ground powder of 2-aminothiazole (1.0 g, 0.01 mole) was added to a mixture of 12 ml of Sulphuric acid and 10 ml water. Cooled to 0-5°C in an ice bath equipped with a magnetic stirrer. Sodium nitrite (0.82g, 0.011 mole) dissolve in 5ml of distilled water was added drop wise over a period of 30 minutes into above solution,cooled to 0-5°C. The resulting diazonium solution was used immediately in the coupling reaction. The diazonium solution was added into a mixture of the coupling components β-Napthol/ recorcinol/ m-nitro aniline/ O-amino benzoic acid/ p- amino benzoic acid in 2 M 30ml NaOH (0.01mole) and stirred for at least 2 h. Raising the pH to 7-8 (with aqueous pottasium hydroxide) prior to filtering and water washing to neutral pH. The resulting product was filtered, washed with water, and recrystallized from ethanol to give required products.



Scheme-1: General scheme for Synthesis of Disperse Azo dyes

Result and discussion:

We report herein synthesis of Azo disperse dyes in two steps. Initially diazotization of 2-aminothiazole was takes place in presences of sodium nitrite, then this diazotized product undergo coupling reaction with β-Napthol/ recorcinol/ m-nitro aniline/ O-amino benzoic acid/ P- amino benzoic acidyield azodyes in good yield. Initially, we take β-Napthol for coupling reaction with diazotised product of 2-aminothazole but yield of the product was very poor. Then we try to improve yield by maintaining the pH of solution by adding dilute acid but product are soluble in acidic solution. Then we add aqueous solution of KOH and adjust the pH to 7-8 we got excellent yield. In this way we improve the yield of reaction by using simple way.

FT-IR Study of synthesized Dyes A-E:

The synthesized dyes were characterized by IR spectral analysis by using KBr. IR spectra of dyes recorded band at 1475-1515 cm^{-1} is due to N=N stretching of azo group. The absorption band appeared at 1537-1589 and band at1529-1581cm^{-1} indicate C=C and C=N stretching of thiazole ring. In addition of peakes, dyes (**A**) and (**B**) contains –OH group, shows broad band at 3050-3500 and 2569-3410cm^{-1} respectively. A Dyes (**C**) to (**E**) shows N-H stretching frequency at 3518-3545cm^{-1} due to NH_2group. The FT-IR spectra fully support for the formation of azo dyes.

Table 1: FT-IR starching frequencies of synthesized dyes in cm-1

Dyes No.	-OH	N=N	N-H	C=C	C=N
A	3050-3500	1505	--	1556	1650
B	2569-3410	1475	--	1589	1629
C	--	1430	3529	1529	1655
D	--	1510	3545	1537	1681
E	--	1515	3518	1540	1643

UV-Visible study of synthesized dyes A-E:

The visible absorption spectra of the synthesized dyes was taken in acetone, dimethyl formamide (DMF), methanol and chloroform, this was to compare the various solvent polarity for the synthesized dyes. From the results summarized in **Table 2** it can be seen that the dyes showed higher absorption wavelength in DMF. The effects of solvent polarity on the maximum absorption

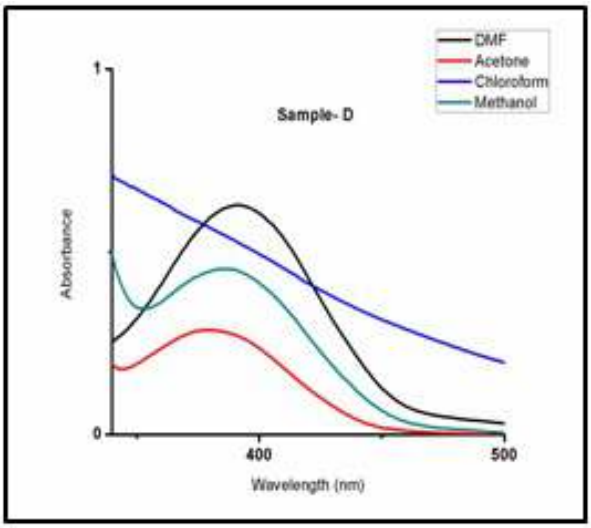
wavelength of the dyes were also studied. The results shows that most of the dyes showed positive solvatochromism when the solvent is changed to a more polar solvents. For example dye **(A)** absorbs at 480 nm in chloroform but when the solvent is changed to a more polar solvent, DMF it absorbs at 490nm indicating a bathochromic shift of 10nm. Also dye **(B)** exhibits this same effect as it absorbs at 442nm in chloroform and 450nm in DMF. Dyes **(C)** show solvatochromism of 13nm, as it absorbs at 358 nm in chloroform and 371 in DMF also dye **(D)** show the greatest effects of positive solvatochromismbecause it absorbs at 350nm in chloroform and 391nm in DMF indicating a bathochromic shift of +41 nm.

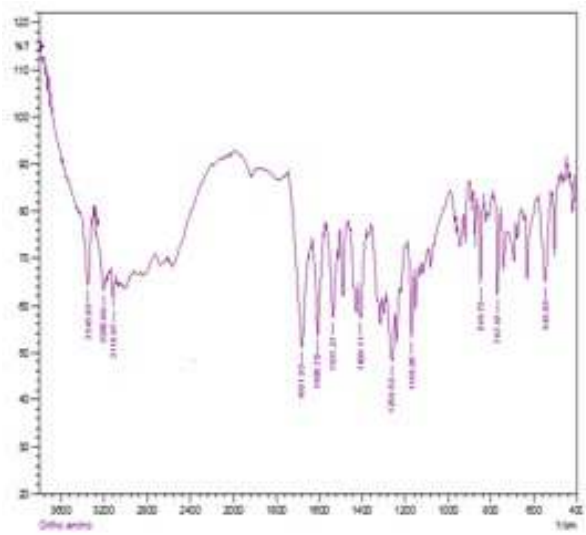
Some of the dyes however, showed negative sovatochromism when solvent is changed to a more polar solvents. For example dyes **(E)** absorbs at 455nm in chloroform and when the solvent was changed to DMF it absorbs at 450nm indicating a negative solvatochromism (hypsochromic shift) of -5nm.

Table 2 Visible Absorption Spectra of Synthesized Dyes λ_{max}(nm)

Dye No.	Acetone	DMF	Methanol	Chloform
A	485	490	476	480
B	448	450	447	442
C	363	371	350	358
D	378	391	386	350
E	450	450	460	455

Figure-1: UV Visible and FT-IR Spectra of Synthesized Dyes D





Conclusion:

A series azo disperse dyes were synthesized in this work. This synthesis was obtained using conventional methods with improvement in yield by adjusting pH (with aqueous pottasium hydroxide). Dyes were characterized by IR spectral studies. The solvatochromic behavior in various solvents was evaluated. The results indicated that λmax values of dyes were strongly dependent on polarity of solvents and as polarity increases they show bathochromic shifts. The absorption maxima of dyes shifted bathochromically in the sequence DMF>methanol>acetone>chloroform, except dye **(E).** The values of λmax were less in low polar solvent and more in high polar solvent.

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Breadboard simulator

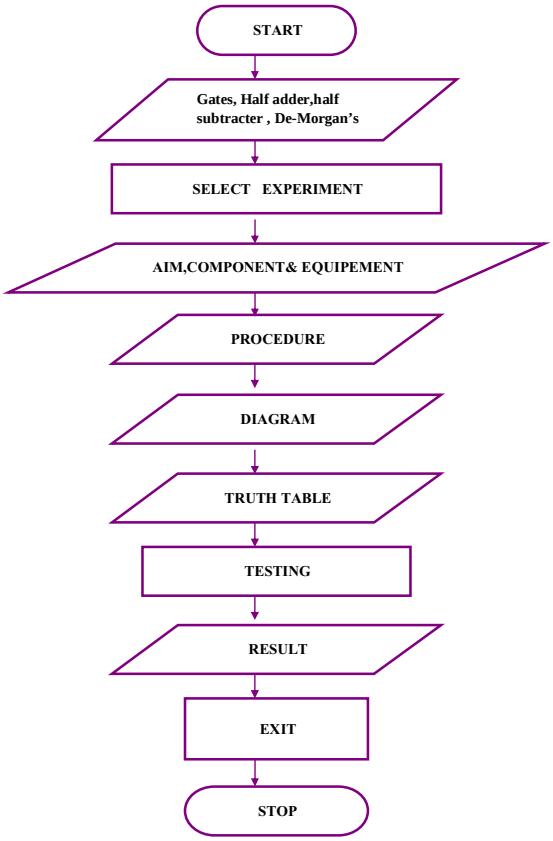
Authors & affiliations:
M. A.Chaudhari , H. A.Patil and L. Y. Bhole*
Department of Computer Science and Information Technology,
M. J. College, Jalgaon, (M.S) India.

ABSTRACT:
The model presents an innovative idea for implementing basic electronic circuitry without using electronic chips & components. This work presents the working of all electronic circuits through software. This software is useful for students to understand basic electronic concepts through animation, which makes teaching interesting and easy to understand. The software includes functioning of all basic gates like adder, subtractor etc. It provides drop down menu through which any circuit can selected for study.
Keywords: breadboard, simulator

Introduction:

The software present an innovative idea for implementing the basic electronic chips and components. This work represents software working of all electronic circuits through software. This software is useful for user to understand the basic electronic concepts through animation. A gate is defined as a digital circuit which follows some logical relationship between the input and output voltages. It is a digital circuit which either allows a signal to pass through as stop, it is called a gate.George Boole in 1980 invented a different kind of algebra based on binary nature at the logic, this algebra of logic called BOOLEAN ALGEBRA. A logical statement can have only two values, such as HIGH/LOW, ON/OFF, CLOSED/OPEN, YES/NO, TRUE/FALSE, CONDUCTING/NON-CONDUCTING etc.

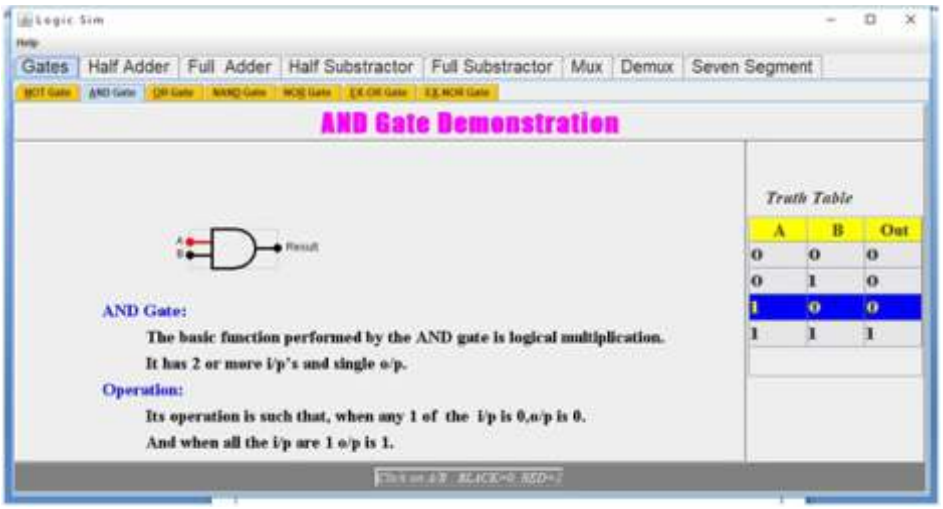
Methodology:



This software deals with the basic digital electronics practicals by using logic gates. In this software we can perform the digital practical like all the basic gates like AND, OR, NOT Likewise, Half adder, Full adder, sFullsubstractor Half subtrctor etc. By selecting any one of the practical from menu option system show us its first aim, components, equipment then we have to click on procedure menu which display procedure of that practical. In the same way we can see diagram, truth table & result.

Result and Discussions:

Software Testing is important element of Software quality assurance and represents ultimate review of specification, design and coding. A successful test is one of that uncover as yet undiscovered error. Testing demonstrate that software functions appear. To have been met. Also data collected as testing is conducted provides a good indication of software reliability and quality. It is general principle of testing that all test should be traceable to customer requirements.



Test to be performed	Pass	Fail
1.Checking input of changes of each basic gates.	✓	-
2.Blinking of Truth Table	✓	-
3.Displaying of diagram	✓	-
4.Displaying aim of each function	✓	-

Conclusion:

This is easy way to perform the digital experiment of electronic Like basic gates AND, OR etc, adder, subtracter, etc. This system works only for digital electronic components. We will try to add all type of electronic components like micro processor, sinusoidal etc.

Teachers can use this system for demonstration.

It is useful for all electronics department.

This system does not give facility to print experiment sheets.

we will try to add this facility.

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Doctor-coordinator

Authors & affiliations:
A.K.Zinzurao, A.R.Kasar and A.C.Patil*
Department of Computer Science and Information Technology
M. J. College, Jalgaon, (M.S.) India.

ABSTRACT:
A Doctor is a someone who maintains or restores human health through the practice of medicine. Doctor helps in healing people when they sick or have themselves. They diagnose the condition, and prescribe medicines and other treatment that need to be taken to get back to normal health.
To help the doctor we have created this website. This website is useful to access current status of patient. This status will be filled by the nurse of particular ward. In this website, We have given the login/signup links to access accounts of particular doctor/nurse.
Keywords: Doctor login, Nurse Login, New patient Registration, ICU, General etc.

Introduction:

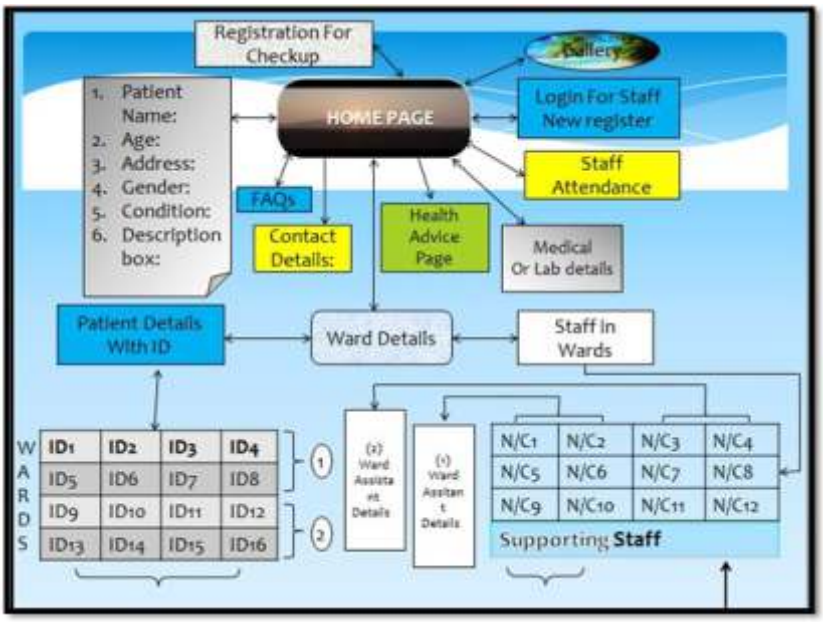
"Doctor-Coordinator" is the best website to view the current status of the patient. The Specialist Doctor can view its patients current status anytime and anywhere by using desktop or on his/her mobile phone.
With the help of his doctor can cure patient accurately and doctor can also give the instruction to the staff of nurse.
The Benefits of the website is given below:

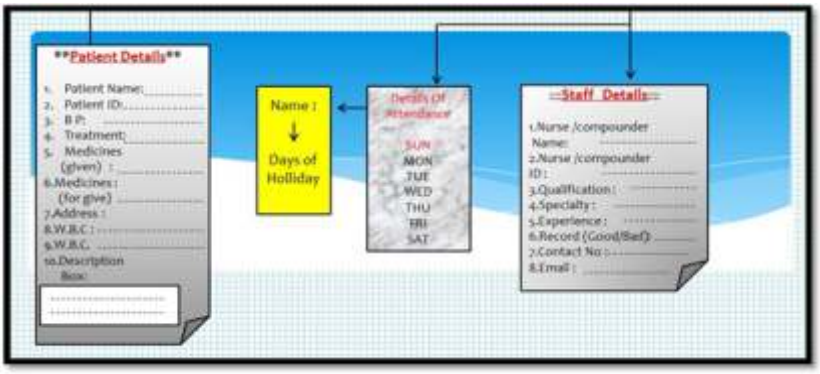
- Using this website, a particular doctor can access the current status of the patient anytime and anywhere by using desktop or on his/her mobile phone.
- This makes the doctor helpful to cure patient.
- The idea about this website is that staff (nurse/ ward boy) have right to fill the status of the patient by accessing his/her own account.
  Example – what are the tablet given by the doctor to patient, heartbeats, Blood pressure and other information. etc.
- Every nurse have special id and password to access their account.
- Nurse can register new patient easily.
- Nurse can also visit this website by scaning QR code.

Methodology:

Today in the area of the health care, a major issue is the provision of adequate and effective health for the elderly, as people aged 65 and older are the fastest growing segment in the population. Technology holds good potential for improvements in the field of health care used in the intelligent environments.
In this website, Doctor can view the information/current status of the patient.

Structure of this Website:





Result:

The result of this website is given below :

- This website is made to access current status of the patient. This website is useful for doctor and nurse only. This is not public website.
- **This include two levels of users:-**
  - Doctor level
  - Nurse level
- The Website include : -
  - Maintaining patient details
  - Providing Security ie. noother can access the Doctor/Nurse login information
  - Doctor can give the advice to nurse such that what will the next treatment to patient.
  - Providing and maintaining Two kinds of compartment :-
  - ICU
  - General

Conclusion:

A doctor is also a human being, doctor cannot check the patient status in every 15 minutes or he/she cannot view the patient current status as he wanted to see anytime an anywhere. This website is useful for the doctor to view the current status of the patient.

From the above mentioned result, it is concluded that "Doctor-Coordinator" is the website which is

coordinator between patient and doctor. Here Doctor is the admin of the website and doctor have rights can to access the server in which website is present.

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File utility manager

Authors & affiliations:
J.D.Kulkarni, P.C.Mahajan and A. S.Patil*
Department of Computer Science and Information Technology,
M. J. College, Jalgaon, (M.S) India.
email: apshreya@gmail.com

ABSTRACT:
The people using computer mostly deals with files & various operations related with files to store data into files, retrieve data from files, send/receive files etc. So, the problems they might face are hacking of data, a time delay due to transmission of data, copy of garbled data etc. Sometimes splitting of large files is required to transfer them over the network/Internet & at the same time merging of these files is required after their reception. Comparison of files is required to check their sizes, extensions etc. Encryption & decryption is required for security purpose. Voice recording can be used for entertainment as well as information sharing purpose.

So it will be very beneficial for computer users if all above mentioned operations are done with the help of a single & simple software.

As per the time limits , the purpose of the system

1.Large sized files can be easily splitted & then merged, for easy transfer.
2.Fixed as well as random memory size can be chosen by the user for splitting the specified file.
3.Text files can be encrypted or decrypted for the security purpose.
4.Two files can be compared & appropriate message is displayed as per the result.
5.Voice can be easily recorded.

Introduction:

As the present world is very much dependant on Computers, the job of the programmers is increased. The programmers deals with handling of files, performs various operations on files to store & retrieve the data. Normally every programmer deals with transferring of files. So the problems they might face is hacking of data , a time delay due to transmission of data , copy of garbled data, etc. System provides a set of great utilities to the users. The security issues like Encryption, Decryption, etc.

are provided, so that data is safely transferred without the fear of information leaking. Transmission of huge data files may engage network for a long time resulting the starvation of other clients. To avoid this inconsistency, the Split & Join utilities are provided in the system

Following utilities are provided by File Utility Manager.

File can be encrypted, folders can be locked for security purpose :-

The security issues like Encryption, Decryption etc are provided by the System, so that data is safely transferred without the fear of information leaking.

Splitting of file is useful while file size is big :

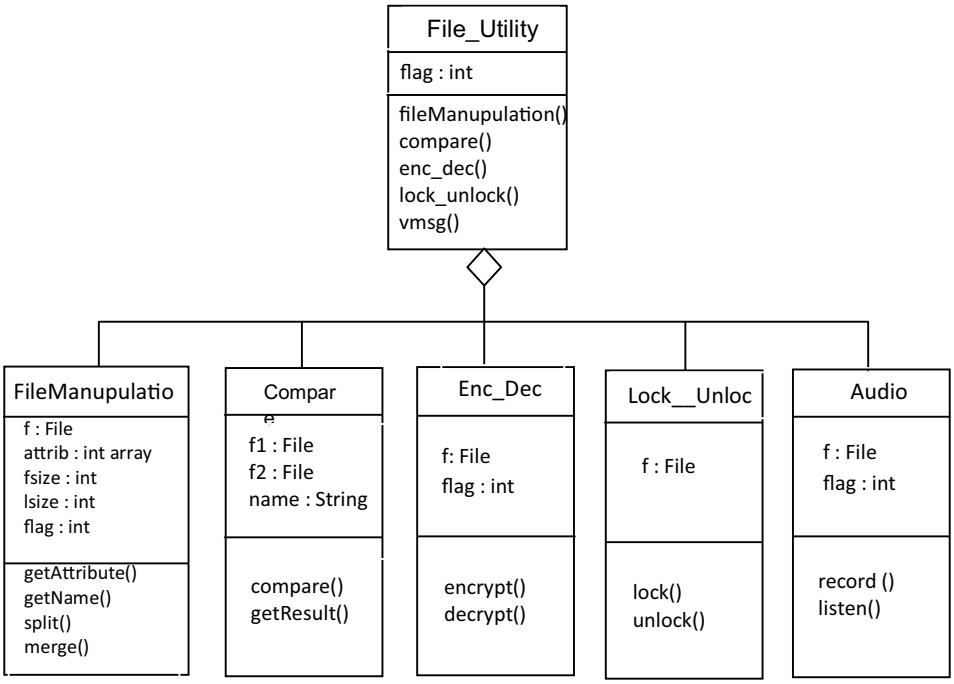
Transmission of huge data files may engage network for a long time resulting the starvation of other clients. To avoid this inconsistency, the Split & Merge utilities are provided in the system.

The system is user friendly :

The system saves the cost of training to its users. The system interacts with the users efficiently. To use the system the user interface provided is very easy to interact. Proper messages will be displayed to the users while using the system. The system interacts with its users by giving options.

Portability :
The system works on any machine on any O.S since we used Java language which is platform independent. The system is independent of the hardware aspects. Just the necessity is of acquiring it in the application architecture to give the functionality.
Simplicity of system :
The system uses English like words while interacting with its users. So that users can feel simplicity of the system. The errors done by the users are displayed to them using English like statements.

Class Diagram:



Conclusion:
While developing the system, a lot of efforts have been taken. From this system we came to know several facts. The logical schema of the system should be designed properly. The analysis of the system should be independent of its implementation. The system should be thoroughly examined before developing it. The sound knowledge of the developing environment is must. Analysis of system plays a vital role. We have successfully completed this project. Working on this project is new experience for us.

From the results obtained, it can be seen that it is possible to meet our objective for the "File Utility System".

The management is fully satisfied with this project and by using this software the user is benefited.

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I-library

Authors & affiliations:
S. D. Ghongade, K.D. Kathore and G.V. Jawale*
Department of Computer Science and Information Technology,
M. J. College, Jalgaon, (M.S) India.

ABSTRACT:
Vision impairments can result from variety of causes, including congenital conditions, injury, eye disease and brain trauma or other conditions such as diabetes and multiple sclerosis. India has the largest blind population of the 37 million people were visually impaired across the globe among which 15 million people are from India. Blind students can read only by audio books, Braille or with the help of a personal assistant. The scope of this project is to provide technical solution and to assist the visually impaired people to access various text resources and enhance their knowledge. This is an Innovative idea for the blind people to listen number of audio books of their choice using android application. Voice assistant in android application will help the blind people in making and efficient use of an application in listening number of audio books that are to be provided on the application. All type of audio books will be provided for the blind people. Not only blind people,person with normal vision can also handle the application using the voice commands.

Introduction:
If you are person with a normal vision you probably spend your time staring at screens. Laptops, Computers, Mobiles phone and Tablets are being our windows to the world. We primarily interact with them easily with our eyes but if you don't have any vision then we feel we are totally cut down to this technology, knowledge and Internet. But the truth is that we don't need site to use such application. Thanks to this simply accessible feature of modern technology that makes these blind peoples to come in contact with this technology and can easily handle these devices. One can easily access the smartphone browse the websites and much more with the help of your voice itself.
There are many such applications in favor of blind people so that they can access smartphones and search various content on internet. So it was necessary to develop such application which can bring them in contact with the knowledge and literature.
So, I-Library is an innovative Concept for Blind people so that they can easily access the mobile application and listen number of audio books of their choice and gain more and more knowledge about literature, social science and other fields of knowledge.

Scope of Work:
As blind cannot primarily interact with the devices using their vision so it was necessary to develop an application that can work on theirvoice commands. They should be easily available on the devices which are affordable to blind people. So this application is built in android platform using JAVA CORE language.
We have gone through various internet blogs and YouTube videos so that we can easily build and application helpful for the blind people. We have collected some data from internet blogs which proved to be the most helpful one for planning concept of this application. The application whole and soul works on voice assistant so that no other person's help is needed while using this application. The exciting feature of voice assistant in android proved to be the best part so we didn't use any other voice API for this application. The inbuilt voice assistant was called and activated for the achieving the goal.

Methods:
This application is for blind people so it was important to develop such application which blind people can easily access using their voice commands. This application is built in android platform using

android studio JAVA CORE language. The exciting and dynamic feature of android studio helped in developing the android application.

- The text to speech is activated using following codes.

```
private void askSpeechInput(){
Intent intent = new Intent(RecongizerIntent,ACTION_RECOGNIZE_SPEECH);
Intent.putExtra(RecognizerIntent.EXTRA_LANGUAGE_MODEL,
RecognizerIntent,LANGUAGE_MODEL_FREE_FORM);
intent.putExtra(RecognizerIntent.EXTRA_LANGUAGE, Local.getDefault());
intent.putExtra(RecognizerIntent.EXTRA_PROMPT,"Hii speak Something");
Try{
startActivityForResult(intent,REQ_CODE_SPEECH_INPUT);
}0 catch (ActivityNotFoundException a){
Toast.makeText(this,"sfsf",Toast.LENGTH_SHORT).show();
}
```

- The particular time is given to user to give response this response is recognized by the code which is called as such

```
ring= MediaPlayer,create(MainActivity.this,raw,hello);
ring,start();
ring.setOnCompletionListener(new MediaPlayer.OnCompletionListener(){
@override
public void onCompletion(MediaPlayermediaPlayer){
}
});
simpleList =(ListView) findViewById(R.id.simpleListView);
CustomerAdaptercustomAdapter =new CustomAdapter(getApplicationContext(),name,img);
SimpleList.setAdapter(customerAdapter);
```

- The application is static application therefore the application don't need any external database.

Result and Discussion:

Analysis:

Blind people due the lack of vision face the problem while getting in contact with the knowledge and literary work. It was important to get through this and make the blind people aware about the knowledge treasure that normal can easily access and get through it. Many such applications are developed in the sake blind people for browsing websites, using smartphones and other activities which normal person can carry out easily.

Application:

I-Library is built in android studio using java core language. We have built this static application so no external database is used for the storing the audiobooks. No external voice API is used for this application as per exciting feature of android studio we have just activated the voice assistant which is just inbuilt in the android studio. The application prompts each and every stage of browsing so that it becomes easy for the user to handle the application easily and effectively.

Conclusion:

This application is built using the android studio in java core language so that it can easily be handled on every smartphone device. This application will totally work on voice assistant so that from start to end this application will be easy to handle for the blind people. The prompting feature of android application makes it easy for the blind people to get to know about the categories and types of books the listener wish to listen.

Acknowledgement:
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