ISBN:978-98-84663-33-9

Life Science Research and Sustainable Development

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M.Sc. Ph.D.

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RUT Printer and Publication Jalna 431 203

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ISBN: 978-98-84663-33-9

Type Setter : Shri Yogesh Urdukhe

Cover Page Designed by : Shri Nitin Muley

Edition : April 2023

Price: Free

RUT Printer and Publication Jalna 431 203

Preface

J.E.S College, Jalna in collaboration with Dr Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhajinagar had organised two days National Conference on 3rd and 4th March, 2023. The motto of the conference was 'Present and future perspective of Life Sciences research for Sustainable Development and Biodiversity Conservation'

Prof. Waykar, Hon. Dean of Science Faculty, Dr Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhajinagar was the chief guest for the inaugural ceremony whereas Dr. Ganesh Agnihotri, Hon. Principal, J.E.S., College, Jalna was the the chairperson of the inaugural function.

Prof. Waykar in his inaugural address said that before civilization man was very close to the nature and the nature was pristine. Pure water, and fertile soil was available everywhere. But now due to developed technology and industrialization nothing is pure as we didn't focus on sustainable development. So we are facing a lot of problems today.

Prof. Waykar further added that the explosion of population is putting extra burdon in natural resources. We have built dams, polluted the environment, cut trees and using natural resources indiscriminately.

Hon. Dean in his address mentioned that today we are using fertilizers and pesticides extensively to increase the agricultural produce but it's destroying in microbials available in the soil so soil is becoming less fertile and there are no nutrients in the food due to poor quality of the soil. The challenge before us is to provide untoxic food to our population, he added. We have done tremendous damage to the biodiversity.

The govt. of India had to run a Cancer train to transport cancer patients in the state of Punjab this is because there is tremendous poisonous elements in the soil that people consume with food. Green Revolution produced enough food for us but farmers used exessive fertilizers and pesticides in the farming and we are facing the results of wrong farming methods today. Our short sighted development plans are responsible for such disasters in the world.

In his Presidential address Principal Dr. Ganesh Agnihotri mentioned that today is the World Wild day so we have chosen this day for the National conference. He further added that if we back our culture sustainable development will automatically happen. We will have to manage all the resources properly. People today are suffering from throat cancer due to extensive use of plastic cups for tea and coffee. He stated that if we are looking for the change then we should go back to the nature that will solve all our problems.

The resource person for the first session was Dr. Hiware sir, the former professor and Head of the Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, and Aurangabad.

Dr. Hiware talked about Agro Climatic Zones in India according to different states.

He said that the major problem in agriculture today is the small and fragmented holding. Soil erosion is another problem we are facing, he added. Furthermore the agriculture is unorganised sector and climate change is making it even worse. The increase in the global temperature is the most important issue we are dealing with today. Changes in the seasonal cycles are failing our crops and farmers and are committing suicides.

Prof. Hiware told that we need to give alternative to the farmers. Sericulture is the best alternative and agricultural agriculture based industry. Its product is silk. Sericulture is an important labour intensive forest. Dr. Hiware added that it is the only cash crop that provides attractive income to the farming community in general and to the small and marginal farmers in particular.

There is no impact of climate change on the sericulture. The beauty of this crop is farmers can take 10 crops in a year and if a crop fails there is no impact on the farmer's income. So farmers get income in every month. While speaking about the labour, he said that farmers have to work only for twenty days. They don't have to go in sun to work. After the leaves are brought, they have to work inside the shade house.

Silk is the queen of the fabrics. It is known for its purity and luster. It is natural based protein fiber, this is how Dr. Hiware described silk as a fiber. In Maharashtra we have Mulberry and Tassar silk culture.

While stating the importance of sericulture he said that it has a high employment potential and it is a women friendly occupation. It is an important agro based enterprise adding value to the lives of farmers. Sericulture needs low investment but it gives high returns. This is an eco-friendly and integrated farming. Sericulture has the potential to stop the migration of people to the bug cities. The demand of the silk has always been several times more than its supply.

Prof. Hiware provided all the details about the sericulture in his address- cum- presentation. His guidance gave new ensight to the students and the participants of the conference. He talked about various enterprises related to the sericulture and silk production to the weaving.

All the delagates liked his presentation very much.

The second session of the conference started at 2pm after the lunch.

Prof. N. N Bandela was the keynote speaker on the topic of Bioindicators and their applications in the environment studies.

Prof. Bandela said that Bio-indicators are biological indicators of environmental quality characterizing environmental conditions. He further added that Bio- indication is a research activity allowing us to obtain a picture of the ecological situation on the basis of its important element e.g. species, ecological form, population and association of community.

There has been drastic changes on Ecology and Ecosystem after the Vietnam War. After Tsunami, earthquakes in Killari and COVID there have been tremendous changes in the ecosystem.

While speaking about the pollution he said that we can only dilute the pollution,we cannot eradicate it. If we have to conserve the biodiversity, we will have to take tremendous efforts, Prof. Bandela said.

Dr. Bandela stated that students should do research which will benefit the society and country. He gave various examples to explain the imfact of pollution and loss of biodiversity in the country and around the world.

Prof. Ajay Kumar Jadhav from Department of Microbiology, Govt. Institute of Science, Chatrapati Sambhajinagar was the resource person for the fourth session. Dr. Ajay Kumar spoke about 'The role of Micro-organisms in sustainable development.' He gave detailed presentation regarding the how microorganisms are so important in the sustainable development. He said that only 1% of all Micro-organisms cause illnesses and only 4% of all bacteria cause plant diseases. Dr. Ajay Kumar told that we should never underestimate the power of Microbes in our life.

Dr. Mamta Goyal presented vote of thanks after the last session.

Participant: 89 Paper Published in Book – 22

Paper Presented in conference - 60 (Oral and Poster)

Organising Secretory

Prof. Laxmikant Shinde

Content

Mutation Breeding: Tool for crop improvement

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Keywords:
Mutation breeding, mutagens, applications, achievements.

Abstract:
This study is comprehensive overview of the various techniques and workflows available to researchers today in the field of molecular breeding, and how these tools complement the ones already used in traditional breeding. Both genetic and phenotypic screens are evaluated. Mutation breeding technique has played a major role in generation of climate smart varieties. Plant mutagenesis is rapidly coming of age in the aftermath of recent developments in high-resolution molecular and biochemical techniques. By combining the high variation of mutagenesis populations with novel screening methods, traits that are almost impossible to identify by conventional breeding are now being developed and characterized at the molecular level.

Introduction:

Mutation breeding technique has played a major role in generation of climate smart varieties. These crop varieties have been shown to with stand wide range of environmental fluctuation. Globally millions of hectares of cultivated land have been devoted for the cultivation of this mutant crop varieties and intern billons of revenue have been generated (Jain, 2010).

The main objective of mutation breeding is to increase food production and provide sustainable nutrition (Goyal *et al*., 2009 and Wani *et al*., 2011). World Food Security (FAO) on food plan action observed that, "Food security at the individual, household, national and global level exists where all people at all times have physical and economic access to sufficient, safe and nutritious food to meet their dietary needs and food preferences for an active and healthy life''. Plant breeders and farmers are under pressure to sustain food production under the climatic changes. The food prices are continuously increasing up worldwide in both developed and developing countries.

The mutant varieties have been grown on large scale by farmers in their fields, and increase in food production resulted from cultivation of the mutant varieties could be translated into increased food security, since this would be accessible for the people in need. Mutation breeding programme should be clearly planned and should be large enough with sufficient facilities to screen large population (Wani *et al.,*2017; Raina *et al.,* 2016).

In mutation breeding, desirable mutations are induced in crop plants with the use of physical or chemical mutagens (Raina *et al.,* 2017). The variability generated through induced mutations are either released as new variety or used as the parent for subsequent hybridization programmes. Conventional breeding in combination with other techniques such as mutagenesis, biotechnology, genetic engineering or molecular breeding utilize local genetic resources for developing new cultivars that could handle frequent climatic changes (Amin *et al*., 2016). Mutation breeding is known to induce genetic variability in the crops that show higher yield and wider adaptability (Khursheed *et al.,* 2016).

Types of mutations

The term "mutation" used in 1900 by Hugo de Vries, to describe phenotypic changes which were heritable. However, the term 'mutation' is now used in a rather strict sense to cover only those changes which alter the chemical structure of a gene at molecular level, are commonly called gene mutations or point mutations. Structural changes in the chromosomes viz., deletions, duplication, inversions and translocations also bring about phenotypic changes in plants and animals, are called chromosomal mutations.

Mutations are classified depending upon the magnitude of phenotypic effect produced by them into two groups (Gaul 1964).

(i) **Macro mutations**: In this type produce a large recognizable phenotype effect on individual plants. These are oligogenic in nature and can be easily selected in the M2 generation

(ii) **Micro mutations**: These types of mutations produce a small phenotypic effect that can be identified only on the basis of a population. These are polygenic in nature and selection for such mutations can be delayed till M3 or later generations.

Types of Mutagens: In mutation breeding the agents available and responsible for induction of mutations called mutagens. They are categorized into two classes: a) Physical and b) Chemical mutagens.

Table .1: Physical mutagens

Mutagen	Source	Characteristics	Hazard
X-rays	X-ray machine	Electromagnetic radiation; penetrates tissues from a few millimeters to many centimeters	Dangerous, penetrating
Gamma rays	Radioisotopes and nuclear reaction	Electromagnetic radiation produced by radioisotopes and nuclear reactors; very penetrating into tissues; sources are ^{60}Co (Cobalt-60) and ^{137}Cs (Caesium-137)	Dangerous, very penetrating
Neutrons	Nuclear reactors or accelerators	There are different types (fast, slow, thermal); produced in nuclear reactors; uncharged particles; penetrate tissues to many centimeters; source is ^{235}U	Very hazardous
Beta particles	Radioactive isotopes or accelerators	Produced in particle accelerators or from radioisotopes; are electrons; ionize; shallowly penetrating; sources include ^{32}P and ^{14}C	May be dangerous
Alpha particles	Radioisotopes	Derived from radioisotopes; a helium nucleus capable of heavy ionization; very shallowly penetrating	Very dangerous
Protons	Nuclear reactors or accelerators	Produced in nuclear reactors and accelerators; derived from hydrogen nucleus; penetrate tissues up to several centimeters	Very dangerous
Ion beam	Particle accelerators	Produced positively charged ions are accelerated at a high speed (around 20%–80% of the speed of light) deposit high energy on a target	Dangerous

Table.2: Chemical mutagens

Mutagen group	Example	Mode of action
Alkylating agents	1-methyl-1-nitrosourea (MNU); 1-ethyl-1-nitrosourea (ENU); methyl methane sulphonate (MMS); ethyl methane sulphonate (EMS); dimethyl sulphate (DMS); diethyl sulphate (DES).	React with bases and add methyl or ethyl groups and, depending on the affected atom, the alkylated base may then degrade to yield an abasic site, which is mutagenic and recombinogenic, or mispair to result in mutations upon DNA replication.
Azide	Sodium azide	Same as alkylating agents.
Hydroxylamine	Hydroxylamine	Same as alkylating agents.
Antibiotics	Actinomycin D; mitomycin C; azaserine; streptonigrin	Chromosomal aberrations also reported to cause cytoplasmic male sterility.
Nitrous acid	Nitrous acid	Acts through deamination, the replacement of cytosine by uracil, which can pair with adenine and thus through subsequent cycles of replication lead to transitions.
Acridines	Acridine orange	Intercalate between DNA bases thereby causing a distortion of the DNA double helix and the DNA polymerase in turn recognizes this stretch as an additional base and inserts an extra base opposite this stretched (intercalated) molecule. This results in frame shifts, i.e. an alteration of the reading frame.
Base analogues	5-bromouracil (5-BU); maleic hydrazide; 5-bromodeoxyuridine; 2-aminopurine (2AP)	Incorporate into DNA in place of the normal bases during DNA replication thereby causing transitions (purine to purine or pyrimidine to pyrimidine); and tautomerization (existing in two forms which interconvert into each other, e.g. guanine can exist in keto or enol forms).

Discussion

In mutation breeding, concept of induced mutation for the improvement of crop fore run to the beginning of 20th century. During the past 89 years, for the crop improvement through mutation breeding plays a crucial role and also support the pace made using traditional methods of plant breeding (Amin *et al.,* 2015). Due to emergence of induced mutation which plays a vital role in the development of crop varieties with desirable features all over the world, cultivation of different crop varieties has continued to be successful over the past five decades.

The plant breeding programs due to use of induced mutants across all over the world has led to official release of 3,362 mutant plant varieties from 240 different plant species in more than 75 countries throughout the world (FAO/IAEA, 2020). Varieties developed by mutation breeding increase the biodiversity due to presence of variation among them which serves as a baseline for

conventional plant breeding and directly contribute toward the conservation and use of plant genetic resources.

To induce genetic variability in various crops different mutagens have been used by various breeders. Since the discovery of mutation effect of X-rays in the 1920s Lewis John Stadler has recognized induction of mutation as a potential technique for crop improvement (Shu *et al.,* 2012). The first disease resistant mutant was reported in barley in 1942 (Usharani & Ananda Kumar, 2015). This finding leads to further work on mutagenesis and helps to develop and release different mutant of several crops.

Chemical mutagens have been successfully employed in mutation breeding programs to artificially generate variations for the development of new varieties with improved traits, such as an increased yield, reduced plant height, resistance to disease and other desirable agronomic characters (Khursheed, *et al*., 2015, Tantray *et al*., 2017). Chemical mutagens primarily induce single point mutation so, it has been most commonly used in reverse genetic studies, and current technologies can also be easily adapted for their discovery (Jankowicz-Cieslak & Till, 2016).

Gathering of information on newly released mutant varieties is further complicated by the fact that mutant varieties have been released in approximately sixty countries (Table 1). Additionally, in most of these countries induced mutations are used for improvement of various crops, often in different plant breeding stations.

Table 1: Number of officially released mutant varieties listed by country

Country	Common name and number of released varieties	Total
Algeria	soybean (1)	**1**
Argentina	groundnut (2), lemon (1), orange (1), peach (1), wheat (1)	**6**
Australia	blue lupin (1), lupin (1), oat (2), serradella (1), soybean (1), subterranean clover (1)	**7**
Austria	apple (1), barley (9), durum (6), faba bean (1)	**17**
Bangladesh	black gram (1), chickpea (1), jute (1), mungbean (4), oriental mustard (3), rapeseed (2), rice (5), tomato (3), tossa jute (3)	**23**
Belgium	azalea (8), barley (1), chrysanthemum (7), ficus (2), guzmania (1), potato (1), red clover (1), ryegrass (1)	**22**
Brazil	chrysanthemum (3), common bean (3), rice (1), wheat (2)	**9**
Bulgaria	barley (4), durum (4), pepper (3), lentil (1), maize (8), peach (1), pepper (1), soybean (3), sweet pepper (2), tobacco (1), wheat (2)	**30**
Burkina Faso	rice (2)	**2**
Canada	apple (2), apricot (1), barley (5), begonia (2), common bean (12), flax/linseed (3), rapeseed (1), rose (2), Russian wildrye (1), sweet cherry (5), tobacco (1)	**35**
Chile	barley (1), wheat (1)	**2**

China	alfalfa (1), apple (1), barley (7), bougainvillea (2), canna lilies (4), chinese cabbage (4), chinese garlic (1), chrysanthemum (21), common bean (1), cotton (8), crown vetch (1), cucumber (1), dahlia (2), flax/linseed (3), foxtail millet (1), groundnut (29), jute (1), lotus (3), maize (42), millet (20), mulberry (6), orange/mandarin (5), pea (1), pear (5), radish (1), rapeseed (7), rice (191), rose (35), sesame (1), shadawang (5), sorghum (3), soybean (54), sugar beet (2), sugarcane (2), sunflower (1), sweet potato (4), taro (1), tea (1), watermelon (2), wheat (124), white ramie (1)	**605**
Costa Rica	common bean (1), cowpea (1), rice (2),	**4**
Cote d'Ivoire	rice (25)	**25**
CSFR/Czech Rep.	barley (27), common bean (1), crimson clover (1), maize (3), rose (1), soybean (1), vetch (1), mustard (1)	**36**
Denmark	barley (21)	**21**
Egypt	chickpea (1), common bean (1), sesame (2)	**4**
Estonia	barley (4), potato (1)	**5**
Finland	barley (4), oat (4), rye (2), wheat (1)	**11**
France	apple (5), barley (12), black currant (1), carnation (4), dahlia (5), durum (1), forsythia (2), plum (1), rice (5), weigela (3)	**39**
Germany/ FRG/GDR	alstroemeria (11), azalea (3), barley (44), carnation (4), chrysanthemum (34), common bean (2), faba bean (1), geranium (1), meadow fescue (3), meadow foxtail (2), ribes (1), rose (3), rye (2), snapdragon (1), soybean (1) spinach (1), streptocarpus (22), wheat (2),	**138**
Ghana	cassava (1)	**1**
Greece	barley (1), durum (1)	**2**
Guyana	rice (26)	**26**
Hungary	chrysanthemum (1), maize (1), rice (3), soybean (1), wheat (1),	**7**
India	barley (14), bitter gourd (1), black gram (3), bougainvillea (10), castor bean (3), chickpea (4), chinese mustard (1), chrysanthemum (46), citronella (6), common bean (1), cotton (9), cowpea (6), dahlia (11), eggplant (1), egyptian clover (1), gladiolus (2), green pepper (1), groundnut (13), hibiscus (2), hyacinth bean (1), khasianum (1), lentil (1), mulberry (1), mungbean (5), mustard (1), okra (1), opium poppy (1), oriental mustard (3), papaya (1), pea (1), pearl millet (5), pigeon pea (5), polyanthes (2), portulaca (10), portulaca per. (1), rice (40), ridged gourd (1), rose (15), sesame (3), sorghum (1), sugarcane (5), tobacco (1), tomato (4), tossa jute (3), turmeric (2), wheat (4), white jute (2), wild sage (3)	**259**
Indonesia	mungbean (1), rice (6), soybean (3), tobacco (1)	**11**
Iraq	barley (7), faba bean (2), rice (3), sesame (3), tobacco (2), wheat (6)	**23**
Italy	almond (1), common bean (2), durum (13), eggplant (3), green pepper (1), olive (1), pea (6), potato (1), rice (1), sweet cherry (3), vetch (1), wheat (2)	**35**

Japan	abelia (1), apple (1), azalea (1), azuki bean (1), barley (8), begonia (6), burdock (4), carnation (1), chinese matgrass (1), chrysanthemum (14), creeping bent grass (1), eustoma (3), hibiscus (1), japanese pear (2), job's tears (1), lettuce (2), loquat (1), mat rush (2), mint (1), potato (1), rice (46), rose (3), roselle (4), soybean (6), sugarcane (1), tomato (4), turnip/jpn rape (1), wheat (2)	**120**
Kenya	cowpea (2)	**2**
Korea	barley (1), rice (2), sesame (6), soybean (2),	**11**
Korea, rep.of	rice (5)	**5**
Malaysia	banana (1)	**1**
Mali	sorghum (8)	**8**
Mongolia	wheat (3)	**3**
Myanmar	groundnut (1), rice (2), tossa jute (1)	**4**
Netherlands	achimenes (8), african violet (1), alstroemeria (24), apple (flowers) (1), azalea (3), barley (1), begonia (6), calathea (1), carnation (7), chrysanthemum (80), dahlia (18), euphorbia (1), gladiolus (2), hyacinth (1),kalanchoe (3), lily (2), onion (2), streptocarpus (7), tulip (8)	**176**
Nigeria	rice (3)	**3**
Norway	barley (2)	**2**
Pakistan	chickpea (5), cotton (5), mungbean (9), rapeseed (1), rice (6), wheat (6)	**32**
Peru	barley (1)	**1**
Philippines	rice (4)	**4**
Poland	barley (1), blue lupin (1), chrysanthemum (6), faba bean (5), gerbera (1), pea (14), scarlet runner (1), yellow lupin (1),	**30**
Portugal	rice (1)	**1**
Romania	rice (1)	**1**
Russia	barley (2), millet (1), onion (1), pea (1), tulip (1)	**6**
Senegal	rice (2)	**2**
Sri Lanka	groundnut (1), rice (1), sesame (1)	**3**
Sweden	barley (20), mustard (3), pea (1), rapeseed (2)	**26**
Switzerland	wheat (1)	**1**
Thailand	banana (1), carnation (1), chrysanthemum (2), rice (4), soybean (1)	**9**
Turkey	barley (1), soybean (2)	**3**
UK	barley (31), streptocarpus (1)	**32**
Ukraine	barley (1)	**1**
USA	barley (13), begonia (11), bermuda grass (4), carnation (1), centipede grass (2), chrysanthemum (1), common bean (26), crape myrtle (2), crested wheatgrass (1), grapefruit (2), groundnut (1), hop (3), hoya (4), lespedeza (2), lettuce (3), lilac (1), oat (12), peppermint (2), rice (23), rose (2), snapdragon (3), st. Augustine grass (2), tobacco (1), wheat (3)	**125**

USSR	amarant (1), barley (26), brome grass (1), buckthorn (1), buckwheat (8), castor bean (1), chamomile (1), chrysanthemum (17), common bean (4), cotton (2), cress (1), cucumber (1), durra (1), faba bean (4), fig (1), flax/linseed (3), fodder beet (5), grape (1), iris (5), kale (1), lettuce (1), maize (12), millet (3), oat (3), onion (1), pea (8), pepper (1), plavine (1), pomegranate (2), poplar (1), rapeseed (2), raspberry (1), rice (6), sainfoin (2), sorghum (1), sour cherry (4), soybean (9), sudan grass (1), sunflower (1), tobacco (4), tomato (2), vetch (1), watermelon (1), wheat (36), white lupin (13), yellow lupin (2)	**204**
Vietnam	groundnut (1), indian jujube (2), maize (2), peppermint (1), rice (18), soybean (5),	**29**
Yugoslavia	pepper (1)	1

Among these countries, while in six countries, the number of released mutant varieties exceeded 100. The topcountries on the list are China, India, former USSR and Russia, The Netherlands, USA and Japan (Table 2). However, the list would change if the mutant varieties developed in the former FRG and GDR (in total 138 varieties including one variety recently released in Germany were combined).

Table 2: Number of officially released mutant varieties in the top six countries (total 2,252)

Country	Number of released mutantcultivars	Percent of total
China P.R.	605	**26.8**
India	259	**11.5**
USSR + Russia	210	**9.3**
Netherlands	176	**7.8**
USA	128	**5.7**
Japan	**120**	**5.3**

The plant species with induced mutant varieties is a long one and recently reached 175 entities (Table 3) as compared with 154 species in 1995 (Maluszynski, *et al*, 1995). This was mainly because of an increase in the application of mutation techniques for the improvement of ornamental and decorative plants in developing countries, where these plants have become important "cash crops". It is remarkable that the number of mutant varieties of vegetative propagated crops has only slightly increased in spite of the availability of many *in vitro* culture methods, which should have facilitated the development of new varieties.

A new FAO/IAEA Coordinated Research Project has been established this year to identify constraints in the production of mutant varieties of fruit trees and to develop methods and protocols for more efficient use of mutation techniques and related biotechnologies. The most significant increase, compared to 1995, (Maluszynski, M., L. van Zanten, A. Ashri *et al*, 1995) was observed in the number of new mutant varieties in crop species (494 new), mainly in seed propagated crops (366 new mutantvarieties). The distribution pattern among seed propagated crops did not change very much. Mutant varieties of cereals are on the top of the list (1072) followed by legumes (311), industrial (81), vegetables (66), oil crops (59) and other seed

propagated crops (111). Significant increase was observed in the number of newly released rice and wheat mutant varieties. Progress in the use of induced mutations for oilseed crops improvement was recently reviewed by Bhatia *et al***.** 1999.

Table 3: Number of officially released mutant varieties in different species

Latin name	Common name	No. of mutant varieties
Abelia sp.	abelia	1
Abelmoschus esculentus (L.) Moench	okra	1
Achimenes sp.	achimenes	8
Agropyron cristatum (L.) Gaertner	crested wheat grass	1
Agrostis sp.	creeping bent grass	1
Allium cepa L.	onion	4
Allium macrostemon Bunge	chinese garlic	1
Alopecurus pratensis L.	meadow foxtail	2
Alstroemeria sp.	alstroemeria	35
Amaranthus sp.	amaranth	1
Antirrhinum sp.	snapdragon	4
Arachis hypogaea L.	groundnut	48
Arctium lappa L.	burdock	4
Astragalus huangheensis	shadawang	5
Avena sativa L.	oat	21
Begonia sp.	begonia	25
Beta vulgaris L.	fodder beet	5
Beta vulgaris L.	sugar beet	2
Boehmeria nivea (L.) Gaudich.	white ramie	1
Bougainvillea sp.	bougainvillea	12
Brassica campestris L.	turnip/jpn rape	1
Brassica juncea L.	oriental mustard	6
Brassica napus L.	rapeseed	15
Brassica oleracea (L.) var. acephala	kale	1
Brassica pekinensis Rupr.	chinese cabbage	4
Bromus inermis Leyss.	brome grass	1
Cajanus cajan Millsp.	pigeon pea	5
Calathea crocata	calathea	1
Camelia sinensis Kuntze	tea	1
Canna indica L.	canna lilies	4
Capsicum annuum L.	pepper	10
Carica papaya L.	papaya	1
Chrysanthemum sp.	chrysanthemum	232
Cicer arietinum L.	chickpea	11
Citrullus lanatus Mansf.	watermelon	3
Citrus limon (L.) Burm.	lemon	1

Citrus paradisi Macf.	grapefruit	2
Citrus sinensis (L.) Osbeck	orange	1
Citrus sp.	orange/mandarin	5
Coix lachryma-jobi L.	job's tears	1
Colocasia esculenta Schott.	taro	1
Corchorus capsularis L.	jute	2
Corchorus capsularis L.	white jute	2
Corchorus olitorius L.	tossa jute	7
Coronilla varia L.	crown vetch	1
Cucumis sativus L.	cucumber	2
Curcuma domestica Val.	turmeric	2
Cymbopogon uinterianus Jouitt	citronella	6
Cynodon sp.	bermuda grass	4
Cyperus malaccensis Lam.	chinese matgrass	1
Dahlia sp.	dahlia	36
Dianthus caryophyllus L.	carnation	18
Dolichos lablab L.	hyacinth bean	1
Xremochloa ophuiroides Hack	centipedegrass	2
Xriobotrya japonica Lindl	loquat	1
Xuphorbia fulgens Karw.	euphorbia	1
Xustoma grandiflorum (Raf.) Shinn.	eustoma	3
Fagopyrum esculentum Gili	buckwheat	8
Festuca pratensis Huds.	meadow fescue	3
Ficus benjamina exotica	ficus	2
Ficus carica L.	fig	1
Forsythia x intermedia	forsythia	2
Gerbera jamesonii Bolus	gerbera	1
Gladiolus sp.	gladiolus	4
Glycine max L.	soybean	90
Gossypium sp.	cotton	24
Guzmania paecockii Ruiz et Pav.	guzmania	1
Helianthus annuus L.	sunflower	2
Hibiscus sp.	roselle	3
Hibiscus sp.	hibiscus	4
Hippophaea rhamnoides L.	buckthorn	1
Hordeum vulgare L.	barley	269
Hoya carnosa R.Br.	hoya	4
Humulus lupulus L.	hop	3
Hyacinthus sp.	hyacinth	1
Ipomoea batatas (L.) Poir.	sweet potato	4
Iris sp.	iris	5
Juncus effusus L.	mat rush	2
Xalanchoe sp.	kalanchoe	3
Lactuca sativa L.	lettuce	6
Lagerstroemia indica L.	crapemyrtle	2

Lantana depressa	wild sage	3
Lathyrus sativus L.	plavine, grass pea	1
Lens culinaris Medik.	lentil	2
Lepidium sativum L.	cress	1
Lespedeza cuneata Dum.	lespedeza	2
Lilium sp.	lily	2
Linum usitatissimum L.	flax/linseed	7
Linum usitatissimum L.	flax	2
Lolium sp.	ryegrass	1
Luffa acutangula Roxb.	ridged gourd	1
Lupinus albus L.	white lupin	13
Lupinus angustifolius L.	blue lupin	2
Lupinus consentini Guss.	lupin	1
Lupinus luteus L.	yellow lupin	3
Lycopersicon esculentum M.	tomato	13
Malus pumila Mill.	apple	9
Malus sp.	apple (flowers)	1
Manihot esculenta (L.) Crantz	cassava	1
Matricaria chamomilla L.	chamomile	1
Medicago sativa L.	alfalfa	1
Mentha arvensis L.	peppermint	1
Mentha arvensis L.	mint	1
Momordica charantia L.	bitter gourd	1
Morus alba L.	mulberry	7
Musa sp.	banana	2
Nelumbo nucifera Gaertner	lotus	3
Nicotiana tabacum L.	tobacco	11
Olea europaea L.	olive	1
Onobrychis viciifolia Scop.	sainfoin	2
Ornithopus compressus L.	serradella	1
Oryza sativa L.	rice	434
Panicum miliaceum L.	millet	4
Papaver somniferum L.	opium poppy	1
Pelargonium grandiflorum hybrid	geranium	1
Pennisetum sp.	pearl millet	5
Phaseolus coccineus L.	scarlet runner bean	1
Phaseolus vulgaris L.	common bean	54
Pisum sativum L.	pea	32
Polyanthes tuberosa L.	polyanthes	2
Populus trichocarpa L.	poplar	1
Portulaca grandiflora L.	portulaca	10
Portulaca grandiflora L.	portulaca per.	1
Prunus armeniaca L.	apricot	1
Prunus avium L.	sweet cherry	8
Prunus cerasus L.	sour cherry	4

Prunus domestica L.	plum	1
Prunus dulcis Webb	almond	1
Prunus persica L.	peach	2
Psathyrostachys juncea (F.) Nevski	Russian wildrye	1
Punica granatum L.	pomegranate	2
Pyrus communis L.	pear	5
Pyrus pyriforia Nakai	japanese pear	2
Raphanus sativus L.	radish	1
Rhododendron simsii Planch.	azalea	2
Rhododendron sp.	azalea	13
Ribes nigrum L.	black currant	1
Ribes sp.	ribes	1
Ricinus communis L.	castor bean	4
Rosa sp.	rose	61
Rubus idaeus L.	raspberry	1
Saccharum officinarum L.	sugarcane	8
Saintpaulia sp.	african violet	1
Secale cereale L.	rye	4
Sesamum indicum L.	sesame	16
Setaria italica (L.) Beauv.	foxtail millet	1
Setaria sp.	millet	24
Sinapis alba L.	white mustard	5
Solanum khasianum Clarke	khasianum	1
Solanum melongena L.	eggplant	4
Solanum tuberosum L.	potato	4
Sorghum bicolor L.	sorghum	13
Sorghum durra Stapf	durra	1
Sorghum sudanense (Piper) Stapf	sudan grass	1
Spinacia oleracea L.	spinach	1
Stenotaphrum secundatum Kuntze	st. Augustine grass	2
Streptocarpus sp.	streptocarpus	30
Syringa vulgaris L.	lilac	1
Trifolium alexandrinum L.	egyptian clover	1
Trifolium incarnatum L.	crimson clover	1
Trifolium pratense L.	red clover	1
Trifolium subterraneum L.	subterranean clover	1
Triticum aestivum L.	wheat	197
Triticum turgidum ssp. durum Desf.	durum	25
Tulipa sp.	tulip	9
Vicia faba L.	faba bean	13
Vicia sativa L.	common vetch	3
Vigna angularis Willd.	azuki bean	1
Vigna mungo L.	black gram	4
Vigna radiata (L.) Wil.	mungbean	19
Vigna unguiculata Walp.	cowpea	9

Vitis vinifera L.	grape	1
Weigela sp.	weigela	3
Zea mays L.	maize	68
Ziziphus mauritiana Lam.	indian jujube	2

Among the total 2,252 mutant varieties, there were 1,585 verities developed Mdirectly' after mutagenic treatment and selection in the subsequent generations. However, in many cases mutants or already released mutant varieties have been used as sources of desired characters in cross breeding programmes; in this way, 667 new varieties were developed. Of 1,585 directly developed mutant varieties, a great majority (1,411) were obtained with the use of radiation as the mutagen (Table 4).

Table 4: Number of officially released mutant cultivars developed with different types of radiation

Type of mutagen	Number of released mutant cultivars	Percent of total
Radiation*	1411	**100.00**
gamma rays*	910	**64.49**
x-rays*	311	**22.04**
gamma chronic	61	**4.32**
fast neutrons**	48	**3.40**
thermal neutrons	22	**1.56**
other	**24**	**1.70**

**including various treatments*; ***including "neutrons"*
Source: Plant Breeding and Genetics Section Joint FAO/IAEA Division/ Mutant Variety Database, 2020.

Applications of mutation breeding

Mutation breeding has been used for improving both oligogenic as well as polygenic characters. It has been employed to improve morphological and physiological characters, disease resistance and quantitative characters including yielding ability. The various applications of mutation breeding may be briefly summarized as under.

In mutation breeding techniques desirable mutant alleles are induced, those do not present in the germplasm or which may be present, but may not be available to the breeder due to political or geographical reasons. Mutation breeding relieves the complete dependence of breeders on the natural germplasm. But it should be remembered that mutation breeding cannot minimize the necessity of germplasm collections; it only serves as a useful supplement to the available germplasm.

Mutation breeding is useful in improving specific characteristics of a well-adapted high yielding variety. This is particularly so in the case clonal crops due to their highly heterozygous nature; in such acase, mutagenesis is the only method available to improve the specific characteristics of cloneswithout changing their genetic makeup.

Mutagenesis in self-pollinated species, is useful in improving the specific characteristics of otherwise adapted and superior varieties. However, in such species mutagenesis may not be

simpler or quicker than the standard backcross procedure if the characteristic is available in avariety. This is more so because the desirable mutations are often associated with undesirable side effects due to other mutations, chromosomal aberrations, sterility, etc. As a result, one or few backcrosses with the parent variety may be necessary to bring the desirable mutant allele in an acceptable genetic background.

Mutagenesis has been successfully used to improve various quantitative characters, including yield. Several varieties have been developed by this technique. However, there is no critical comparison available to show that the same improvement would not have been broughtabout by the conventional hybridization programmes. F1 hybrids from intervarietal crosses may be treated with mutagens in order to increase genetic variability by inducing mutations and by facilitating recombination among linked genes. But this method has not been widely used.

In developing countries, mutation breeding is widely used, but in Europe it is mainly confined to clonal and ornamental crops. For example, mutagenesis is the principal source of genetic variation in *chrysanthemum* and banana breeding programmes. This is because most breeders believe that the characteristics of mutation breeding, viz.,

a. The need for large (105 to 106) M2 populations,
b. Associated detrimental effects of mutations, and
c. The existence in germplasm of the so called 'novel' mutant alleles,

mitigate against the incorporation of this technique into conventional breeding programmes.

In mutagenesis, the yields of new varieties released over a period of years (developed through conventional breeding approaches) show anaverage increase of -1 % in case the major field crops. Development of a new variety using mutagenesis would require about 7 years; therefore, the mutant variety must show an increase of -7% in yield over the parent variety. An increase of this magnitude is unlikely from modificationof a single gene or trait unless it is critical for plant performance, e.g., disease or insect resistance.

Limitations of mutation breeding

Apart from all this desirable genetic recombination initiated in mutation breeding, there has certain limitations of the technique; these limitations are summarized as under.

1. The frequency of desirable mutations is very low, about 0.1 per cent of the total mutations. Therefore, large M2 and subsequent populations have to be grown and carefully studied. This involves considerable time, labour and other resources.
2. The breeder has to screen large populations to select desirable mutations. Therefore, efficient, quick and inexpensive selection techniques are required to screen large populations.
3. Mutation breeding is more easily applied to such characters where quick screening techniques are available, e.g., disease resistance. But in the case of characters where elaborate tests are required, e.g., quality characteristics, mutation breeding is virtually impractical. For this reason, mutation breeding has been more successful with those characteristics where the mutant phenotype is distinct and easily detectable.
4. Desirable mutations are commonly associated with undesirable side effects due to other mutations, chromosomal aberrations, etc. The mutant lines often have to be back crossed to the respective parent varieties to remove these defects. This increases the time requirement of mutation breeding programmes and involves additional labour, time and expenditure.
5. Often mutations produce pleiotropic effects. The chief procedure for reducing or eliminating pleiotropic effects is to transfer the gene into different genetic backgrounds by hybridizing

the mutant with a randomly selected range of elite varieties. Alternatively, when the pleiotropic effect is on a specific trait, e.g., delayed flowering, appropriate genes for correction of the defect, e.g., genes for early flowering, can be introgressed into the mutant strain.

6. Mutations in quantitative traits are usually in the direction away from the selection history of the parent variety; this conclusion was reached by Brock in 1965 and is generally regarded as valid. This may tend to limit the degree of improvement attainable in a quantitative trait that has been the object of selection for a long period of time, e.g.,yield.
7. Most of the mutations are recessive; detection of recessive mutations is almost impossible in clonal crops and is difficult in polyploidy species. Consequently, in polyploidy species, larger populations have to be grown and larger doses of mutagens have to be applied. Mutagenesis has been most commonly applied to diploid species that reproduce sexually, more particularly to self-pollinated species.

Conclusion

Induced mutagenesis is one of the most powerful breeding tools for creating novel genetic variation and accelerating the process of trait selection. Over the last several decades, mutation breeding is successful in developing large number of mutants with improved agroeconomic traits of diverse plant species.

The availability of accessible genetic variation is highly important to initiate crop improvement programme, the plant breeding supplemented with induced mutagenesis has proved to be coherent and robust.

The technique has been successfully applied in cereals, pulses, medicinal, horticulture plants and fodder crops etc. The different traits of interest like yield, physiological activity, nutritional quality, secondary metabolites, plant biomass etc. has been targeted and improved through mutation breeding. With the advent of modern biotechnological tools and genetic markers, the unbound possibilities of mutation breeding are expanding. In future, it is highly recommended to integrate molecular advancement into the mutation breeding programmes for improving the selection accuracy and target trait specificity.

Variation is among the major factor without which we cannot imagine the improvement of crop in any aspect. Among various method of breeding in crop plant mutation breeding i.e. induced mutation is one of the preeminent methods of creation of variation/genetic variation. Conventional method of breeding takes long time to improve a crop variety due to a very slow increase in genetic variation.

To overcome this induced mutation, play a crucial role which helps in creation of genetic variation in a short period. Over last several year's mutation breeding is getting popular and is adopted by several countries. It improves several qualitative and quantitative characters of crop plant and is successfully applied in several cereal, grain legume, oil seed, vegetable, fruits, medicinal plant, ornamental plants and fodder crops. With the advancement of various plant breeding, genetics, and biotechnological tools mutation breeding contribute toward the increase in global food and agriculture production which ultimately overcome global hunger and improve the nutritional status of the globe.

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Phytochemical screening of the leaves of *Achyranthes aspera Linn , Phyllanthus niruri Linn , Leea indica Burm.f* Tal-Dist-Palghar , Maharashtra, India

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Keywords:
Achyranthes aspera Linn, Phyllanthus niruri Linn, Leea indica Burm.f, phytochemical screening, Leaves, Medicinal Plants

Abstract:
Medicinal plants are the important source of potential drugs in our country . The plants *Achyranthes aspera Linn, Phyllanthus niruri Linn, Leea indica Burm.f* is widely used in Ayurvedic system of medicine as antidiabetic , antiasthamatic, antiviral,antibacterial, antitumor, analgesic, antifungal, anticarcinogenic , etc . The present study deals with phytochemical screening of leaves of the selected plants for its identification and to distinguish it from the co-existing weeds . The phytochemical screening of leaves is done by using solvent like chloroform . The qualitative analysis revealed the presence of alkaloids, phenol, flavonoids, glycoside, tannin, reducing sugar in the leaves. Since there is no proper information regarding this plants , our efforts were devoted to study the phytochemical analysis of this plants . Thus , present study shows that this plants having various medicinal properties and can be use for the treatment of various disease . The study is done to reveal the good impact of ethnomedicinal plants on the health . In future allopathic medicines can replace by homeopathic medicines . So this study was undertaken for proper identification , collection and investigation of the plants .

INTRODUCTION :

Plants were identified with the help of Cookes flora. *Achyranthes aspera* belongs from family Amaranthaceae includes 2500 species and 174 genera distributed worldwide. *Phyllanthus niruri* belongs from family Phyllanthaceae includes 2000 species and 54 genera. *Leea indica* belongs from family Vitaceae includes 910 species and 14 genera. All selected plants are well known plants drugs in Ayurvedic, Unani-Tibbi, Siddha, Allopathic, Homeopathic, Naturapathic and Home remedies (Khare C.P. , 2007). Ethnomedicines are derived from plants and used by various tribal people of the various regions. Plants have been used for health and medicinal purposes since ancient times . Medicinal plants are the richest source of secondary metabolites which can act as bioactive components (AbhayKumar Kamble, 2018) . This plants contains numerous biologically active compounds which are helpful in the treatment of various diseases and improving the life (Samy, et, al., 2008) . This plants posses properties like anti-inflammatory, antifungal, antibacterial,antiperiodic, antidiabetic, antiasthamatic, antiviral, anticarcinogenic, etc. (Wakhloo,

et. Al., 1979) . 80% of the number of inhabitants in creating nations rely upon customary drugs, by using plant parts by WHO (Vines 2004) . Etnomedicines is concerned with cultural interpretations of health, disease and illness and also addresses the health care seeking process and healing practices. Research interest and activities in the areas of ethnaobiology and ethnomedicine haave increased tremendously in the last decades. Apart from that, these plants plays a critical role in the development of human cultures around the whole world (Akash, Navneet, B.S.Bhandari, 2020). For developing phytomedicines as a major area of concern, it would be essential to adopt a holistic interdisciplinary approach, have a scientific basis of the understanding of the plant systems, new innovations and their conservation for utilisation in future on a sustainable basis (Sharma, 1997).

MATERIALS AND METHODS :

Collection of plant sample:

The leaves was collected from Tal-Dist-Palghar, Maharashtra,India

Preparation of the extract

The leaves of the selected plants were washed thoroughly in tap water to remove dust particles. The leaves were dried in shade at room temperature for 1 month and coarsely powdered by mechanical grinder. The dried powdered sample was soaked in solvent like chloroform for 5

days. After 5 days, the extracts were filtered through no.1 Whatman filter paper and stored in air tight container for further screening.

Qualitative analysis of phytochemicals

Preliminary phytochemical screening was carried out (Harborne, 1980)

1. Test for alkaloids (Mayer's test)

To 1ml of extract, 1 ml of Mayer's reagent (Potassium iodide solution) was added.

Formation of whitish yellow or cream coloured precipitate indicates the presence of alkaloids.

2. Test for steroids (Libermann Burchard test)

To 1ml of extract, 2ml of acetic anhydride and 2ml of concentrated sulphuric acid were added.

Formation of violet to blue or green colour indicates the presence of steroids.

3. Test for terpenoids (Salkowski test)

To 1 ml of extract, 2ml of chloroform and few drops of sulphuric acid were added.

Formation of reddish brown ring indicates the presence of terpenoids.

4. Test for flavonoids (Alkaline reagent test)

To 1 ml of extract, few drops of dilute ammonium solution and few drops of concentrated hydrochloric acid were added.

A yellow colouration indicates the presence of flavonoids.

5. Test for saponins (Froth test)

To 1 ml of extract, 5 ml of distilled water was added and shaked vigorously.

Formation of froth indicates the presence of saponins.

6. Test for phenols (Lead Acetate test)

To 1ml of extract, 1 ml of lead acetate solution was added.

Formation of precipitate indicates the presence of phenols.

7. Test for tannins (Lead acetate test)

To 1ml of extract, 1ml of lead acetate was added.

A formation of white precipitate indicates the presence of tannins.

8. Test for cardiac glycosides (Keller killiani test)

To 1ml of extract, 5ml of distilled water was added and evaporated to dryness. Then to the Sample 2ml of glacial acetic acid containing trace amount of ferric chloride solution was added. Then 1ml of concentrated sulphuric acid was added along the sides of the tube.

Formation of brown ring underlayed with blue colour indicates presence of cardiac glycosides.

9. Test for amino acids (Ninhydrin test)

To the 1ml of sample, 3 to 4 drops of Ninhydrin solution was added and boiled in water bath for 10 minutes.

Formation of purple or blue colour indicates the presence of amino acids.

10. Test for proteins (Biuret test)

To the 1ml of extract, 1ml of 40% sodium hydroxide solution and 2 drops of 1% copper sulphate solution were added.

Formation of violet colour indicates the presence of proteins.

11. Test for carbohydrates (Barfoed test)

To the 2ml of extract, 1ml of Barfoed's reagent was added and boiled in water bath for few minutes.

Formation of reddish brown precipitate indicates the presence of carbohydrates.

12. Test for reducing sugars (Fehling's test)

To the 1ml of extract, equal quantities of Fehling solution A and B were added and heated.

Formation of brick red precipitate indicates the presence of reducing sugars.

OBSERVATION TABLE :

Qualitative phytochemical analysis of the leaves of *Achyranthes aspera Linn , Phyllanthus niruri Linn, Leea indica Burm.f*

TESTS	*Achyranthes aspera*	*Phyllanthus niruri*	*Leea indica*
ALKALOIDS	+	+	+
STEROIDS	-	-	-
FLAVONOIDS	-	+	+
TERPENOIDS	-	+	-
SAPONINS		+	-
PHENOLS	+	+	-
TANNINS	+	+	+
CARDIAC GLYCOSIDES	+	-	-
AMINO ACIDS	-	-	-
PROTEINS	-	-	-
CARBOHYDRATES	-	-	-
REDUCING SUGARS	+	-	+

RESULTS :

The qualitative analysis were done to find out which phytochemicals found in the selected plants. The selected plants shows the traces of the secondary metabolites in their leaves. The plant *Achyranthes aspera* shows the presence alkaloids, phenols, tannins, cardiac glycosides and reducing sugar. The plant *Phyllanthus niruri* shows the presence of alkaloids, flavonoids, terpenoids, saponins, tannins. The plant *Leea indica* shows the presence of alkaloids, flavonoids, tannins, reducing sugar.

CONCLUSION :

The three selected plants possess the different kinds of the phytochemicals in their leaves. Medicinal plants are the richest source of secondary metabolites which can act as bioactive components. And this phytochemicals have medicinal properties which can be used for treating and preventing various disease.This plants acts as antidiabetic,antiasthamatic, antiviral,antibacterial,antitumor, analgesic, antifungal, anticarcinogenic agents. In future allopathic medicines get replace by homeopathic medicines. It will reduce the dependence on the pharmaceutical drugs. Phytochemical analysis will surely reveal the beneficial properties . This study will definitely utilize the plants which considered to be weeds. It will be helpful in searching for bioactive agents those can be used in the synthesis of useful drugs.

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Acetobacter diazotrophicus as Nitrogen fixing Endophytic bacteria in Sugarcane Collected from Rahuri, Ahemednagar

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Keywords:
Acetobacter diazotrophicus, Endophytic bacteria, Rahuri, Ahmedangar.

Abstract:
Rahuri taluka of Ahmednagar district is located between latitude 19.3951 and longitude 74.6521 meter. The climate of Rahuri is characterized by hot summer and general dryness. Nitrogen fixation is an important process in plants providing nitrogen as the most valuable macronutrient required by the plants. *Acetobacter diazotrophicus* as an endophytic nitrogen fixing bacteria is collected from Rahuri taluka of Ahmednagar district (M.S). Total 16 rhizospheric samples were collected from various localities of the taluka and soil testing was done to determine pH and Water Holding Capacity (WHC). An average pH of soil samples collected from the study area was ranging between 5.7 to 6.5 pH; while WHC was ranging between 33.20 to 41.80 %. After isolation of bacteria from soil sample, cell morphology and colony morphology was studied. With the help of special media and morphological characters preliminary identification of *A. diazotrophicus* was done. Out of these 16 bacterial strains 05 bacterial strains did not show characters identical with *A. diazotrophicus;* while other strains resembled *A. diazotrophicus.* Morphologically 11 bacterial strains *viz.* RH 01, RH 02, RH 03, RH 04, RH 06, RH 08, RH 09, 09, RH 11, RH 13, RH 14 and RH 15 were identical showing similar morphological characters. While 05 bacterial strains RH 05, RH 07, RH 10, RH 12 and RH 16 were different from one another and they are grouped in Group-II. Colonies were creamy white, cicular, small to large sized, opaque with smooth margins. *A. diazotrophicus* is able to increase nutrient supply, soil fertility and crop growth of sugarcane. The study of *A. diazotrophicus* will be useful for further researchers and it will be better alternative for chemical fertilizers.

INTRODUCTION

Rahuri taluka of Ahmednagar district is located between latitude 19.3951 and longitude 74.6521 meter. The climate of Rahuri is characterized by hot summer and general dryness. The average temperature in Rahuri taluka is 25.9°C and the average rainfall is 511 mm. The area under Rahuri is 1061.5 square kilometre (Mahadule *et al.,* 2020). Gulve and Gadekar (2022) studied watershed development program in Ahmednagar district. Nitrogen fixation is an important process in plants providing Nitrogen as the most valuable macronutrient required by the

plant. Crop rotation with legumes has been recognized to increase soil fertility and agricultural productivity (Cheng, 2008). Santi1 *et al.,* (2013) studied biological nitrogen fixation in non-legume plants. Endophytic bacteria can influence plant growth & productivity through Nitrogen fixation. Conceptually, plant growth promoting endophytic bacteria may affect plant growth either directly or indirectly (Sansanwal *et al.,* 2017). Large and diverse populations of N2-fixing bacteria are associated with sugarcane.

Endophytic bacteria establish in between and within the spaces of all plant parts and not causing any plant disease. They create array of relationship include mutualism, cannibalistic, commensalistic andtrophobiotic in nature. Endophytic bacteria play a major role in developing plant growth enhancement, phytoremediation, phosphate solubilization, nitrogen fixation, modulation of plant metabolism and phytohormone signaling. There is an increasedinterest in the use of endophytes for their agricultural applications that promote plant growth under cold, drought or contaminated soil structure conditions or induce disease resistance in plants (Muthukumar *et al.,* 2018). Endophytic bacteria are alternative to agrochemicals (fertilizers and pesticides) in developing environment friendly agriculture (Adeleke and Babolola, 2021). Therefore, e ndophytic bacteria play an important role in microbial ecology, associating environmental factors, and their roles that contribute to their effectiveness in promoting plant growth for maximum agricultural cropproductivity was highlighted (Adeleke and Babolola 2021). *A. diazotrophicus* is a nitrogen-fixing endophytic bacterium, originally isolated from sugarcane. Its colonizing ability was evaluated in field of agriculture to promote the growth and development of crop plant. A preliminary study regarding contributions of the bacterial endophyte *A. diazotrophicus* to sugarcane nutrition was reported by Sevilla *et al.,* (1998). *A. diazotrophicus* was found mainly inside cortical cells of stems and inside xylem vessels. No L-glucuronidase activity was observed in non-inoculated plants. *A. diazotrophicus* is able to increase nutrient supply, soil fertility and crop growth of sugarcane. The study of *A. diazotrophicus* will be useful for further researchers and it will be better alternative for chemical fertilizers. *A. diazotrophicus* colonizing the root intercellular spaces and the interior of root epidermal cells. They proposed that *A. diazotrophicus* could be distributed from the base of the stem to other organs via stem xylem vessels, since they also detected xylem colonization in the basal region of the stalk in non-inoculated sugarcane plants (Dong *et al.,* 1997). Hence during the present investigation report of *A. diazotrophicus* was collected from sugarcane from Rahuri taluka of Ahemednagar district

MATERIALS & METHODS

a) Collection of bacterial samples: Rhizosphere samples were collected from 16 different locations of Rahuri taluka of Ahmednagar district in sterile zipped locked polythene bags. Those samples were brought to the laboratory and kept at 4^{O}C for further investigations. Soil pH was calculated using pH meter, while Water Holding Capacity (WHC) was determined as described (Kalra, 1995).

b) Isolation of bacterial samples: One gram of soil was suspended in 10 ml distilled water to prepare soil suspension. It was inoculated on specific *Acetobater* manitol agar media (Hi-Media) and incubated at 25±2^{O}C for 48 Hrs. which allow only the growth of *A. diazotrophicus*.

c) Morphological Characterization: Confirmation of the bacteria was done by relevant morphological characterization (Phalke *et al.,* 2017). Growth of colonies was observed after 48 Hrs. Morphology characterization of bacterial cell was studied in respect to cell size, shape and gram staining. While Colony morphology was studied in respect to color, shape, size, appearance

and colony margins on the special culture media as described by Phalke *et al.,* (2017). Cultures were preserved at 20^{O}C for further studies.

RESULTS & DISCUSSION

Soil samples were collected from 16 different localities of the study area and the value of soil pH and WHC are presented in Table 01. An average pH of soil samples collected from the study area was ranging between 5.7 to 6.5 pH. This indicates acidic nature of the soil in the study area. WHC was observed between 33.20 to 41.80 %; Maximum soil pH was recorded at Chandegaon (6.5); while minimum at Tambhere (5.7). Maximum WHC was recorded in the sample collected from Chincholi (41.80%); while minimum at Tambhere (33.20%) Overall average pH of all samples is 6.1 out of 16 localities, 10 localities *viz.* Ambi (6.3), Aradgaon (6.2), Chandegaon (6.5), Chincholi (6.3), Davangaon (6.4), Kendal Budruk (6.2), Manori (6.2), Nimbhere (6.1), Songaon (6.1) and Musalwadi (6.3) showed high pH than that of the average pH of all samples and 6 localities showed less pH than the average pH *viz* Bodhegaon (5.8), Bramhani (5.9), Digras (5.7), Satral (5.8), Tambhere (5.7) and Taharabad (5.9). The average WHC of all samples is 37.07; out of which 7 soil samples showed high WHC than the average and 9 samples showed less WHC than the average.

Table 01: Localities selected for collection of Sugarcane Rhizosphere sample.

Sample Code No.	Location	Soil Type	pH	Water holding capacity (WHC) in Percentage
RH01	Ambi	Black	6.3	36.80
RH02	Aradgaon	Laterite soil	6.2	37.40
RH03	Bodhegaon	Black	5.8	35.60
RH04	Bramhani	Black	5.9	36.48
RH05	Chandegaon	Lommy soil	6.5	39.20
RH06	Chincholi	Red and black soil	6.3	41.80
RH07	Davangaon	Black	6.4	40.60
RH08	Digras	Laterite soil	5.7	37.40
RH09	Kendal Bk.	Black	6.2	33.80
RH10	Manori	Black	6.2	35.90
RH11	Nimbhere	Black	6.1	39.30
RH12	Satral	Black	5.8	33.70
RH13	Songaon	Black	6.1	35.70
RH14	Tambhere	Laterite soil	5.7	33.20
RH15	Musalwadi	Black	6.3	41.60
RH16	Taharabad	Black	5.9	34.72
Average			**6.1**	**37.07**

Table 02: Morphological characters of Nitrogen fixing Endophytic bacterial strains collected from Rhizosphere

Strain	Cell Morphology			Colony Morphology				
	Gram Staining	Cell Size (Avg.)	Cell Shape	Color	Shape	Size (Avg.)	Appearance	Margins
RH01	-ve	1.55 µm	Rod	Creamy White	Circular	1.25 mm	Glistering	Entire
RH02	-ve	1.60 µm	Rod	Creamy White	Circular	1.32 mm	Glistering	Entire
RH03	-ve	2.40 µm	Large rod	Yellow	Irregular	1.34 mm	Opaque	Rough
RH04	-ve	1.65 µm	Rod	Creamy White	Circular	1.27 mm	Glistering	Entire
RH05	+ve	2.56 µm	Large rod	Creamy pale orange	Irregular	1.44 mm	Glistering	Rough
RH06	-ve	1.45 µm	Rod	Creamy White	Circular	1.39 mm	Glistering	Entire
RH07	+ve	2.48 µm	Large rod	Creamy pale orange	Irregular	1.54 mm	Opaque	Rough
RH08	-ve	1.88 µm	Rod	Creamy White	Circular	1.31 mm	Glistering	Entire
RH09	-ve	1.60 µm	Rod	Creamy White	Circular	1.28 mm	Glistering	Entire
RH10	+ve	2.68 µm	Rod	Yellow	Irregular	1.49 mm	Glistering	Rough
RH11	-ve	1.70 µm	Coccus	Creamy White	Circular	1.37 mm	Opaque	Entire
RH12	+ve	2.54 µm	Large rod	Yellow	Irregular	1.45 mm	Opaque	Rough
RH13	-ve	1.72 µm	Rod	Creamy White	Circular	1.29mm	Glistering	Entire
RH14	-ve	1.72 µm	Rod	Creamy White	Circular	1.31mm	Glistering	Entire
RH15	-ve	1.72 µm	Coccus	Creamy White	Circular	1.35mm	Glistering	Entire
RH16	+ve	2.62 µm	Large rod	Creamy pale orange	Irregular	1.58mm	Opaque	Rough

Morphological details of the bacterial samples are presented in Table 02. Morphologically 11 bacterial strains *viz.* RH 01, RH 02, RH 03, RH 04, RH 06, RH 08, RH 09, 09, RH 11, RH 13, RH 14 and RH 15 were identical showing similar morphological characters. These strains are grouped as Group-I. While 05 bacterial strains RH 05, RH 07, RH 10, RH 12 and RH 16 were different from one another and they are grouped in Group-II. Bacterial cells of Group-I were gram negative. While bacterial cell of Group-II were Gram positive in staining. The cell size of Group-I varies from 1.45 µm to 1.88 µm. Group-II cell size was larger than that of Group-I which was varying between 2.40 µm to 2.68 µm. All the bacterial strains of Group-I was rod shaped but in Group-II, RH 05, RH 07, RH 10, RH 12 was large rod shaped while bacterial cell of RH 16 was coccus in shape.

All the bacterial colonies of Group-I strains were creamy white colored on the special media. And in Group-II, Strain RH 05, RH 07, RH 16 showed creamy pale orange color while RH 10 and RH 12 showed yellow color while. The bacterial colonies of Group-I strains were circular in shape while Group-II showed irregular shape. Colony size of the Group-I was ranging between 1.25 mm to 1.39 mm while colony size of Group-II was ranging between 1.44 mm to 1.58 mm. Appearance of the Group-I bacterial strain is glistering while Group-II showed opaque colonies. The bacterial strain margins of Group-I showed smooth margins while Group-II showed rough and smooth margin. These morphological characters resembled *A. diazotrophicus*.

Similar bacterial cell and colony morphology of *A. diazotrophicus* was described by various research workers (Fuentes-Ramirez *et al.,* 1998; Madhaiyan *et al.,* 2004; Chawla *et al.,* 2014). Fuentes-Ramirez *et al.,* (1998) reported colonization of sugarcane by *A. diazotrophicus* inhibited by high N-fertilization. Reis *et al.,* (1994) suggested improved methodology for isolation of *A. diazotrophicus* and confirmation of its endophytic habitat. Reis *et al.,* in 2015 described role of nitrogen fixing family Acetobacteraceae in agriculture. Similar characterization of *Gluconacetobacter diazotrophicus* is reported by Ahmed *et al.,* (2016) isolated from sugarcane cultivated in Upper Egypt. Kuchekar and Pawar (2019) also studied morphological characterization of *Azotobacter* spp. from various localities of Aurangabad district (MS). James *et al.,* (1994) reported presence of *A. diazotrophicus* as nitrogen-fixing bacterium in sugarcane.

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Arbuscular Mycorrhizal Status of *Heteropogon contortus* L. From Family Poaceae

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Keywords:
Arbuscular, colonization, rhizospheric

Abstract:
The arbuscular mycorrhizal fungi play an important role in absorption of nutrient and enhancing the growth of the plant. It also helpful for stress resistance, drought resistance, etc. the arbuscular mycorrhizal fungi forms symbiotic association with the roots of the host plants and also with the soil. The present investigation was done with the arbuscular mycorrhizal status of one plant species *Heteropogon contortus* wild, belonging to the family Poaceae.The present work on the plant species mainly done to know the root colonization with hyphal, vesicles and arbuscles along with study of diversity of arbuscular mycorrhizal fungal spores from the native rhizospheric soil sample. The roots of the plant along with rhizospheric soil sample were collected from the area surrounding Mauli Mahavidyalaya, Wadala, N. Solapur, Solapur.The analyzed roots showed presence of 90% arbuscular mycorrhizal fungal root colonization and consist of 350/100 gm of AMF spores in rhizospheric soil samples with great diversity in which genus Glomus was much dominating than others.

1. Introduction

The term mycorrhiza was first coined by German Botanist Arber Frank (1985). Mycorrhiza is symbiotic association between fungi and the roots of the higher plants. Such relation are non pathogenic, parasitic with roots of plant (Kar, A.K. 1993). Mycorrhiza plays an important role in increasing mobilizing ability of nutrients such as Phosphorous, Nitrogen, with other organic material for the host plant (Atul – Nayyar *et al*, 2009). Mycorrhiza maintain the intermediate link between plants root and soil (Mulani, R.M. and Wankhede S.B., 2013).Mycorrhiza gives drought resistance ability to herbaceous plants (Bagyaraj D.J. and Varma A., 1995; Sadhana B. 2014). Such great diversity in arbuscular mycorrhizal fungi has been investigated by many workers in medicinal plants, herbaceous plants (Bagyaraj D. J. 2014; Kannan K. and Lakshminarashiman C., 1988; Kumar , A. Chhavi et al 2013; Mulani R.M. *et al* 2004; Mulla R.M. 1994; Mulani R. M. and Waghmare S.S. 2012;).

Heteropogon contortus L. is a perennial grass grows in grass land throughout the world as a dominating grass species. *H. contortus* minor component for a vegetation community (Johnston, 1963). The invasions of these species can alter the structure, function and component of ecosystem throughout the world (D Antonio and Vitousek, 1992). The stem of the plant s flat, rough and pale green in color. The leaves are flat and folded which develops along the length of stem. Flowers are develops in spike; the species mostly forms seeds without pollination.

2 .Materials and Method

2.1 Study site. For present investigation study site selected was area of Mauli Mahavidyalaya, Wadala (17052’25.0’’N and 75^{0}50'04.0''E). N. Solapur, Solapur, Maharashtra.

2.2 Root and rhizospheric soil sample collection

The roots of the plants was collected along with the rhizospheric soil sample of *Heteropogon contortus* L. widely growing in area of Mauli Mahavidyalaya, Wadala , N. Solapur , without any harm to the root system. The collected material brought into laboratory, washed the roots thoroughly with tap water until the removal of adhering soil. The washed roots were cut into 1cm length each. The arbuscular mycorrhizal root colonization was studied by using the method suggested by Phillips and Haymann (1970). The root segments were kept in 10% KOH and autoclaved it for 15 minutes at proper pressure. After that KOH was removed and dipped these root segments into 1N HCL for 5 minutes. After that root segments were stained in cotton blue with lactophenol overnight. In next day such stained root segments were screened under microscope, for its arbuscular mycorrhizal root colonization.

The whole mount of root segments showed presence of arbuscles, vesicles and mycelium as the roots were positively mycorrhizal with colonization. The percentage of root colonization was calculated by using following formula (Giovanneti M and Moss B, 1980).

$$\text{Percentage of root colonization} = \frac{Numberofrootsegmentscolonized}{Totalnumberofrootsegmentsscreened} \text{ X100}$$

Along with calculation of root colonization, the air dried rhizospheric soil sample was preserved for study of spore population. Isolation of spore was done by using method of Gerdmann and Nicolson, (1963). 100 gm of air dried rhizospheric soil dissolved in water with few drops of tween-20. The above solution was continually stirred with glass rod and finally allowed to settle down for about 20 to 25 minutes.

Such settled solution then sieved through different sized sieves as 710mμ, 210mμ, 150mμ, 75mμ, 45mμ, 25mμ respectively. The content of each sieves now poured into separate Petri dishes for examination of spores. The identification of spores were done by using the manual given by Schenck and Perez, (1990).

3 Results and Discussion

The roots of *Heteropogon contortus* L. Showed the presence of all type of arbuscular mycorrhizal root colonization as vesicular, arbuscular and mycelial with about 90%. The present vesicles were with globular, rounded and elongated while hyphae were with branched structure. The hyphae and vesicles were predominately observed.

The analysis of rhizospheric soil sample showed presence of 350 spores/100gm of soil. In rhizospheric soil sample different AMF spores were observed in which *Glomus* was the genera found predominately present as compared to other generas. *Glomus* was with *G. microcarpum, G. aggregatum, and G. mossae.*

The roots of *Vetiveria zizanioides* L.c ollected from the botanical garden of Swami Ramanand Teerth Marathwada University, Nanded showed the presence of 90%arbuscular mycorrhizal fungal root colonization while the soil analysis showed presence of 240 spores in 100gm of soil (R.M. Mulani and S.B. Wankhede, 2015). The grasses show fibrous root system and are efficient in nutrient absorption. However some grasses are heavily mycorrhizal (de la Pena, E. Echeverria *et al, 2006)*.Muthukumar and Udaiya (2000) noticed that 53 grasses were collected from Western Ghats of South India only 18 species showed arbuscular mycorrhizal colonization.Javaid*et al (*1995) also worked on the mycorrhizal root colonization in grass species.

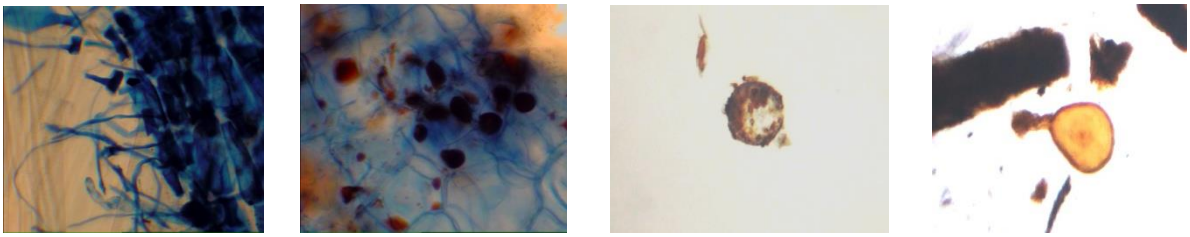

Figure: AM fungal root colonization with AMF spores

4. Conclusion

The present study mainly reveals that the plant *Heteropogon contortus* L. which belongs to family poaceae shows presence of arbuscular mycorrhizal root colonization with arbuscules, hyphal and vesicular infection. The rhizospheric soil collected screened for its spores content revealed that presence of 350 spores per 100gm of soil, in which Glomus species was abundantly found followed by other genus.

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Biochemical Studies Of *Avitellina* Tapeworm Infecting *Capra Hircus(L)* From Parbhani City M.S. India

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Keywords:
Biochemistry, Cestodes, *Capra hircus*, *Avitellina*.

Abstract:
Parasitic biochemistry has great practical importance in understanding of the complex association involved between host parasite relationship .Bio-molecules such as protein, glycogen and lipids are determined in parasite *Avitellina* and infected intestine of host *Capra hircus*. Results, from the present experimental study shows that the percentage of lipid is high in parasites as compared to protein and glycogen from the infected intestine of host. These parasites absorbing most of nourishment from host and fulfilling its need and causing interruption in the proper development of host. Present investigation deals with the biochemistry of parasite *Avitellina* in ruminant *Capra hircus*

Introduction:

India's livestock sector is one of the largest in the world and accounting for 26.40% and goats *Capra hircus* plays an important role in economy (Anonymous, 2012).

Goat rearing is a tribal profession of nomads and many other farming communities in Parbhani city. Goats contribute to the substance of small holders and landless rural poor. Goats due to improper management and unhygienic conditions are suffering from various parasitic infection ranges from acute diseases frequently with high rates of mortality. Cestode parasites when live in the intestine of hosts, they utilize food from the gastrointestinal tract. The metabolism of these cestodes depends on the feeding habits and the rich nourishment available in the gut of the host. These cestode use this nourishment for their normal development and growth.

Biochemistry is the study of structure, composition and chemical reactions of substances in living systems. Parasitology has developed into a multi-dimensional approach in helminth research. They serve as valuable models for the study of fundamental biological phenomena. The biochemistry and physiology of Cestode has been comprehensively reviewed by Smyth and McManus (1989) and specific aspects have been reviewed by Barratt (1981), McManus (1987) and McManus and Bryant (1986).

Glucose is an important source of energy for cestodes, inhabiting the alimentary tract of vertebrates (Mishra et al 1991). Cestodes possess stored carbohydrate metabolism, with enormous amount of stored carbohydrate (Daugherty 1966, Fairbairn, Markov 1939 and Read et Rothman, 1957 b). Cestode parasites stores relatively large quantities of polysaccharides, which in most cases has been assumed to be glycogen (Read 1949b and Reid 1942). Proteins have many different

biological functions. They are ubiquitous in their distribution and there is really no satisfactory scheme of classifying them. The largest groups of proteins are the enzyme proteins provide rich environment for the nourishment of cestodes. The cestodes utilize different degrees of protein for producing energy. Literature reveals that the parasites able to adapt themselves to the parasitic mode of life only due to protein usually constitutes between 20 and 40 % of the dry weight have been reported (John Barrett 1981) . The higher content of lipid is found in older proglottids (Brand and Van T. 1952) . It is revealed from the present study that there is high content of lipids in the parasites and the parasites are taking advantage of host by absorbing most of the nourishing material. The present investigation deals with the biochemical studies of *Avitellina* cestode in ruminants *Capra hircus*.

Material and Methods

The cestodes were collected from the alimentary tract of *Capra hircus* and then washed with distilled water. Collected cestodes were dried on the blotting paper keeping them to remove excess water and transferred to watch glass and weighed on sensitive balance. After 50-60 C^o for 24 hrs, the dry weight was also taken. The estimation of protein content in the cestode parasites were carried out by Lowry's method (1951), the glycogen estimation were carried out by Kemp et al. (1954) method and lipid estimation by Folch et al (1957) method.

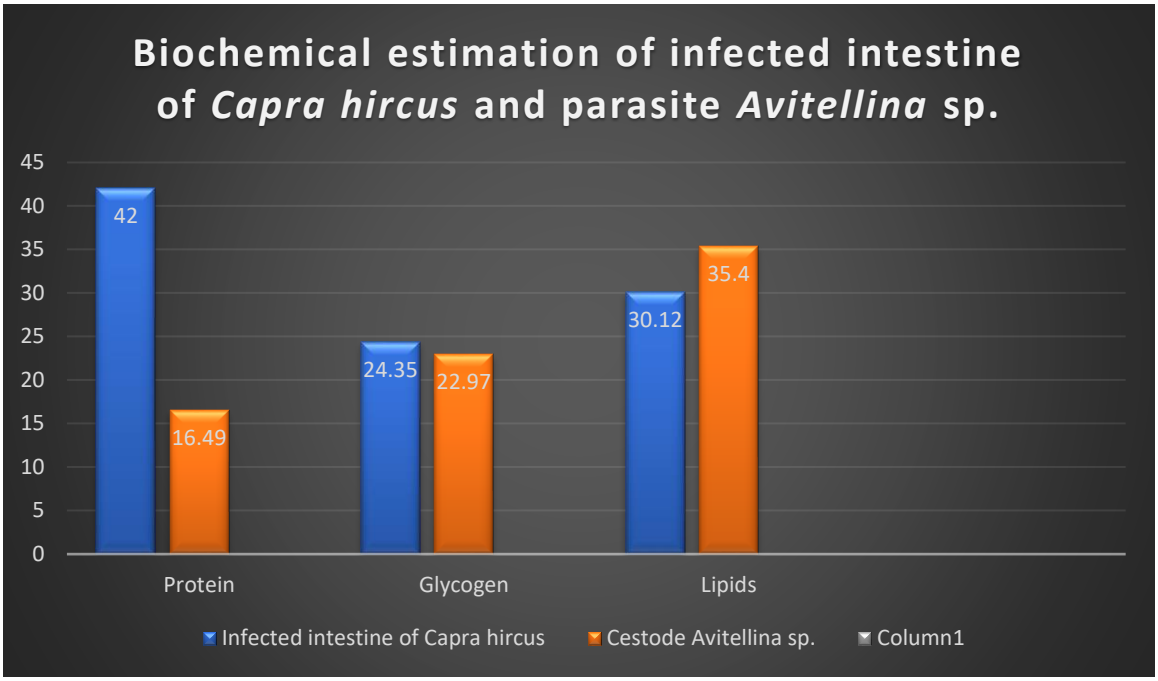
Result and Discussion-

In the present investigation ,biochemical estimation in Cestode parasites i.e. *Avitellina* sp. was carried out . It shows that the protein content of cestode *Avitellina* sp. obtained 16.49 mg/g weight of tissue protein while infected intestine of ruminant *Capra hircus* obtained 42 mg/g weight of tissue from above result it is concluded that the worm *Avitellina* maintain a good balance in protein content with the host *Capra hircus*. Protein content is lower in cestode parasites as compare to host. The glycogen content of *Avitellina* sp. obtained 22.97 mg/100 ml of solution. While infected intestine of ruminant *Capra hircus* obtained 24.35 mg/100 ml of solution but the lipid content of *Avitellina* sp. obtained 35.40 mg/ gm of weight of tissue while in infected intestine of goat ,*Capra hircus* obtained 30.12 mg/gm of weight of tissue.

From the present experimental study it has been observed that the percentage of lipid is high in parasites as compared to protein and glycogen. These parasites absorbing most of nourishment from host and fulfilling its need and causing hindrance in the proper development of host (B. V. Jadhav et al. 2008)

Table No. 1: Biochemical estimation of *Avitellina* sp. From host *Capra hircus*

Name of parameter	Host infected intestine (*Capra hircus*)	Cestode parasite (*Avitellina* sp.)
Protein	42.00 mg/gm. wt. of tissue	16..49 mg/gm. wt. of tissue
Glycogen	24.35 mg/ 100ml of sol^n	22.97mg/ 100ml of sol^n
Lipids	30.12mg/gm wt. of tissue	35.40 mg/gm wt of tissue



Conclusion

From the above biochemical estimation it is concluded that the percentage of lipid is high in parasite *Avitellina* as compared to protein and glycogen. The parasite *Avitellina* absorbing most of nourishment from host *Capra hircus* causing interruption in it's proper development thus reduces it's food value.

Acknowledgement

The authors is thankful to Principal ,B.Raghunath ACS college Parbhani (Maharashtra) for providing the laboratory facilities to complete this research work.

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Molecular nature and functional characterization of the QTLs/genes for rice yield related traits

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Keywords:
Rice, Yield, QTL mapping, Gene Cloning

Abstract:
Rice is a staple food for more than half of the world's population. More than 90 per cent of the world's rice is grown and consumed in Asia, where 60 per cent of the global population lives. Rapid growth in human population throughout the world is boosting demand for a corresponding increase in grain yield and there is need to increase production 50 per cent more by 2025. Therefore, an increase in rice production remains a challenge today. To achieve this ambitious goal various rice varieties with greatly improved agronomic traits such as high yield potential should be developed. Over the past 20 years, the development of DNA markers and genomic sequencing technology have led to rapid progress in the QTL mapping and cloning of genes underlying grain shape and grain weight in rice. These are made by analysis of segregating plant populations derived by crossing parents with contrasting characteristics. In recent years, tremendous progress has been attained and many QTLs for rice yield traits have been isolated and functionally analyzed in detail, which provides new sights into the molecular mechanisms of the formation of rice yield traits. To date, ~45 QTL/genes associated yield and its contributing traits have been isolated by map-based cloning strategies. The association or linkage of particular molecular marker variants (alleles) with a heritable phenotypic trait enables us to identify which region of a particular chromosome is important for the phenotype.

1. Rice

Rice is a staple food for more than half of the world's population (Delseny*et al.*, 2001). Rice has been cultivated for more than 7,000 years (Yunfei*et al.,* 2007 andZong*et al.,* 2007). The estimate of world paddy production in 2011 is 7.2 × 108 t (4.8 × 108 t, milled basis), and the global rice yield must reach 8 × 108 t in 2025 to meet the demand for rice consumption (FAOSTAT, 2010). This additionalamount of rice has to be produced by using less land, less water, less labor, and fewer chemical inputs. Therefore, an increase in rice production remains a challenge today.More than 90 per cent of the world's rice is grown and consumed in Asia, where 60 per cent of the global population lives. Rice accounts for 35 to 60 per cent of the calories consumed by 3 billion people in Asia alone. Rapid growth in human population throughout the world is boosting demand for a corresponding increase in grain yield (Liang *et al.,* 2010) and there is need to increase production 50

per cent more by 2025. For rice consuming countries there is need to produce 40 per cent more rice by 2030 (Khush, 2005 and Zhu *et al.,* 2011). To achieve this ambitious goal various rice varieties with greatly improved agronomic traits such as high yield potential, stress tolerance etc., should be developed.

Rice is unique in its ability among cereal crops to grow in a wide range of environments. Depending on the hydrology, the rice environment can be classified intofour major ecosystems are generally recognized (Khush, 1984) as follows: (1) irrigated, (2) rainfed lowland, (3) upland, and (4) floodprone. These environments vary with respect to elevation, rainfall pattern, depth of flooding and drainage, hydrological status, soil type and by the adaptation of rice to agro ecological factors (Huke and Huke, 1997 and Maclean *et al.,* 2002). Approximately, 55% of the world rice area planted to rice is irrigated and is the most productive rice growing system, perhaps contributes 75% of the world rice production. Large areas of rice are grown under lowland and upland rainfed conditions.As a complex agronomic trait, grain yield of a rice plant is multiplicatively determined by three component traits: number of panicles per plant, number of grains per panicle, and grain weight.Different mappingpopulations have been used to explore the QTLs controlling yield related traits.

Rice to be grown successfully under a variety of climatic conditions across the globe; breeders maintain rice at high genetic diversity. Globally, rice is grown on 154 million hectares (Mha), and approximately 45 % of this area is under rainfed conditions that have very low-yield potential (Verulkar*et al.,* 2010). About 80 million ha of irrigated lowland provide 75% and about 60 million ha of rainfed lowlands supply about 20% of the rice production. Rice needs more water compared to other crops, on an average about 2,500 liters of water is needed to produce 1 kg of rough rice. Irrigated rice receives an estimated 34-43% of the total world's irrigation water. Worldwide water for agriculture is becoming increasingly scarce, day by day due to uncertain and uneven rainfall distribution patterns, shrinking groundwater resources, increasing level of salts in soil solution and diverting the fresh water resources to competing urban and industrial uses. In the coming future, water availability may be more affected due to ongoing changes in global climate. Because of semi-aquatic ancestry, rice is extremely sensitive to water shortage. Drought is the major constraint to rice production in rainfed areas across Asia. Drought can be simply defined as reduction in yield due to shortage of water (Bernier *et al.,* 2008). Throughout the world, about 34 per cent (~54 million hectare) of the total land under rice cultivation is under rainfed condition (Maclean *et al*., 2002). Asia occupies 32.1 per cent rainfed low land rice of the total rice area which currently averages production of 2.3 tonnes per hectare (Tuong and Bouman, 2003). Drought is the most devastating among abiotic stresses and it depresses yield by 15-50 per cent depending on the vigor and period of stress in rice (Srividya*et al*., 2011). The global reduction in rice production due to drought averages 18 million tonnes annually (O'Toole, 2004). Rice is sensitive to drought stress during reproductive growth and even moderate stress can result in drastic reduction in grain yield (Hsiao,1982; O'Toole, 1982 andVenuprasad*et al.,* 2008).

2. Yield and its component traits

As a complex agronomic trait, grain yield of a rice plant is multiplicatively determined bythree component traits: number of panicles perplant, number of grains per panicle, and grainweight. Number of panicles is dependent on the ability of the plant to produce tillers (tilleringability), including primary, secondary, and tertiary tillers. Number of grains per panicle can also be attributed to two subcomponents: number of spikelets, which is mainly determined by the numbers of primary and secondary branches, and seed setting rate of the spikelets. Grain weight is largely determined by grain size, which is specified by its three dimensions(length,

width, and thickness), and the degree of filling(Xing *et al*.,2010).Rice varieties differ tremendously in thelevels of grain yield, with immense variabilityin the combinations of component traits owingto the vast diversity of genetic constitutions.In addition, yield levels of rice varietiesare also greatly influenced by the environmentalconditions and the field management practices.There are also remarkable interactionsbetween genotypes and environments such thatvarieties are adapted to specific environmentalconditions.

Besides these traits the other physiological traits of plant system also contributed to enhance yield. The traits like Plant height (PHT), Total Number of tillers (NT), Effective number of tillers (ENT), Flag leaf length (FLL), Flag leaf width(FLW) and Panicle length(PL) also contributed to increases the grain yield. Many studies show that these traits contributed to yield indirectly by enhancing the rate of photosynthesis and thus increases the rate of transportation of photo assimilates from source to sink and thus contributed to enhance yield.

3. Molecular markers and dissection of the molecular bases of rice yield traits

The inheritance of quantitative traits classicallyinvolves multiple genes, each having a small effectthat is sensitive to environmental changes.These traits are known in general as having lowheritability and thus have earned the reputationof being difficult to investigate. However,the development of molecular marker, genome mapping, and QTL analysis technologies hasgreatly facilitated the investigation of geneticbases of quantitative traits. In rice, researchershave constructed high-density genetic linkagemaps based on restriction fragment length polymorphism(RFLP) and simple sequence repeat(SSR) markers (McCouch*et al*.,1988; Kurata*et al*., 1994 and McCouch*et al*.,2002). With the rapid development of different types of DNA markers, marker-assisted selection (MAS) has been playing a prominent role in plant breeding. Both random genomic marker and genic marker could be used in MAS. The random genomic markers like RFLP have been used to construct high density linkage maps based on restriction fragment length polymorphism and simple sequence repeat (SSR) markers for genotyping experiments. However, are limited in MAS application due to their relatively low accuracy in selection caused by the genetic recombination between the marker and the target gene. The genic or functional markers, derived from polymorphic loci within genes affecting phenotypic variation, would overcome the problem of the recombination, and thus are highly predictive of phenotype, and will facilitate efficient selection of favorable alleles in breeding programs (Andersen and Lubberstedt, 2003). However, there are still many challenges such as existence of any particular alleles in a given breeding line and availability of user friendly DNA markers for MAS application in complex traits (Xu*et al.,* 2005). Identification of agronomically important genes and mining of the alleles in natural populations are primarily required to develop the genic or functional markers (Takeda and Matsuoka, 2008).

Fan*et al.* (2009)has been developed a cleaved amplified polymorphic sequence (CAPS) marker based on the C-A mutation in GS3 loci, a gene contributed for grain size. This CAPS marker is highly associated with grain length, thus could be used for selection of rice grain length in breeding. However, its efficiency in MAS is still limited as the PCR product needs to be digested by a restriction endonuclease and this procedure is relatively expensive and elaborative once applied to a largebreeding population. It would be useful to develop somePCR-based functional markers for grain length improvement.Knowledge of the allelic diversity in *GS3* and their effects would be helpful for genetic manipulation of grain size in rice. Takano-Kai *et al.*(2009) also demonstrated that the C-A mutation in *GS3* gene played a critical role in seed size differences among the modern subpopulations of *O. sativa*.

Yan CJ *et al.*(2009) compare the nucleotide sequencesencoding the OsCKX2 enzyme on the *Gn1a* locus, a gene for grain size revealed a 16-bp deletion in the 50-untranslated region and an 11-bp deletion in the coding region in the two high-yielding rice cultivars Habataki and 5150, respectively, as compared to those in the Koshihikari cultivar. On the basis of sequence variation among different alleles, two sequence-tagged site (STS) markers, *Gn1a*-M1 and *Gn1a*-M2, were developed for genotyping *Gn1a* alleles.

In another study of Wang (2011)three polymorphic loci, namely SR17, RGS1, and RGS2, were found on the second intron, the last intron and the final exon of *GS3*, respectively. The mutation in the cysteine codon (TGC) in the small grain group was changed to a termination codon (TGA) in the large-grain group. Numerous insertion/deletion and nucleotide polymorphisms at these three loci were identified besides the C-A mutation at the locus SF28 within *GS3* in a wide collection of rice germplasm.

4. QTL Fine Mapping

To narrow down the targated QTL region of interest a fine mapping strategy was used in several studies. This will help to easy transfer the targated QTL into new cultivars of interest and their identification into it. Based on sequence information the different types of molecular markers like SSR, SNP, Indels had been designed in the targated QTL region using bioinformatics tools. Onces the markers were identified their cosegression pattern used to identification of flanking region which was associated with our trait of interest.

4.1 Approaches based on advanced populations

F2, DH or RIL populations are powerful and adequate to fine map qualitative genes if there are plenty of polymorphism markers. To date, only a few genes have been identified and isolated using F2 populations: e.g. *SRS3*, *DEP2*/*EP2*, *DEP3*, *EP3*, *APO1* and *GIF1*. The mutation of loss-of-function or gain-of-function versions of these genes causes a significant variation, in which individuals of the target population can be easily classified into groups. However, it is difficult to precisely estimate the genetic effect of a single QTL in a primary mapping population due to the genetic background noise in a F2 population. Moreover, it is difficult to fine-map a major QTL in a F2 population, l*et al*one a minor QTL. In view of this point, NILs were proposed by Tanksley and Nelson (1996) for isolating and cloning a candidate QTL/gene. The target QTL region is segregated and the genetic background is fixed in the NILs. Compared with the primary mapping population, NILs eliminate the genetic background noise. Thus, the related trait controlled by a target QTL shows the characterization of a Mendelianfactor with a segregation ratio of 3:1 in the NIL-F2 population. In addition, a QTL can explain more variation in the NIL than in the primary population. Hence, NILs are widely used for QTL fine-mapping and isolation. Many QTLs have been fine-mapped and cloned using consecutive backcrossing strategy, known as CB-NILs populations (Ashikari*et al.*, 2005; Fan *et al.*., 2006;Song *et al.*, 2007; Xing *et al.*, 2008; Xue*et al.*, 2008 and He *et al.*, 2010). However,construction of CB-NILs is time consuming; usually taking three years to develop NILs in rice after QTL information is obtained.

The RIL populations, such as F6 and F7, are obtained by single-seed descent method. TheF6 plant and its F7 progeny show a nearly identical phenotype. However, if one unknown gene/QTL for a trait is located in the heterozygous region, its progeny should segregate in the target trait which is easily observed. Thus, the progeny of the heterozygous plant consists of the NILs of the unknown gene/QTL. This type of NIL is known as trait performance based NILs (TP-NILs) as they are obtained according to trait performance without any QTL information in advance.The TP-NILs strategy depends completely on the variation in trait performance. Hence, it is an efficient way to identify the major QTLs that cause large trait variations.

Another strategy for NIL development is the heterogeneous inbred family (HIF-NILs) method. HIF plants based on primary QTL mapping information are screened from inbred recombinants with marker genotypes. Self-pollinating the heterozygous plant produces NIL-F2. Such NILs are suitable for mapping and isolating either major or minor QTLs. A major QTL (*SPP1*) and two minor QTLs (*qGL7* and *qGL7–2*) for grain length have all been successfully fine mapped using such NILs (Liu *et al.,* 2009; Bai*et al.,* 2010 and Shao *et al.,* 2010). In genetics, both strategies of TP-NILs and HIFNILs employ the same method in searching a plant carrying a heterozygous region harboring a QTL in the high generations of RIL. The difference between them is that TP-NILs are obtained simply based on the varied trait performance within a RIL, and HIF-NILs are based on the genotype of the QTL region. These three strategies of NIL development are all successfully used for QTL fine-mapping. The genetic makeup of TP-NILs and HIF-NILs combines their two parents' genomes, whereas CBNILs carry an identical genetic background to the recurrent parent except for the target QTL region.

Chromosome segment substitution lines (CSSLs) are an advanced population, developed with a similar strategy to that for CB-NIL. Based on MAS, a set of CSSLs, in which donor segments cover the whole rice genome, can be obtained. CSSL population is a powerful tool in detection of either major or minor QTLs, and has therefore been very popular in rice and other crops over recent years (Cheng *et al.,* 2011 andGuo*et a.,*. 2011).

An ideal panicle structure is important for improvement of plant architecture and rice yield. Peng*et al.* (2014) identified a quantitative trait locus (QTL), designited qPPB3 for primary panicle branch number, using recombinant inbred lines (RILs) of PA64s and 93-11, With a BC3F2 population derived from a backcross between a resequenced RIL carrying PA64s allele and 93-11, qPPB3 was fine mapped to a 34.6-kb genomic region on chromosome number 03.

4.2 Approaches based on natural population

At present, the large amount of germplasm preservedin gene banks (ex situ) and in situ throughout theworld provides the groundwork for identifying new genescontrolling yield and other valuable traits (Tanksley andMcCouch, 1997).Simple sequence repeat (SSR) and simple nucleotide polymorphism (SNP) are the most informative genetic markers useful for genetic diversity studies (Russell *et al.,* 2000, Sjakste*et al.,* 2003and Hamza*et al.,* 2004) and mapping.

Association mapping, also known as linkage disequilibrium(LD) mapping, is a new method of mapping QTLs that takes advantage of historic LD to link phenotypes to genotypes.Association mapping based on natural populations (unrelatedindividuals) is widely used for QTL mapping due to the rapidLD decay in maize (Yu and Buckler, 2006). Agrama*et al.*(2007) used the mixed linear model (MLM) method to disclose the associations between 123 SSR markers and yield components in rice. Analogically, rice, a highly selfing species,is also an ideal candidate for association mapping due tothe following features: rich resources of germplasm and beinggenotyped once and repeatedly phenotype. Two major grain-size QTLs, *GS3* and *qSW5*, were fine-mapped to regions of 120 kb and 72 kb, respectively. However, the peak signals of association loci often appeared near (but not within) the known genes. These situations are consistent with slow LD decay over 100– 250 kb (Mather *et al.,* 2007 and McNally *et al.,* 2009), which may explain the low resolution mapping. Compared to linkage analysis, GWAS has not identified many QTLs in rice. Surprisingly, some cloned major QTLs (e.g. *Ghd7* and *Ehd1*) regulating rice flowering, could not be identified by GWAS. However, recent mutations resulting in large trait changes would be detected by linkage analysis, but not by association mapping due to its rare occurrence in the natural population. For instance, the *GW2* wider-grain allele is found in very few varieties, e.g. WY3 and Oochikara (Takano-Kai *et al.,* 2009).It is expected

thatassociation mapping will detect more genes/QTLs with higherresolution compared to linkage analysis. Some major QTLs cloned by linkage analysis have been fine-mapped to 100 kb(Huang *et al.,* 2010). Huang *et al*. (2010) used ~3.6 million SNPs with 517 rice landraces. Lakew*et al*.(2010) demonstrated association mapping of drought-related traits in barley using SSR and SNP markers. Yan*et al*. (2011) reported that SSR markers provided more information on genetic diversity and performed better at clustering all lines into groups than SNPs. Since both the DNA markers are efficient at association mapping and population studies. Zhao *et al.*(2011) reported GWAS using 44,100 SNPs with 413 rice accessions. In another experiment byZhao *et al*. (2010) studied 130 rice accessions with 170 SSR markers to identify marker–trait associations by MLM for grain quality. There are numerous reports on genomewide association studies (GWAS) in rice using SNPs.

A major QTL for grain length, qGRL-1.1 has been mapped to a 108-kb region between markers RM431 and CHR1.1 on chromosome 1 (Singh *et al*., 2012). GW3 and GW6 are major grain weight QTLs that have been fine mapped on chromosome 3 and 6, respectively. GW3 has been narrowed down to a 122-kb physical distance containing 16 open reading frames. The cloned GS3 gene is located in this region and it remains to be determined whether they represent the same locus.

Notably, several QTLs that affect the vascularbundle system in rice have been further cloned by positionalcloning. For instance, genes such asAPO1andNAL1have beenshown to involve in enhanced translocation capacity ofvascular bundles (Fujita *et al*., 2013).Fujita *et al.* (2013) identified the major QTL for number of spikelet per panicle i.e.NAL1 indirectly involved in translocation capacity of photosynthats had been fine map to 18kb between markers Ind4 and Ind 12. Recently another QTL for spikelet per panicle, qSPP6 map to a 429kb and cosegregate with markers RM20521 and Ind1.LSCHL4 a QTL for flag leaf shape and chlorophyll content has been mapped on chromosome 4 to a 18.97kb within T2957-2 and RM348 marker.qFLW7.2, a new major QTL for flag leaf width, was fine mapped within 27.1 kb region on chromosome 7. Both qFLW7.2 and qPY7 were located in the interval of 45.30 ~ 53.34 cM on chromosome 7, which coincided with the relationship between yield per plant (PY) and flag leaf width (FLW)(Zhang *et al*., 2015).

5.Cloning and functional characterization of Yield related traits

Higher yields of rice have always been apredominant goal in rice breeding techniques. However,the inheritances of rice yield and its componentsare still unknown, and no information regardingsuitable alleles can be directly provided for improvingthe rice yield level. Over the past 20 years, the development of DNA markers and genomic sequencing technology have led to rapid progress in the mapping and cloning of genes underlying grain shape and grain weight in rice (Ashikari*et al*., 2006). To date, ~40 QTL/genes associated yield and its contributing traits have been isolated by map-based cloning strategies. Most of them are still poorly understood, particularly with regard to their functions at the biochemical and cell biological levels.

Dwarf1 (D1), also known as the rice heterotrimeric G protein alpha subunit (RGA1), D2, D11, and D61. Mutations in these genes result in dwarf plants and have detrimental pleiotropic effects on organ growth, including a reduction in seed size. D1/RGA1 was the first gene to be cloned that had substantial effects on seed-size regulation (Ashikari*et al*., 1999 and Fujisawa *et al*., 1999). An 833-base pair (bp) deletion of D1 disrupts the coding region of the heterotrimeric G protein alpha subunit and results in dwarf plant phenotypes with smaller grain. Genes affecting brassinosteroid (BR) biosynthesis and signal transduction have also been shown to regulate grain size in rice. D2 and D11 encode two cytochrome P450 oxidoreductase enzymes involved in BR biosynthesis (Hong *et al*., 2003) and D61 encodes a BR receptor, an ortholog of BRI1 in Arabidopsis

(Arabidopsis thaliana) (Yamamuro*et al*., 2000). The D2 protein represents a novel type of P450 (CYP90D2) that catalyzes the steps from 6-deoxoteaster-one to 3-dehydro-6-deoxoteasterone and from teasterone to 3-dehydroteasterone in the late BR biosynthesis pathway (Hong *et al*., 2003). The D11 P450 (YP724B1) enzyme is required for the supply of 6-deoxotyphasterol and typhasterol in the BR biosynthesis network (Tanabe *et al*., 2005).

Gn1a, a major QTL, has been cloned andcharacterized for the grain number per panicle trait inthe Koshihikari/Habataki-derived advanced backcrosspopulation. Functional analysis revealed that Gn1aencoded cytokininoxidase/dehydrogenase (OsCKX2),an enzyme that degrades the phytohormonecytokinin(Ashikari*et al*., 2005). Grain weight is usually representedby 1,000-grainweight in breeding techniques andis determined by grain width, length, and thickness.ManyQTLs associated with rice grainweight have beenidentified in the last decade.Of these,GS3, a majorQTLfor grain length and weight and also as a minor QTL forgrain width and thickness, has been mapped onchromosome 3. These cloned genes governing rice yield componentsnot only elucidate themolecular mechanisms regulatinggrain number per panicle and grain weight and widthtraits but also directly provide information on sequencevariation. This will enable us to design gene-taggedmarkers in the application of MAS for yield traits.

To clone Ghd8, the NIL-F2 population (NIL-F2(HR5)) wasobtained from a heterozygous F7 plant derived from a crossbetween an old version of Zhenshan 97 (ZS) and HR5 (Zhang *et al*., 2006) was used. In rice, each HAP subunit is encoded by a gene family;therefore, various possibilities were available to form differentHAP complexes, which regulate multi-developmental processes.This explanation may account for the pleiotropic effectsof these Ghd7/Ghd8 genes.

GRAIN SIZE 3 (GS3) is a major QTL for grain length and weight and a minor QTL for grain width and thickness and functions as a negative regulator for grain size (Fan *et al*., 2006). GS3 was originally detected from the progeny produced by a cross between Minghui 63 and Chuan7. The GS3 protein contains an organ size regulation (OSR) domain in the N terminus, a transmembrane domain, a tumor necrosis factor receptor/nerve growth factor receptor (TNFR/NGFR)-like domain, and a von Will-ebrand factor type C (VWFC) domain in the C terminus. The OSR domain functions as a negative regulator of grain length and deletion mutants of this domain result in the formation of long-grain rice. The C-terminal TNFR/NGFR and VWFC domains act as positive regulators of grain length and loss-of-function mutations of these domains lead to the development of very short grain (Fan*et al*., 2009 and Mao *et al*., 2010). A molecular maker based on GS3 has been developed for the selection of long-grain lines in rice breeding programs (Wang *et al*., 2011). The GS3 ortholog in maize (Zea mays) has also been cloned and characterized. ZmGS3 has functional domains in common with the rice GS3 protein and ZmGS3 has been shown to be involved in maize kernel development (Li *et al*., 2010).

GIF1 was primarily mapped with simple sequence repeat markers using 300 F2 gif1 individuals and was further placed in a 32kb region between the markers CAPS4 and CAPS8 (Wang *et al*., 2008). GIF1 was also shown to cosegregate with CAPS7 using 900 F2 mutant plants. The grain incomplete filling 1 (gif1) mutant was isolated from a screen for mutants with grain-filling defects. The GIF1 mutant also has more grain chalkiness as a result of loosely packed starch granules. GIF1 encodes a cell-wall invertase required for carbon partitioning during early grain filling. A frameshift mutation caused by a 1-bp nucleotide deletion in GIF1 results in premature termination of its open reading frame. The GIF1 gene is expressed in a more restricted pattern in the flowers of cultivated rice varieties than in the flowers of wild rice, which is apparently a consequence of accumulated changes in the regulatory sequence of the promoter as a result of

domestication. The identified quantitative trait locus (QTL) Ghd7, isolated from an elite rice hybrid and encoding a CCT domain protein, has major effects on an array of traits in rice, including number of grains per panicle, plant height and heading date. Enhanced expression of Ghd7 under long-day conditions delays heading and increases plant height and panicle size. Natural mutants with reduced function enable rice to be cultivated in temperate and cooler regions. Thus, Ghd7 has played crucial roles for increasing productivity and adaptability of rice globally. To precisely map the Ghd7 locus, theyassayed 1,082 plants showing the recessivephenotype for all the three traits (short,early heading and small panicle) from population1 with two simple sequence repeatmarkers, RM5451 and RM1135, bracketingthe Ghd7-containing region. By further genotypingof the recombinant plants using markersRM3859, RM5436, C39 and RM7110, they localized the Ghd7 locus to the intervalbetween RM3859 and C39, cosegregatingwith RM5436. Ghd7 haspleiotropiceffect similar to those of Ghd8.GRAIN WIDTH 5 (GW5) has been identified as a major QTL for Seed Width on chromosome 5 (qSW5) for the determination of rice grain width and weight (Shomura*et al*., 2008; Wan *et al*.,2008 andWeng*et al*., 2008). A survey of GW5/qSW5 polymorphisms in various rice landraces has revealed that deletions in this gene may have played an important role in the selection of increased grain size from artificial and natural crossings during rice domestication (Shomura*et al*., 2008). The GW5/qSW5 gene encodes a nuclear protein of 144 amino acids with an arginine-rich domain. Because GW5/qSW5 physically interacts with polyubiquitin, it is likely to act as a regulator in the ubiquitin– proteasome pathway and regulates cell division of the outer glume of the rice spikelet.

In particular, researcher mapped a major QTL for grain width, GW2, with the WY3 allele at GW2 contributingto increased grain width. Previous studies have mapped two QTLsfor grain width near the GW2 region on the short arm of chromosome2 (Fan *et al*., 2009) suggesting they may be the same QTL and thatthe GW2 locus may contribute to increased grain width in the various rice varieties. Map-based cloning of the GW2 QTLby using two parental varieties that showed highly significant differencesin grain size to more easily identify the QTL. The crossed of Japonicavariety, WY3, with a very large grain (1,000-grain weight, 41.9 g ±1.3 g) and a high-quality elite indica variety, Fengaizhan-1 (FAZ1),with a small grain (1,000-grain weight, 17.9 ± 0.7 g) toproduce an F2 population. GW2 was initially detected from a cross between a large-grain japonica rice variety, WY3, and a small-grain indica rice variety, Fengaizhan-1 (FAZ1). A 1-bp deletion in the GW2 gene in WY3 results in the introduction of a premature stop codon in its exon 4, causing the large-grain phenotype in WY3. GW2 negatively regulates cell division by targeting its substrates to proteasomes for regulated proteolysis; loss of GW2 function results in an increase in cell number in the spikelet hull and acceleration of the grain-milk filling rate, thus enhancing grain width, weight, and yield. There are two homologs of the rice GW2 in maize, referred to as ZmGW2-CHR4 and ZmGW2-CHR5, both of which contribute to the phenotypic variation in kernel size and weight (Li *et al*., 2010).

The another group of cloned genes associated with grain shape and weight includes the SMALL AND ROUND SEED (SRS) loci identified in the japonica rice subspecies. Mutations in SRS1 result in reduction in both cell length and cell numbers in the longitudinal direction, and elongation of the cells in the lateral direction of the lemma of rice flowers. Deletions of 38 bp in srs1-1 and 31 bp in srs1-4 disrupt the coding region. Other srs1 mutant alleles are caused by alterations in the stop codon and mRNA splicing sites (Abe *et al*., 2010). The SRS1 mRNA and proteins are abundant in young leaves, internodes, and panicles. SRS1 encodes a protein of 1365 amino acids with no known functional domains (Abe *et al*., 2010). The small and round seed

phenotype of srs3 is a result of the reduction in cell length of the lemma. The SRS3 protein contains a kinesin motor domain and a coiled-coil structure and is a member of the kinesin 13 subfamily (Kitagawa *et al*., 2010). The cell length of the lemma in srs5 mutants is shorter than that in the wild type plants. A 1-bp substitution in the fourth exon of SRS5 is responsible for the phenotype. SRS5 encodes alpha-tubulin and may regulate cell elongation in a pathway independent from the BR signaling network.

Besides these other class of genes related to yield has been cloned such as, GRAIN SIZE on chromosome 5 (GS5) is a major QTL affecting grain width, grain filling, and grain weight (Li *et al*., 2011). It encodes a serine carboxypeptidase and functions as a positive regulator of grain size. Analysis of genomic DNA sequences and promoter swaps in transgenic plants reveals that nucleotide changes in three segments of the GS5 promoter seem to be responsible for the variations in grain width (Li *et al*., 2011). Grain width 8 (GW8) was identified from a cross between HXJ74 and Basmati385 as a major QTL affecting grain width and grain yield (Wang *et al*., 2012). A recent gene-cloning project has revealed that GW8 encodes SQUAMOSA pro-moter-binding protein-like 16, referred to as OsSPL16, which belongs to the protein family of SBP domain-containing transcription factors. There are six polymorphisms in the DNA sequence of OsSPL16 between HXJ74 and Basmati385. Among them, a 10-bp deletion in the promot-er region has been shown to be responsible for the slender grain trait of Basmati385 (Wang *et al*., 2012).

To explore the relationship between floral induction and yield formation and the molecular mechanism ofpanicle development in rice, a novel mutant, ghd10, was identified from japonica variety Wuyunjing 7 plants subjectedto ethyl methanesulfonate (EMS) treatment. Hu *et al*.(2013)used the F2 segregation populations were used for a $\chi 2$ test. For map-based cloning, an F2 segregation population for mapping derived from a cross between the ghd10 mutant and the indica cultivar NJ06 was constructed to identify the gene in the mutants. The parents and 6,846 F2 individuals were planted in a paddy field, among which 978 with the mutant phenotype were used to map theGhd10. The ghd10 mutant exhibited delayed flowering time, tall stalks andincreased panicle length and primary branch number. Map-based cloning revealed that Ghd10 encodes a transcriptionfactor with Cys-2/His-2-type zinc finger motifs. Ghd10 is orthologous to INDETERMINATE1 (ID1), which promotesflowering in maize (Zea mays) and is identical to the previously cloned genes Rice Indeterminate1 (RID1), Early heading date2 (Ehd2) and OsId1. Ghd10 mutation has pleiotropic effects on grain yield, heading date and plant height and functions like Ghd7 and Ghd8/DTH8. A longer vegetative growth period allows more photosynthate(the source) to be transferred to grains, i.e., the sink capacity increases, after flowering. When a plant has a large sink capacity and the flow between the sink and the source is unimpeded, the yield formation potential increases. For genetic analysis to determine whether a dominant or recessive, single or multiple gene controls the ghd10 phenotype, reciprocal crosses between ghd10 and the japonica cultivars NIP, WYJ7 and CJ06 were conducted.

In recent year a QTL ghd10 has been cloned using mutant plant population on chromosome 10. This was the QTL identified for plant height and panicle development in Wuyunjing 7 mutant population. The markers k10-5 and k10-3cosegrigated with ghd10 (Hu *et al*., 2013).Fujita *et al*. (2013) identified a gene, SPIKELET NUMBER (SPIKE), from a tropical japonica rice landrace that enhances the grain productivity of indica cultivars through pleiotropic effects on plant architecture. Map-based cloning revealed that SPIKE was identical to NARROW LEAF1 (NAL1), which has been reported to control vein pattern in leaf. Phenotypic analyses of a near-isogenic line of a popular indica cultivar, IR64, and overexpressor lines revealed increases in

spikelet number, leaf size, root system, and the number of vascular bundles, indicating the enhancement of source size and translocation capacity as well as sink size.

The basic premise of high yield in rice is to improve leaf photosynthetic efficiency and coordinate the source sink relationship in rice plants. Quantitative trait loci (QTLs) related to morphological traits and chlorophyll content ofrice leaves were detected at the stages of heading to maturity, and a major QTL (*qLSCHL4*) related to flag leaf shape andchlorophyll content was detected at both stages in recombinant inbred lines constructed using the *indica*rice cultivar93-11 and the *japonica* rice cultivar Nipponbare. Map-based cloning and expression analysis showed that *LSCHL4* is allelicto *NAL1*, a gene previously reported in narrow leaf mutant of rice(Zhang *et al.*, 2014).

Table 1: The cloned grain yield related genes/QTLs

Traits	QTL/Gene	Chr	Intervals (kb)	Population (No.)	Causal mutation	Reference
plant height and panicle development	ghd10	10	k10-5 and k10-3	Wuyunjing 7 plants subjected to ethyl methane sulfonate (EMS) treatment	Point mutation	Shikai Hu, 2013
GN	Gn1a	1	3A28-3A20	CB-NIL(13000)	Deletion	Ashikari*et al.*, 2005
TN/GN	*MOC1*	6	17-2-12-2	F2 (2010)	insertion	Li *et al.*, 2003
GW/GS	*GS3*	3	GS63-SF19 (7.9)	CB-NIL (5740)	Premature stop	Fan *et al.*, 2006
HI/GN	*APO1*	6	R3819-C11635 (11.4cM)	F2 (-)	Substitution	Ikeda *et al.*, 2007
GN (2284)	*Ghd7*	7	RM5436-C39	CB-NIL (3200)	Deletion	Xue*et al.*, 2008
GW/GF	*GIF1*	4	SSLP1-CAPS8 (86)	CB-NIL (5384)	Premature stop	Wang *et al.*, 2008
			CAPS4-CAPS8 (32)	F2 (3600)		
TN/ GN	*PROG1*	7	S3204-P71 (14)	CB-NIL (3051/14400)	Substitution	Jin *et al.*, 2008
TN/ GN	*PROG1*	7	pr5-pr7 (8.8)	CB-NIL (3051/14400)	Substitution	Tan *et al.*, 2008
GW/GS	*qSW5/GW5*	5	MS40671-M16 (2.3)	CB-NIL (4501/8720)	Deletion	Shomura*et al.*, 2008;
GW/GS	*qSW5/GW5*	5	CW5-CW6 (21)	CB-NIL (4501/8720)	Deletion	Weng*et al.*, 2008

GN	*DEP1*	9	S2-S11-2 (85)	CB-NIL (1311)	Premature stop	Huang *et al.*, 2009
TN/GN	*EP3*	2	STS5803-5-STS5803-7 (46.8)	F2 (987)	Substitution	Piao*et al.*, 2009
TN/GN	*LRK1*	2	RM279-RM5654 (*qGY2-1*)	Over-expression	Over-expression	Zha*et al.*, 2009
GS/GN	*SP1*	11	M7-M8 (8)	F2 (1500)	Deletion	Li *et al.*, 2009
GN	*Ghd8/DTH8*	8	Ind8-47-Ind8-15 (47)	CB-NIL (15000)	Frameshift	Wei *et al.*, 2010
GW/GF	*FLO(a)/FLO2*	4	P18-P22 (81)	F2 (1571/4000)	Substitution	Qiao*et al.*, 2010
GW/GF	*FLO(a)/FLO2*	4	218042-218787 (37)	F2 (1571/4000)	Substitution	She *et al.*, 2010
TN/GN	*IPA1/WFP*(*OsSPL14*)	8	M4-M5 (78)	CB-NIL (5500)	Substitution	Jiao *et al.*, 2010
TN/GN	*IPA1/WFP*(*OsSPL14*)	8	dCAPS825-CAPS311 (2.6)	RIL-F3, F4 (3000)	Substitution	Miura *et al.*, 2010
GW/GS	*SRS3*	5	3130-3151 (21)	F2 (4800)	Substitution	Kitagawa *et al.*, 2010
GW/GS	*EP2/DEP2/SRS1*	7	P5-P7 (30.3);	F2 (821/810/6000)	Deletion	Zhu *et al.*, 2011
GW/GS	*EP2/DEP2/SRS1*	7	M2-M15 (27);	F2 (821/810/6000)	Deletion	Li *et al.*, 2010;
GW/GS	*EP2/DEP2/SRS1*	7	25285As-25340As (55)	F2 (821/810/6000)	Deletion	Abe *et al.*, 2010
HI/GN	*APO1*	6	3628-41-3628-56 (11)	CB-NIL (10000)	Substitution	Terao*et al.*, 2010;
GN	*DEP3*	6	P21-P23 (73)	F2 (887)	Deletion	Qiao*et al.*, 2011
GN	*Ghd8/DTH8*	8	SEQ3-1-SEQ5-1 (70)	TP-NIL (2256)	Frameshift	Yan *et al.*, 2011
GW/GS	*GS5*	5	S2-RM574 (11.6)	CB-NIL (5265)	Substitution	Li *et al.*, 2011
Seed size	*D1*	5	C309 -G1458	F2(13,000)	833-bp deletion	Ashikari, M. *et al.*,1999

Seed size	*D2*	1	9A – 3A(60-kb)	F2 (3000)	Nonsense mutation in d2-1; missense mutation in d2-2	Hong, Z. *et al.* (2003)
Seed size	*D11*	4	G7008 -L353	F2 (3020)	1-bp deletion in d11-1; 1-bp insertion in d11-2; missense mutation in d11-3; abnormal splicing in d11-4	Tanabe, S. *et al.* (2005)
Seed size	*D61(Os BRI1)*	1	C1370	F2	substitutions	Yamamuro Chizuko. e*t al.* (2000)
Grain width	GW2	2	W024-W004	BC3F2 (6013)	1-bp deletion	Song, X.J. *et al.* (2007)
Grain width	GW8/SPL16	8	RM502 and PSM736	F_2 (2,000)	10-bp deletion in the promoter	Wang, S. *et al.* (2012)
Seed size	SRS5	11	chr11-7240 and chr11-9030	F2(2000)	missense mutation	Segami*et al.* (2012)
Grain length	*GL3.1*	3	L012 and L008	(BC2F2 (179)		Peng Qi, 2012
Flag leaf shape and chlorophyll contenet	*LSCHL 4*	4	T2957–2 and RM348,	BC7F2 (1700)		Guang-Heng Zhang, 2014
IR64/ YP9 (IR68522-10-2-2) (7,996 BC4F3)	NAL1	4	Ind4 and Ind12	7,996 BC4F3	natural variation	Daisuke Fujita, 2013
d10-1 (d10-1/d10-1) and Kasalath	D10	1	RM10955-RM3411	(154 mutant F2)	point mutation	Tomotsugu Arite, 2007

d27-ZF802 mutant/ZF802 (5200 F2)	D-27	11	P3-P6		frame shift	Hao Lin, 2009

GN, Grain number; GW, Grain weight; GS, grain size; TN, Tillers number; PL, Panicle length; HI, Harvest index; GF, Grain filling.

6.Association mapping

Plant breeding and genetics rely on geneticmaps. These are made by analysis of segregating plant populations derived by crossingparents with contrasting characteristics. The hybridresulting from such a cross is then allowed toproduce offspring by selfing, and it is in thissegregating family that genetic variation, causingdifferent physical attributes (known as thephenotype), can be assessed. Regions of the plantchromosomes that contain important geneticinformation governing interesting and heritablephenotypic traits are identified by co-inheritancewith molecular DNA markers. Molecular markersare basically different length DNA fragments that areall inherited in a simple Mendelian manner. During the reproductive phase, recombination, orcross-over, occurs between the correspondingchromosomes from the two parents in the cross. Thisleads to reshuffling of the genes from each parent sothat the chromosomes in the offspring will consist ofmixtures, some parts of which are derived from oneparent, and some from the other. The closer twopositions on the chromosome (loci) are together, theless likely it is that a cross-over point is locatedbetween them. The closeness of the linkage betweentwo loci can therefore be measured in a segregatingfamily by observing the inheritance of molecularmarkers, which can then be ordered on the geneticmap. The association or linkage of particularmolecular marker variants (alleles) with an heritablephenotypic trait enables us to identify which regionof a particular chromosome is important for the phenotype. This is important for plant varietydevelopment because it means that plant breederscan use markers to test for the presence of importanttraits rather than testing for the traits themselves.This is called marker assisted selection (MAS).Genetic linkage between two loci (genes or molecular markers) on a genome is revealed by thefact that they are associated, or in LinkageDisequilibrium (LD). This is a genetic term meaningthat the two loci are very closely associated and islocated very close together on the chromosome

6.1 Population structure analysis

Pritchard *et al.* (2000) introduced the so-calledstructured association to reduce confounding due topopulation structure. The approach is based onassigning individuals to subpopulations by using amodel-based Bayesian clustering algorithm,STRUCTURE, and carrying out all analyses asconditional on the inferred assignments. TheSTRUCTURE algorithm is computationally intensiveand may be impractical on large datasets. Price *et al.* (2006) suggested that a principal components analysisbe used to summarize genome-wide patterns ofrelatedness. However, as the population is divided inmore subgroups, the probability of false positives isreduced at the cost of a reduction in statistical power.Moreover, any method that effectively removesconfounding also removes the true positives that arestrongly correlated with population structure (Zhao *et al.*, 2007). For instance, if the causal polymorphismsare perfectly correlated with the underlying populationstructure, distinguishing between true and falsepositives statistically is impossible, and any attempt toremove the latter will remove the former.The structured

association is effective but may notbe sufficient to control the confounding effects (Zhao *et al.*, 2007). Yu *et al.* (2005)introduced a mixed-modelapproach to control the population structure and the genetic relatedness among inbreds. Similar to othermixed-model-based methods, a random effect toestimate the fraction of the phenotypic variation,which can be explained by genome-wide correlations,is included by assuming that the phenotypiccovariance between individuals is proportional to theirrelative relatedness or kinship. Relative relatedness isestimated by using genome-wide marker data (the Kmatrix of pairwise kinship coefficients). In addition tothis random effect, a fixed effect by using thepopulation assignments produced by theSTRUCTURE algorithm (the Q matrix), was includedas a fixed effect in the model. The Q and K seem tocapture different features of the confoundingpopulation structure. However, Zhao *et al.* (2007)found that Q was not required in most cases if K wascomputed by using a method different from the oneused by Yu *et al.* (2005). AM has been proven to be an efficient method inrice using low- and high-density markers.

The bestexamples were presented by Huang *et al.* (2010) and Zhao *et al.* (2011). Huang *et al.* (2010) used whole-genome sequencing to identify singlenucleotide polymorphisms (SNPs) for association analysis, whereas Zhao *et al.* (2011) used Affymatrix chips with 44 100 SNPs. Huang *et al.* (2010) utilized517 rice landraces and about 3.6 million SNPs toanalyze marker-trait association for 14 agronomictraits. They identified a total of 37 significantassociation signals. Association signals for six traitswere located close to previously known genes, whichwere identified by using mutants or in studies ofrecombinant populations. They later reported ongenome-wide association studies of flowering timeand grain yield traits by using 950 worldwide varietiesand detected and identified 32 new loci responsible forflowering time and 10 grain-related traits (Huang *et al.*,2010). Zhao *et al.* (2011) applied association analysisto 413 diverse accessions of *O. sativa* from 82countries for 34 traits. They found SNPs associatedwith panicle length at 31.7 Mb to 31.7 Mb onchromosome 1 and for amylose content and floweringtime at 4.2 Mb to 4.6 Mb on chromosome 6.

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A case study on the effects of the MMRC Car-shed project on the Biodiversity of Aarey Forest

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Keywords:
Aarey Forest, Biodiversity, Environment, Activity

Abstract:
The construction of the MMRC car-shed project in Aarey Forest has the potential to significantly impact the biodiversity of the colony. Therefore, it is important to conduct a case study about the effects of the project on the biodiversity of the Aarey colony.

Aarey Colony is a lush green area located in the suburbs of Mumbai, India. It is home to a variety of plant and animal life and provides a vital ecological resource for the city. In 2014, the government proposed a car-shed project in the area as part of the Mumbai Metro Rail Corporation (MMRC) plan to expand its services. This project has sparked a debate over its potential effects on the biodiversity of the Aarey Forest. This case study will involve an analysis of the project's potential impacts on the flora, fauna, and other species in the area. Additionally, it will discuss the possible solutions that can be implemented to mitigate the negative effects of the project.

The MMRC car-shed project in Aarey Milk Colony has been a contentious issue for environmentalists and activists, who are concerned about the impact on the biodiversity of the region. This study will examine the effects of the MMRC car-shed project on the biodiversity of Aarey Forest, drawing on case studies and research from environmental experts and activists.

INTRODUCTION

As Prakash Bhoir, an environmental activist rightly said "We Adivasis together have created an oxygen factory for Mumbai." Aarey is indeed the lungs of Mumbai. Its serene beauty and calmness can make anyone's heart feel blissful. Spread around 3,000 acres, Aarey Milk Colony of Mumbai is home to 8,000 men, women, and children belonging to the tribal communities of Maharashtra. According to the data published, the aarey adivasis have been struggling for survival since the 1970s. Unfortunately, their struggle has soared since the announcement of the MMRC project. Not only is this project affecting the innocent residents of Aarey but also the rich biodiversity inhabiting it. Let us learn more about the Adivasis and biodiversity of aarey in depth.

Adivasi of aarey

Along with the everlasting beauty of aarey, the people of aarey have their charm. The aarey colony expands to an area of 3000 acres where 8,000 men, women, and children belonging to the tribal communities of Maharashtra reside. Various tribal communities are living in the lungs of Mumbai including Warli, Kokna, Mallar Koli, Katkari, and several other indigenous tribes.

Alt text: Warli tribe
Source: Dinodia Photo

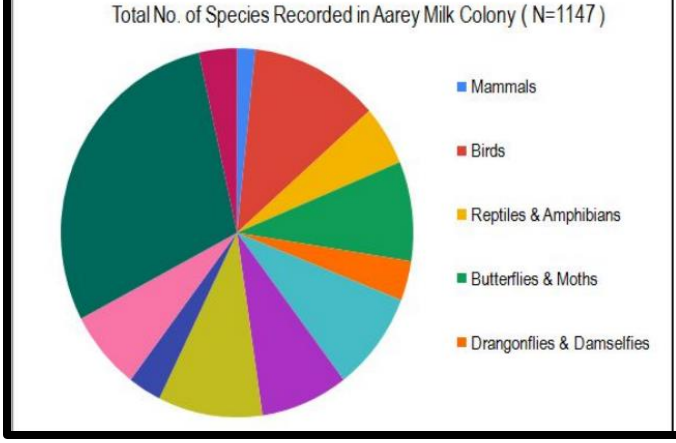
Alt text: Konkana tribe
Source: Wikipedia

Alt text: Mallar Koli tribe
Source: Shutterstock

Alt text: Katkari tribe
Source: India fellow

Aarey (Milk Colony) Forest Biodiversity -

The Aarey Milk Colony was founded in 1949. It is located in Goregaon East, Mumbai, Maharashtra, India, and is more than 3000 acres. It is located in the Sanjay Gandhi National Park's ecologically vulnerable area (SGNP). The Aarey Milk Colony is a mosaic of land uses, including adivasi padas, tabelas (cattle sheds), a milk factory, a hostel, a hospital, a field of crops, and the upper catchment zone (or watershed zone) for the Mithi and Oshiwara Rivers. Aarey is widely recognized for its 300 different animal species and its leopard population.
Throughout the research, 1147 species of Mammals, Birds Invertebrates in total were noted,



Taxon	No. of Species Recorded	Wildlife (Protected) Act Status	IUCN Status
Mammals	19	Sch 1, 2	VU, NT, EN
Birds	136	Sch 1, 2	VU, NT, EN
Reptiles & Amphibians	60	Sch 1, 2, 3	VU, EN
Butterflies & Moths	100	Sch 2	-
Drangonflies & Damselfies	40	-	-
Bug & Beetles	100	-	-
Ants & Other Insects (Bees, Wasps, Flies, Praying Mantis, Antlions, etc)	92	-	-
Spiders & Other Arachnids (Scorpions, mites, ticks)	110	-	-
Other Invertebrates (Crabs, millipedes, centipedes, etc)	35	-	-
Indigenous Flora (Trees &Palms)	80	-	-
Flora (Herbs, Shrubs, Climbers, Grasses, Orchids, Ferns)	335	-	-
Minor Flora (Bryophytes, lichens, Mosses) & Fungi	40	-	-
Total species Recorded	1147		

(Alt text: species information)
(Source: SPROUTS Education Trust)

Aarey is covered with a variety of tiny microhabitats. Zeeshan Mirza, a research associate at the National Centre for Biological Sciences, Bangalore, recently published information about Idiops rubrolimbatus (a trapdoor spider), Lychas aareyensis (a scorpion), and Heterophrictus aareyensis (tarantula) in a scholarly journal. There may be fewer of these species across the country. Aarey attracts a lot of migratory birds and dragonflies from the Himalayas to southern Europe.

The Tarantula (Haploclastus validus), was found after 110 years, the Trapdoor Spider (Idiops bombayensis), was also found after 110 years, and another tarantula (Pleasiophrictus millardi), which was rediscovered after 100 years, is just a few of the species that have been rediscovered in Aarey that were previously thought to have gone extinct.

Leopards of Aarey

There is an image of a large cat in the Waghoba temple in Mumbai's Aarey Milk Colony. The nearby tribal people worship an idol that resembles a tiger. In honor of the cats that live in the 13 square km Aarey Milk Colony with the villagers, a new leopard statue is scheduled to be inaugurated a short distance away in Aarey.

Alt text: Waghoba, an animal deity is worshipped by the tribal communities inside Aarey Milk Colony and Sanjay Gandhi National Park.
(Source- Kartik Chandramouli/Mongabay)

The Indian Wildlife (Protection) Act of 1972 lists the Indian leopard (Panthera pardus fusca) in Schedule I, giving it the highest level of legal protection. The leopard is still listed in Appendix 1 of CITES and is classified as near endangered by the IUCN (IUCN, 2014). The leopard population in Aarey Milk Colony (AMC) is small and often studied together with the leopards in the bordering Sanjay Gandhi National Park (SGNP) which has one of the highest densities of leopards in the world. But with Aarey now a proposed site for a metro project, the wildlife of AMC specifically, is in focus. A metro car shed for the Mumbai Metro Rail Corridor line, which runs between south and north Mumbai, from Colaba to SEEPZ (Special Electronics Export Processing Zone) has been selected as the green spot in the middle of urban Mumbai's western suburbs. No wildlife has been seen at the project site, according to the Environment Impact

Assessment (EIA) study from 2012 for the proposed metro project in Aarey. In contrast to the EIA report, In cooperation with the forest department, a team of scientists and wildlife enthusiasts has been carefully compiling data on wildlife, especially the elusive predators that call Aarey home. An average of four to five adult leopards can be found in Aarey at any given time, with a transient population that travels between Aarey and SGNP. Since this particular monitoring operation in Aarey started in 2015, four adult females, Adarsh Nagar, Bindu, Chandani, and Luna, have been consistently showing up in data recorded throughout various periods.

Alt text : Luna and a male
(Source- Ranjeet Jadhav)

Threats caused to the Biodiversity before the initiation of the Metro car-shed project:
Aarey and Film city is under severe threat from various anthropogenic activities. Some of the activities are listed below:

- Destruction of forest
- Burning of dried leaf litter kills many reptiles, spiders, and other life forms residing within the thick mat of leaf litter
- Soil erosion
- The proliferation of exotic flora
- Dumping of garbage in the water bodies
- Removal of soil for brick making
- Bootlegging
- Poaching of birds, turtles and monitoring lizards for consumption
- Speeding Vehicular traffic
- The proposed widening of roads
- Shifting of Zoo in Aarey

Metro Carshed project in Arrey-
The Aarey Milk Colony serves as both a center for biodiversity and a catchment area for the Mithi River, which runs through Mumbai. For this reason, they have sought that 165 Ha of forest land be denotified. The Maharashtra government is set about having its car shed here despite the protests of the public and the existence of seven other alternatives.

Environmentalists from various organizations, activists, students, and citizens have been fighting hard for more than three years to preserve the Aarey forest However, access to the area where

the metro car shed construction has already started has been denied to these environmental crusaders. Already, over a hundred trees have been removed. Mumbai's air quality index was 244 (classified as "poor") according to the System of Air Quality Weather Forecasting and Research (SAFAR), which is higher than Delhi's, which dropped from 204 to 183 (classified as "moderate"). In fact, Mumbai's PM2.5 levels, which were 109, were nearly double the acceptable limit. The Nature Conservancy's study shows how urban trees may remove up to a quarter of PM pollution within a few meters, and can even serve as a barrier for nearby households, filtering poor air. It is well known that urban greenery can assist in combating rising air pollution. Aarey Forest, which is a portion of Sanjay Gandhi National Park, sustains a unique environment with a wealth of species. Such an ecosystem cannot be quickly restored because it takes more years to develop.

When people learned about it, the protests started right away. This is because Aarey Milk Colony is a prized green area in the concrete jungle i.e, Mumbai. People have emerged to promote the message of conservation and stand against the reverse as a result of rising environmental consciousness. Additionally, it has just been learned that the removal of the trees could cause significant flooding at the international airport since the Mithi River will receive extra water during heavy rains. The list of negative effects we might experience as a result of giving up our greens is numerous.

Threat to biodiversity because of the Metro car shed project

Environmentalists cited a 2019 report to highlight the long-term effects which the proposed Mumbai Metro car shed 3 projects will have on the biodiversity of the Aarey forest in Mumbai. The assessment states that the damages will make the area's flood risks worse.
The research project, which is being conducted by the Kamla Raheja Vidyanidhi Institute for Architecture and Environmental Studies (KRIVA), uses qualitative-spatial analysis to show how current and future land use and cover may affect the watershed and ecosystems of the Aarey terrain. According to Shweta Wagh, an associate professor at KRVIA and urban conservator, the Aarey forest has shrunk from 1,300 ha to 800 ha over the past few decades as a result of changing land usage. The area was initially designated as a "No Development Zone" and is meant to stay that way. The final piece of forest connecting it to the Sanjay Gandhi National Park is where the metro car shed, rehabilitation homes, and zoo are proposed. It functions as a corridor for wildlife. However, constructing the infrastructure will completely cut it off from the national park and endanger the ecological landscape.

The area serves as a buffer between the SGNP and where leopards still wander at night. According to the Save Aarey movement, removing this buffer zone could result in undesirable interactions between people and wildlife. Experts are concerned that as a result of the ecological disruptions, the tribal people would be advised to relocate, which will negatively affect their way of life, wildlife habitats, and the amount of forest cover.

According to the report, Aarey Forest serves as a buffer for the national park, thus any harm to the forest region will affect the park both directly and indirectly. The city will be at risk since the national park will be directly exposed to its river systems and catchment area. To safeguard the national park from an ecological standpoint, Aarey was declared an ecologically sensitive zone

in 2016. Environmentalists worry that the ambiguity created by permits for infrastructure development under the heading "residential needs of local inhabitants" including "eco-tourism amenities" may lead to more forest clearing.

The future of Mumbai will be affected for a very long time by the decision to cut down these trees. Plants and trees provide humans with a safety net as pollution levels rise. The effects of cutting them down will be felt by both the current and upcoming generations that will reside in the city. According to a World Health Organization (WHO) report, air pollution may impact people's health severely. Another effect of deforestation is that there may be more conflicts between humans and animals. If the animals in the Aarey forest are deprived of their habitats, they will eventually move into human-populated regions, endangering the lives of inhabitants.

OBJECTIVES

- To study the effects of the project on forest biodiversity.
- To look at the current scenario of the local community.
- To study deforestation and its impact.
- To study the car shed project in detail.
- To look into the protests organized by the locals and NGOs.
- To study the outcomes of the protest if any.
- To propose solutions for the future project, if any.

P

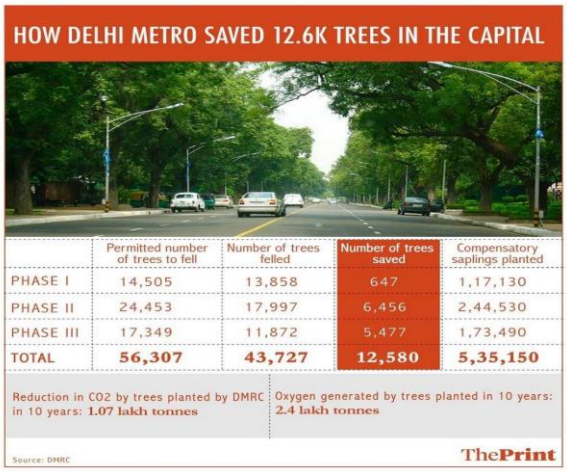
CONCLUSION

After considering everything, the aarey project indeed will have long-term effects on the environment and the city as well. A clear example of it is the air quality index result which was published by the aarey conservation group, depicting how the air quality was 276 PM2.5 on 31st December 2022, which was poor according to the standards. This doesn't look like a single project to harness the serene beauty of aarey but it is indeed an initiation to numerous projects in the

future. The wildlife will soon face major negative side effects and thus the lungs of Mumbai will lose their charm.

Well as a solution the government initially proposed a plan to shift it to kanjurmarg salt pan land but the financial loss the state would face because of relocating is a major issue. Not only will the relocating solution increase the cost but also will ultimately delay the project's completion.
Instead, the MMRC can implement the strategies DMRC i.e, the Delhi metro Rail corporation used**.** One of India's most effective suburban rail systems, the Delhi Metro connects areas on different ends of the national capital region along a network of 377 kilometers (NCR).
The Delhi Metro claims that the project was completed with careful planning that addresses numerous concerns associated with deforestation and other environmental aspects.
Measures taken by the DMRC are as follows:

- **Deforestation, reforestation.**
For every tree fallen the Metro authorities have to pay compensation to the Delhi Forest Department, which then carries out compensatory afforestation. A total of 10 saplings are planted for every tree cut said a senior official of the Delhi Metro Rail Corporation (DMRC), it is a joint venture of the Union territory administration and the central government. The Metro concessionaire planted over 5 lakh saplings to compensate for the 43727 trees cut for the three phases of the network i.e, 11 times. The trees planted, according to DMRC data, have sucked up 1.02 lakh tonnes of CO2 over the past 10 years and given out 1.07 lakh tonnes of oxygen. Another 35 lakh tonnes of carbon emissions were offset through the use of clean alternatives like solar power and reducing commuters' reliance on fossil fuels, the DMRC data stated. Officials of the DMRC environment section said they made a conscious effort to minimize environmental damage as they pushed forth with the Metro project.



For example, a depot at Khyber Pass, which caters to Yellow Line (between HUDA City Centre and Samaypur Badli), was built at a landfill site to avoid tree-felling. This required the DMRC to remove all the garbage from the site and lay it on good earth for the tracks to be laid. However, such measures could only be taken in areas where alignment was even minutely possible.

- **Green buildings**
A DMRC official said all the stations being built under the ongoing Phase III have been designed as "green buildings" with specific provisions for the conservation of energy and water, fewer emissions, and waste management. They have been equipped with more plants, water-efficient fixtures, and low-VOC paints, i.e, paint with fewer volatile organic compounds that are harmful to human health and the environment. Also when certain areas needed to be "dewatered" for the construction of underground Metro lines, the water was shared with the Chandrawal Water Works for the revival of lakes in North Delhi.
Such environmentally friendly measures taken by DMRC were quite beneficial in contributing towards the green environment and thus such initiatives should also be implemented by MMRC.

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Avian Diversity in and around Padmavati lake Dist.Jalna (Maharashtra)

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Abstract

Keywords:
Padmavati lake, avifauna, Passeriformes

Abstract:
In the present investigation avian diversity in and around Padmavati lake has been studied to know the current status of this reservoir. Padmavati Lake is one of the small water-body located in the area of vicinity of Bhokardan Tehsil situated in District Jalna of Maharashtra State. The water of reservoir is favorable for faunal diversity, rich in dissolved oxygen and supports variety of aquatic weeds and fishes, it has been found to be suitable habitat for bird fauna. A total of 33 species of birds spread over 20 families and 7 orders were documented in the lake. Maximum species of birds belonged to Passeriformes followed by Charadriiformes, Pelecaniformes, Ansriformes were recorded. The present study is carried only to prepare the checklist of birds as well as to find out occurrence and to create the awareness for their conservation of the biodiversity of avian fauna of Padmaavti lake.

Introduction

Diversity of avifauna is one of the most important ecological indicators to evaluate the quality of habitats. Now a day, avifaunal diversity has been decreasing due to the destruction of natural habitats and human disturbances. Birds are excellent model organisms for understanding key issues in ecology, animal behavior, evolutionary biology and conservation (Urfi, 2011). The migratory birds of India and abroad have created much interest in the minds of several workers to study their ecological niche. Although, Ali Salim (1987) and some other workers have studied the phenomenon of migration of birds, Similarly a little attention has been given to understand the ecological niche in relation to the migratory birds from this reservoir. Freshwater lakes one of the important types of wetlands, play a vital role in the economics of their respective regions, especially with reference to agriculture, fishing, livestock maintenance and drinking water facilities of the adjacent areas. The geographic location of a wetland may determine how and when birds will use it or use adjacent habitat (Manikannan, 2011). Water birds are commonly grouped into several categories based on their behaviors Gulls, Shore birds, wading birds, water fowl and terns. The birds are flying vertebrates and are of immense important to mankind. They are great useful to farmer as they kill and destroy the pests and help in controlling them. They also act as pollinator, agents for seed dispersion and scavengers.

The present investigation was undertaken for survey on the avian diversity in and around Padmavati lake for future initiatives in conservation. This reservoir locally called as Padmavati

dharan. The water of this lake is primarily used for washing, bathing, fishing activities, irrigation purposes and for other activities by the villagers. This lake harbors a number of aquatic weeds in the submerged as well as floating state on which thrive a large number of aquatic organisms. Apart from this, periphery is covered with bushes and trees which provide suitable habitat for the birds. Due to the abundant food available throughout the year in this area in the form of crustaceans, insects, molluscs, amhibians and even fruits which attracts a variety of birds. A total of 33 species of birds spread over 20 families and 7 orders were documented in and around the lake. Maximum species of birds belonged to Passeriformes followed by Charadriformes , Pelecaniformes, Anseriformes were recorded.

Material and Methods

The present work was carried out from June 2021 to May 2022. Different species of birds were observed with the help of binocular (Vanguard 10×50) and spot identification were done using field guide Salim Ali (2012), The Book of Indian Bird and also taken help of Ornithologist. The birds were observed during the morning (6 to 10 AM) and in the evening (4 to 7 PM).

Results and Discussion

During present investigation 33 species of birds belonging to 20 families and 7 order including land birds and water birds were recorded from Padmavati lake and surrounding areas. Out of 33 species 24 species were residents, 7 species were winter visitors and 2 species were summer visitors. (Table. 1). Similar work done by some earlier researchers like Kurhade (2010) reported 208 species of birds in Jaikwadi reservoirs near Ahmadnagar (MS), Narwade and Fartade (2011) recorded 165 species of birds of Osmanabad district (MS), Rasal and Chavan (2011) reported 61 species of birds in local ecosystem of Aurangabad (MS), Kukade et al. (2011) recorded 68 birds species of Chhatri lake of Amravati district (MS), Harney, et al, (2012) recorded 37 species of birds from Kanhala pond of Bhadrawati, District Chandrapur (MS), Joshi and Shrivastava (2012) reported 64 species of birds in Tawa reservoir of Hoshangabad district (MP).

Table 1.

Sr. No.	Order / Family	Scientific Name	Habit
1	Passeriformes Sturnidae	*Sturnus contra*	R
2	Passeriformes Passeridae	*Anthus rufulus*	R
3	Passeriformes Corvidae	*Corvus splendens*	R
4	Passeriformes Lanidae	*Lanius schach*	R
5	Passeriformes Sturnidae	*Acridotheres tristis*	R
6	Passeriformes Pycnonotidae	*Pycnonotus cafer*	R
7	Passeriformes Motacillidae	*Motacill maderaspatensis*	R
8	Passeriformes Sylvidae	*Chrysomma sinense*	R
9	Passeriformes Campephagidae	*Tephrodornis pondicerianus*	R
10	Passeriformes Muscicapidae	*Turdoides striat*	R
11	Passeriformes Muscicapidae	*Saxicolodies fulicatus*	R
12	Passeriformes Dicrudidae	*Dicrurus macrocercus*	R
13	Passeriformes Passeridae	*Anthus rufulus*	R
14	Charadriformes Charadridae	*Vanellus indicus*	R
15	Charadriformes Scolopacidae	*Actitis hypoleucos*	RM
16	Charadriformes Scolopacidae	*Philomachus pugnax*	R

17	Charadriformes Scolopacidae	*Tringa ochropus*	R
18	Charadriformes Scolopacidae	*Limosa limosa*	R
19	Charadriformes Scolopacidae	*Tringa nebularia*	R
20	Pelecaniformes Phalcrocoracidae	*Phalacrocorax fusicollis*	R
21	Pelecaniformes Phalacrocoracidae	*Phalacrocorax niger*	RM
22	PelecaniformesArdeidae	*Ardea alba*	R
23	Pelecaniformes Ardeidae	*Ardea purpurea*	R
24	Ansariformes Anatidae	*Anas poicillorhyncha*	R
25	Ansariformes Anatidae	*Netapus coromandalianus*	R
26	Ansariformes Anatidae	*Netta rufina*	WM
27	Ansariformes Anatidae	*Anas platyrhynchos*	R
28	Coraciformes coraciidae	*Coracias benghalensis*	WM
29	CoraciformesAlcedinidae	*Alcedo atthis*	R
30	Cuculiformes cuculidae	*Eudynamys scolopaceus*	R
31	Cuculiformes cuculidae	*Centropus sinensis*	R
32	Columbiformes Columbidae	*Spilopelia chinensis*	R
33	Columbiformes Columbidae	*Columba livia*	R

Chavhan and Dhamani (2014) recorded total 76 bird species was recorded in and around Chaprala wild life sanctuary, District- Gadchiroli, Maharashtra, India during December, 2011 to December, 2012. Out of 76 species 90% were common, 09% uncommon and 01% were migratory. Accipitridae was the dominated family of birds with maximum number of species was Accipitridae with 11 species (14%) followed by Corvidae represented by 06 species (08%), Passeridae 05 species (07%), Campephagidae 04 species (05%), Alcedinidae, Columbidae, Muscicapidae, Phasianidae, Strigidae and Sturnidae with each 03 species as well as Ciconiidae, Meropidae, Psittacidae with each 02 species (03%), and 26 other families representing least number of species with 1%. Dapke et al., (2015) observed diversity and seasonal abundance of avifauna with vegetation, composition of habitat and foraging pattern in and around Laxminarayan Institute of Technology (L.I.T.) campus, Nagpur, Central India. They recorded 62 species of birds belonging to 11 orders and 38 families during January, 2013 to December, 2014. Out of which 57 were residents, two passage migrants, two winter migrants and one was breeding migrant. Passeriformes most dominating order represented by 36 species. Similar results were observed during present investigation out of 33 species observed during study period, 13 species represented by order Passeriformes, 6 species represented by Charadriiformes, each 4 species were of Pele-caniformes, and Anseriformes and 2 species represented by each order Coraciformes, Cuculiformes and Columbiformes.

Conclusion

Although Padmavati lake is small water-body the site is suitable habitat for the residential and migratory birds. But the birds present in and around the study site are affected by anthropogenic disturbances like washing clothes, direct bathing, washing livestocks, immersing of idols, fishing practices and pollution due to spraying of insecticides on the crops in catchment area. There is a need for execution of necessary precautions by the concerned authorities in order to prohibit destructive activities and conserve the avifaunal diversity in these lakes. Keeping in view the varied avifauna recorded, steps should be taken to do proper maintenance and beautification of the lakes.

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HEMATOLOGICAL CHANGES INDUCED BY FLUORIDE IN FRESHWATER FISH, *CYPRINUS CARPIO*

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Keywords:
Hematological parameters, *Cyprinus carpio,* fluoride, Lethal & Sub lethal concentration

Abstract:
The fish, *Cyprinus carpio* exposed in lethal and sub lethal concentrations of fluoride at varying time interval and effects on various hematological parameters were evaluated. WBC was decreased significantly in both concentrations. Significantly increased in the percentage of HB% at 96h in lethal & sub lethal, there was significantly decreased RBC level in lethal concentrations. While it was significantly increased RBC level at 48h. In lethal, HCT levels were decreased in all the durations. In sub lethal, HCT levels were insignificantly increased in 96h. In the lethal MCV was significantly decreased in all the duration. In sub lethal, MCV was found highly increased insignificantly. In lethal MCH was highly increased insignificantly in all the duration. While in sub lethal MCH was insignificantly increased in 48h & 72h. In the lethal & sub lethal concentrations of fluoride significantly increased in the MCHC levels. The observed changes suggest that fluoride affects on hematological parameters of fish, *Cyprinus carpio.*

Introduction:

Fluorine is the thirteenth most abundant element making up approximately 0.06 to 0.09% of the earth's crust (Environment Canada 1976; Smith, 1983). The main natural source of inorganic fluorides in soil is the parent rock (WHO, 1984). At high doses, fluoride interferes with carbohydrate, lipid, protein, vitamin, enzyme and mineral metabolism (Anon, 1970). High fluoride levels cause cell damage and necrosis which affect organ function. Detailed changes which occur in the bone of animals severely affected with fluorosis have been described by Roholm (1937 and Shupe *et al.,* 1955). Fluoride affects the hematological parameters (Saxena *et al.* 2001), brings about morphological and behavioral changes (Tripathi *et al.* 2004) and affects the cellular architecture in vertebrates (Gupta, 2003). The observation of different haematological parameters provides a good deal in diagnosing the effect of fluoride on fish and in fact would give an insight into changes induced in the circulating fluids. Fluoride damages erythrocytes and induces echinocyte formation (Jain & Susheela, 1986). These damaged erythrocytes are eliminated through the process of phagocytosis, this shown that fluoride decreases RBC & hemoglobin (Pillai & Mane 1985). *Cyprinus carpio* fish is an economical important fresh water fish and commonly cultured in many parts of the world. In the present study, an attempt was made to investigate the toxic effects of fluoride on haematological parameters exposed in lethal and sub lethal concentrations of fluoride at different time interval.

Material and Methods:

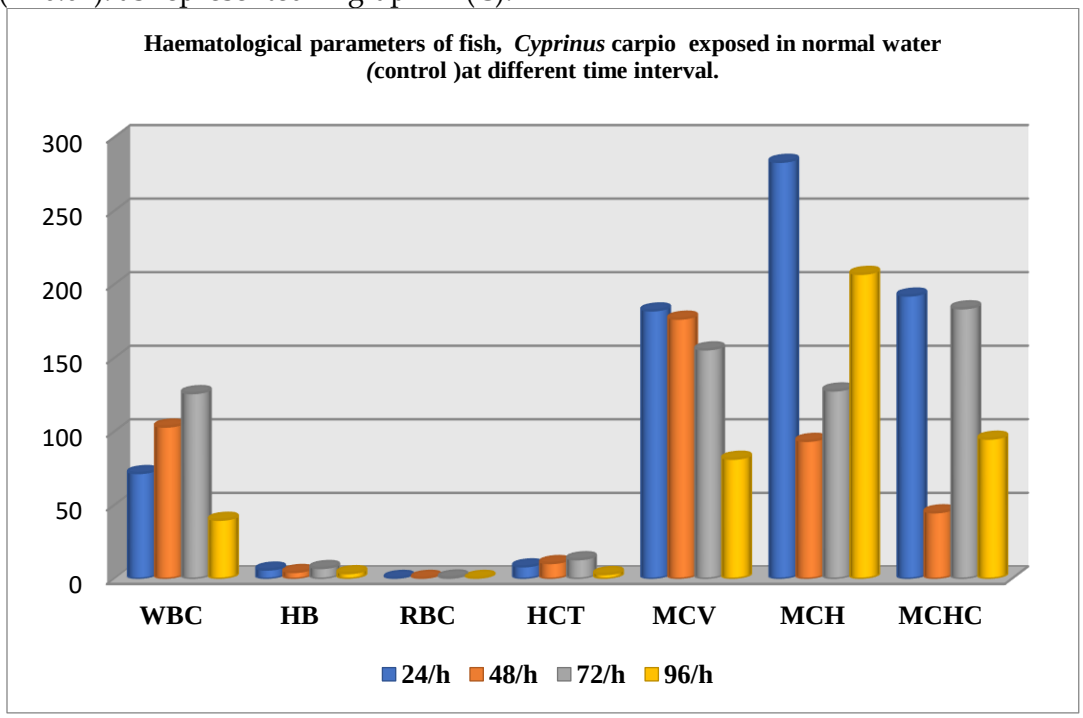
The fluoride waters obtained from nearby areas and were tested in public health Lab. The report indicates that, the bore water (F) contained 4.285mg/lit. *Cyprinus carpio*, ranging between weighing about 250gm.were collected from a nursery pond at Sawargaon in Umri Tahsil of Nanded district. The animals were brought to the laboratory and were acclimatized to lab condition for four days where they fade with rice and groundnut cake. Fish was exposed in lethal & sub lethal con. of fluoride at different time interval. One group served as a control, LC50 was recorded at 96h.

Collection of Blood: - The blood was collected by direct heart puncturing using sterile disposable plastic syringe with a 22-gauge needle (Molnar, 1960). The blood sample was taken in a tube rinsed with EDTA as an anticoagulant, & was mixed gently by rotation and stored in a refrigerator at 4^{o}C. Preserved blood sample was used for hematological studies. Hematological parameters were determined by fully automated Cell Counter (Trivitron Selenium Jr.) & compared with control.

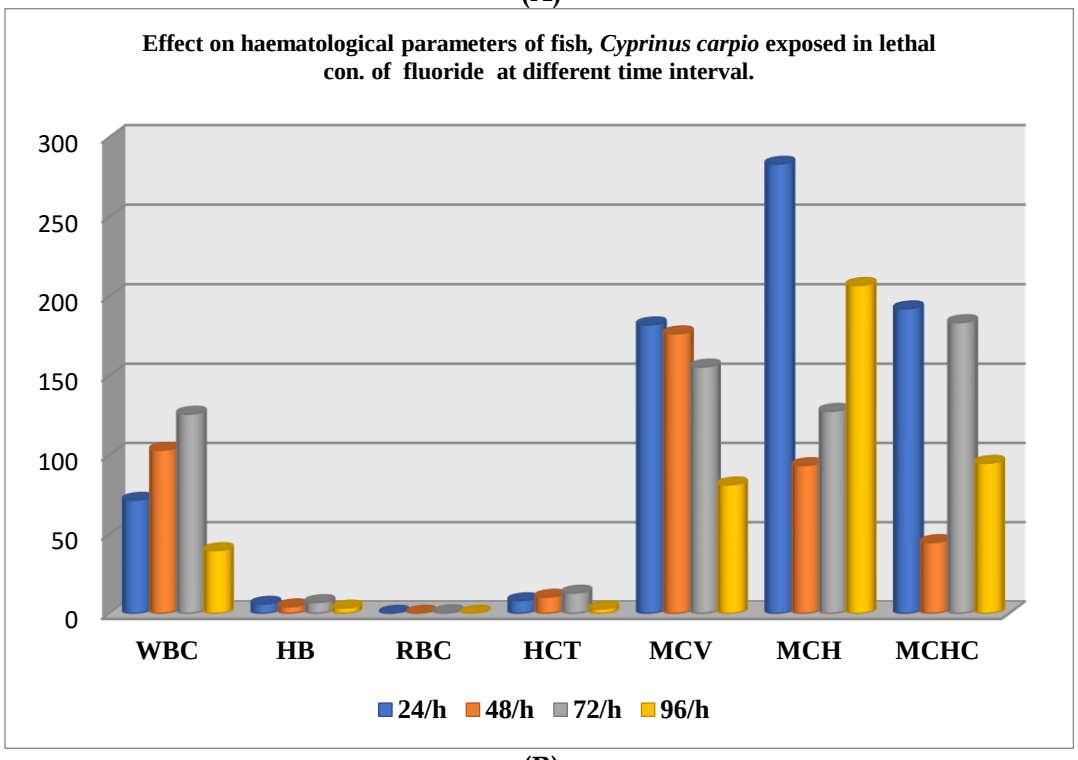
Results: In the present study, in control, WBC count was highest in 72h & 48h followed by 24h & 96h. The HB was highest in 72h, followed by 24h & in 48h. The lowest was in 96h. The RBC count was highest in 72h followed by 48h 96h & in 24h. The hematocrit HCT percentage were highest in the 72h followed by 48h 24h & lowest was in 96h. The MCV was highest in 24h followed by 48h & 72h. Lowest was in 96h. The MCH in control was highest in 24h followed by 96h, 72h, lowest was in 48h. MCHC was highest in 24h followed by 72h and 96h & lowest were in 48h. as represented in graph (A) when, fish, *Cyprinus carpio* was exposed in lethal as well as sub lethal concentrations of fluoride, significant changes were observed on the haematological parameters. WBCs was significantly ($P>0.05$, $P<0.05$) decreased in the 24h, 48h & 72h. In 96h it was highly insignificantly decreased. In 24h, 48 &72h it was significantly ($P<0.05$) decreased. In 96h WBC was insignificantly decreased. The HB was increased insignificantly in 96h. The insignificantly decreased HB was in 24h & 48h. While significantly ($P>0.05$) decreased in 72h. The RBCs was decreased in all the duration of exposure. Significant ($P<0.05$) decreased was observed in 48h. In the 24h RBCs was decreased significantly ($P>0.05$). In 72h decreased insignificantly. In 96h it was insignificant decreased. The HCT, decreased in all duration & significantly ($P>0.05$) decreased in the 48h & 72h. In 48h, it was decreased insignificantly. The MCV was decreased all the duration. MCV was in 24h and 72h, was found decreased significantly ($P>0.05$). In 48h & 96h was decreased insignificantly. The MCH was highly elevated significantly during 48h. In 24h it was decreased significantly ($P>0.05$). In 72h & 96h decreased insignificantly. The MCHC increased in all duration. & highly elevated significantly ($P>0.05$) in the 48h, & in 72h increased significantly ($P>0.05$). In 96h increased insignificantly. In 24h, it was found significant equal ($P=0.05$). As represented in graph (B).

In Sub lethal, WBCs was significantly decreased in all the duration. In 96h it was insignificantly decreased. In 24h it was significantly decreased ($P=0.05$).In 48h &72h it was significantly ($P>0.05$) decreased. In 96h, it was insignificantly decreased. The HB was increased insignificantly in 96h.while insignificantly decreased was in 24h, 48h. It was significantly ($P>0.01$) decreased in72h. The RBC was increased insignificantly in 48h & in 24h it was decreased insignificantly. In 72h, it was decreased significantly ($P>0.05$). In24h & 96h, it was insignificantly decreased. The HCT was increased insignificantly. In the 48h & 72h, it was significantly ($P>0.05$) decreased. The MCV was increased insignificantly in 96h. MCV in 24h & 48h significantly ($P>0.05$) decreased. In 48h, & 96h, it was decreased insignificantly. The MCH was observed elevated in 48h & decreased insignificantly. In 24h it was decreased insignificantly. Followed by 72h it was decreased

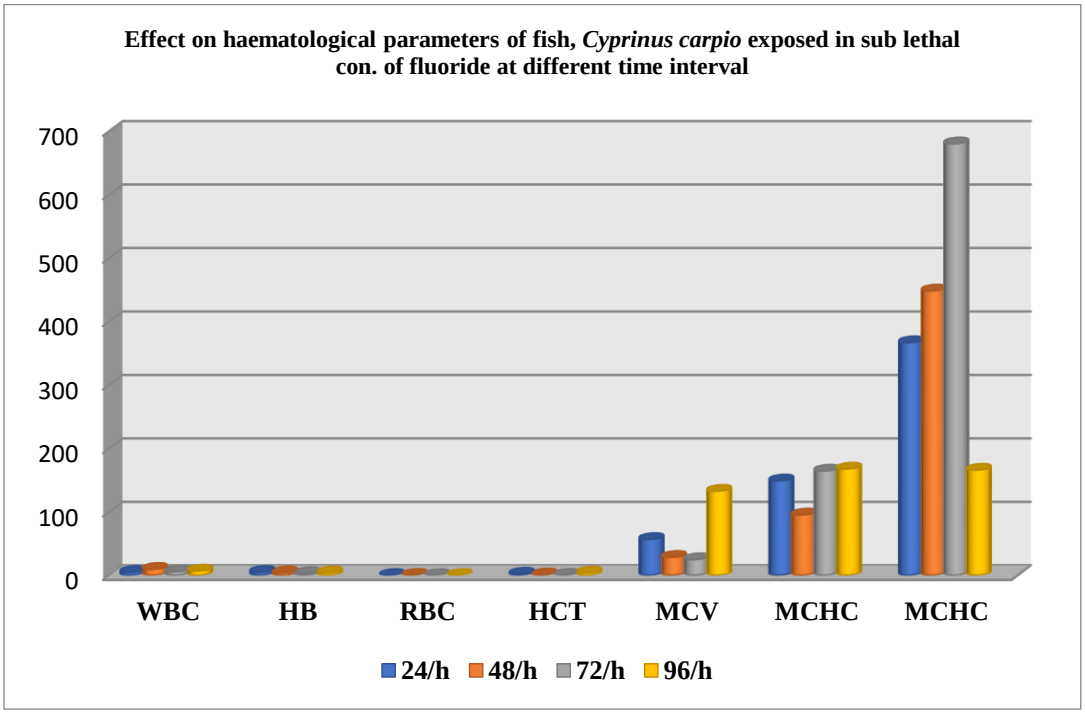
significantly (P>0.05). In 72h & 96h, it was decreased insignificantly. The MCHC was increased in all the duration. It was highly elevated significantly (P>0.05) in 72h & 48h. In 96h, it was increased significantly (P>0.05) .In 24h, it was increased insignificantly. In 48h, it was significant equal (P=0.01). as represented in graph in (C).



(A)



(B)



(C)

Graph:

(A)-Showing normal haematological parameters, (B) & (C) Changes observed in haematological parameters of fish, *Cyprinus carpio* exposed in lethal & Sub lethal concentrations of fluoride at various time intervals.

Discussion:

During the present investigation considerable effect on haematological parameters when exposed to the toxicant, fluoride are observed as per the data pertaining to % Hemoglobin (HB), White Blood Cells (WBC), Red Blood Corpuscles (RBC), Hematocrit (HCT), Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin (MCH) & Mean Corpuscular Hemoglobin Concentration (MCHC), revels changes which influence the haematological parameters of the fish. Hlynezak and Ubranksa (1987) reported a significant decrease in blood hemoglobin and haematocrit of Catfish, Cows & rats exposed to fluoride. The WBC, RBC count & Haemoglobin decreased in both the concentration of fluoride in all treated groups. Similar results were also reported by Mishra & Mohapatra 1987; Kristinson *et al.,* 1997; Jagdish *et al.,* 1998; Mandal *et al.,* 1986; Shanthakumari & Subramaniam 2007; Banupriya *et al.,* 1997, Sushella *et al.*, 2001, Kahl *et. Al.,* 1973 and Pillai *et al.,* 1985) reported the inhibition of RBC on fluoride exposure. The WBC count was significantly reduced in both fluoride concentrations similarly in another study WBC count change by fluoride exposure to 10pp for four month rats by Eren *et al,* (2005).

In this investigation of RBC, PCV, HB %, MCH, MCHC was found decreased. Similar reports were made by Ramanujaam & Mohanty (1977) also reported decreases in RBC, PCV, HB %, MCH, MCHC due to thiodon. Bhatt and Farswan (1992) observed that RBC, WBC, HB%, PCV decreased while ESR increased, due to plant toxicant *Barilius bendalensis* (Hem). In the present investigation we found significant increase in MCHC, WBC cell count. Similar observation was made by Nath and Banerjee (1995) observed a significant increase in MCHC, WBC and ESR in *Heteropheustes fossils* due to toxicity of dimithion. A significant decreased in the Hb%, RBC and PCV% was noticed on exposure of *Catla catla* to Malathion and Dichlorofos (Srinivas *et al.,* 2001). The MCV

increased in sub lethal dose of fluoride in 96h while it was remained elevated until the end of the experiment. Similar results were obtained by Kurovskaya and Osadchaya (1993), who did not report anemia in common carp infested with *Ichthyophthirius multifiliis.* The increase in MCV, MCH in lethal & sub lethal exposure of fluoride elevated in 48h and altered or decreased MCHC was evident in the present investigation. This data confirmed that anemia produced was of macrocytic hypochromatic type. The MCHC is a good indicator of red blood cells swelling and a decrease in haemoglobin synthesis (Wepener *et al.,* 1992a, b; Bhagwant and Bhikajee, 2000). Ololade and Oigni (2009) observed a decrease in MCHC with increased concentration of toxicants (zinc). Martins *et al.* (2004) and Sabri *et al.* (2009) also recorded a significant decrease in RBC, PVC, and HB in parasitized specimens of *Leporinus macrocephalus* and *Clarias garipienus,* respectively.

Conclusion: From the above discussion, it can be concluded that fluoride is more toxic to fish. Exposure to both lethal & sub lethal concentration of fluoride resulted in significant alteration in haematological indices. Thus fluoride affects not only aquatic animals but also terrestrial animals too.

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Heavy Metals in Godavari River Water Samples: An Analytical Study

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Keywords:
Heavy Metals, Metal Contamination, Water Analysis.

Abstract:
Out of the 105 total elements discovered so far, the number of heavy metals in the Godavari River is close to 65. The concentrations of metals vary from location to location. Compared to marine water resources, fresh water resources are scarce. These fresh water sources are becoming more and more contaminated as a result of several natural disasters and human activities, and the amount of heavy metals in them is rising steadily. It is currently the top worldwide concern for both the human race and the underwater environment. These sources mostly contain Cd, Cr, Pb, Ni, and Zn, among other elements. The sequence of the heavy metal concentrations in the sample water taken from the Godavari River is Ni>Pb>Cd>Zn>Cr. Controlling these natural and artificial activities that raise the content of heavy metals in river waters is now very necessary.

INTRODUCTION

Water resources are becoming more and more contaminated as a result of extensive human activity. Our drinking water supplies are mostly impacted by pollution and high metal levels. Water resources include a variety of dangerous contaminants, many of which are pharmacologically active and some of which are either carcinogenic or mutagenic [1, 2]. The Bureau of Indian Standards' acceptable limits for one or more heavy metals were exceeded in samples gathered from two-thirds of the water quality sites spanning India's main rivers [3]. The results are included in a report, the third iteration of an exercise the Central Water Commission (CWC) carried out between May 2014 and April 2018. In some cases, it is impossible to completely eliminate metals in drinking water, however some metals are necessary for health in tiny levels. They are connected to a number of illnesses though when present over safe levels [4, 5]. Everyone is concerned about the problem of the ever-growing threat of water pollution because of contemporary technology, industry, and civilization. The problem of water contamination by trace metals is now widely acknowledged to be vital all over the world, especially in a developing country like India. Very hazardous chemicals are present in industrial effluents that contribute to aquatic pollution [6, 7].

Liquid waste of both organic and inorganic form that is released indiscriminately alters the physio-chemical properties of water and poses a threat to both aquatic ecosystems and critical members of the human food chain. As a result, the current study aims to analyse some of the significant heavy metal concentrations of the Godavari river water, including those of iron (Fe), copper (Cu), chromium (Cr), lead (Pb), cadmium (Cd), zinc (Zn), and fluoride (F).

MATERIAL AND METHODS-

At various targeted Godavari river bank locales, including Paithan, Kaigaon, Gangakhad, and Kopargaon, samples were gathered during various seasons. The right scientific techniques are used to collect samples, which are put in plastic jars that have been cleansed with nitric acid and distilled water prior to sampling. For the analytical study of heavy metals present in the collected water samples, such as iron (Fe), copper (Cu), chromium (Cr), lead (Pb), cadmium (Cd), zinc (Zn), and fluoride (F), the Atomic Adsorption Spectrophotometer (AAS) technique is utilised. It has been identified in concentrations ranging from trace to large amounts.

RESULTS AND DISCUSSION-

The findings of the study's analysis of river water in ppb units are shown in [Table 1 and Figure 1].

Table 1: Changes in Heavy Metals (ppb) in Godavari River Water for the Years 2019–20 (summer and winter)

Station No.	Cu	Fe	Cr	Pb	Cd	Zn	F-
N1	0.486	0.684	2.42	12.50	2.233	1.897	0.345
N2	0.498	0.654	2.33	12.80	2.189	1.756	0.428
N3	0.503	0.643	2.48	12.95	2.323	1.824	0.378
N4	0.516	0.656	2.16	14.16	2.425	1.958	0.453
N5	0.505	0.647	2.35	13.80	1.981	2.210	0.586
N6	0.528	0.659	2.50	12.96	2.351	2.198	0.489
N7	0.508	0.672	2.80	16.55	1.994	1.996	0.721
N8	0.520	0.685	2.95	16.43	1.898	1.975	0.793
N9	0.534	0.698	2.16	15.66	2.356	2.021	0.736
N10	0.511	0.708	2.80	17.58	2.410	2.105	0.754
N11	0.526	0.696	2.96	16.98	2.322	2.112	0.738
N12	0.587	0.688	3.12	16.77	2.408	2.312	0.784

*All values are in ppb (parts per billion).

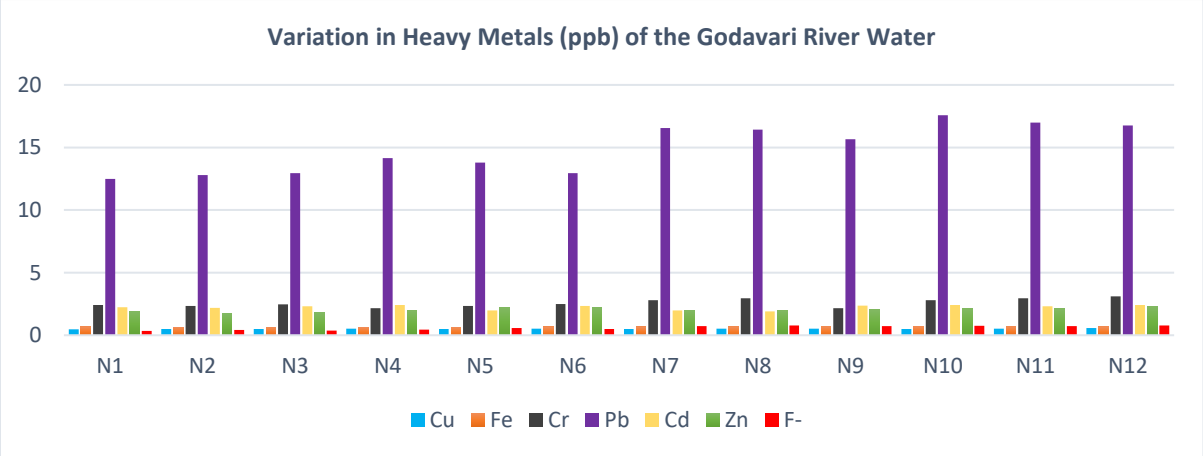

Figure 1 : Variation in Heavy Metals (ppb) of the Godavari River Water

Copper (Cu) -

The range of copper content in ppb level during the summer and winter of 2019–20 was 0.486 (N1) to 0.587. (N2). High levels of copper can have negative effects such as nasal, oral, and ocular irritation, vomiting, diarrhea, and gastrointestinal lesions (GIT). The victims of the aforementioned illnesses were noted at the primary health facilities in the research region during the monsoon season.

Iron (Fe) -

The range of the iron content in ppb level during the summer and winter of 2019–20 was 0.643 (N3) to 0.698. (N9). High iron levels typically provide bitter, astringent, and inky tastes. Additionally, it can taint clothing, plumbing fixtures, and produce scaling that encrusted pipes.

Chromium (Cr) -

Chromium levels in ppb levels throughout the summer and winter of 2019–20 ranged from 2.16 (N4) to 3.12. (N12). The electroplating, metal polishing, and publicly operated treatment plants are the main sources of chromium. Iron and steel foundries, inorganic chemical facilities, tanneries, textile manufacturing facilities, runoff from urban and residential areas are very small contributors (apart from localized pollution).

Lead (Pb) -

The variance in ppb level of lead metal for the year 2019-20 (summer and winter) ranged from 12.50 (N1) to 16.98. (S11). The lead content was raised, releasing free metal ions into the water bodies from cooking utensils and increasing the solubility of old paintwork from buildings during acidic wet deposition.

Cadmium (Cd) -

Cadmium metal levels fluctuated from 1.898 (N8) to 2.425 ppb over the summer and winter of the 2019–20 school year (N4). The outflow of domestic wastewater from residential areas, the impulsive use of pesticides, the use of fertilizers in the palm oil estates that line the banks of rivers, and local air pollution from open burning are all potential sources of cadmium in river water systems.

Zinc (Zn) -

Zinc levels in ppb throughout the summer and winter of 2019–20 ranged from 1.756 (N2) to 2.312 (N12). The amount of zinc was greater in the summer. Since the river's water volume was significantly reduced throughout the summer, it is likely that human activity—such as agricultural runoff, residential activity, wastewater discharges, effluent discharges, and other non-point sources—is to blame for the rise in heavy metal content.

Fluoride (F-) -

Fluoride in ppb levels fluctuated from 0.345 (N1) to 0784 over the 2019-20 school year (summer and winter) (N12). Because fluoride is naturally found in water, it becomes hazardous to animals and humans when it exceeds 1.0 mg/l in drinking water. Molting of teeth and bones has been recorded at levels of 1.5 mg/l.

CONCLUSIONS-

Since rivers have such great ecological, cultural, and touristic importance, protecting them is in everyone's best interest. Understanding the quantity of hazardous substances (heavy metals) entering rivers and their biological magnifying effects on animals, especially those at the base of the food chain, would be made easier with the aid of this study. Additionally, this study will aid in educating nearby farmers or residents about good waste management practices and how to reduce the usage of synthetic inputs. Scientists, decision-makers, administrators, and everyone else concerned in environmental conservation face a problem as a result of the study's findings that the daily accumulation of hazardous waste in rivers has led to biological amplification in the food chain.

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Studies On Water Quality And Aquatic Insects From Bori Reservior, Naldurg Dist- Osmanabad, Maharshtra

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Diversity of Aquatic insects from Bori reservoir

Abstract:
The present investigation deals with the studies on water quality parameters and Diversity of Aquatic insects from Bori reservoir, Naldurg, Dist-Osmanabad, Maharashtra. The work was carried out during the year 2021 (January to December) the abiotic parameters were studied from month to month. The diversity of water insects was studied. The species were found during the study period.

INTRODUCTION

The quality of water play a vital role in the growth of aquatic animals and their population. The water quality standards below and above the optimum level may lead to stress or death among the aquatic animals. The quality of water may be changed due to pollution of water body. The quality of water depends on physical, chemical and microbiological factors.

Aquatic insects are important group of organisms in freshwater bodies and plays an imp role in the processing and cycling of nut nets. According to Larrnberti Moore (1948), Aquatic insects belongs to several feeding groups i.e. filter feeders, deposit collectors scrapers, shredders and Predators. Aquatic insectors are are bioindicators of water pollution Wieder Holm, 1948 : Metcaffe (1989). These insects form a link between the nutritional cycles of aquatic ecosystem. The workers like Sinha and Sinha, Kaushik et al (1990), Pandey et al (1992), Singh (1993). Arvind Kumar (1994), works on seasonal water quality of fresh water bodies.

Due to lack of information about the water and Aquatic insects interrelationship the work was undertaken.

MATERIALS AND METHODS

The Aquatic insects were collected by insect collecting net made up of nylon cloth having mesh size 40-80 cm^2. The samples were cleaned and preserved in 5 percent formalin. The identification of insects was done with the help of standard literature of Tonapi (1959), Michael (1973), Macan (1959).

Monthly collection of water samples was done by using plastic containers of 5 lit. size. The samples were brought to laboratory for analysis. The metrological parameter i.e. air temp. and humidity were determined in the field. The samples were analysed by using standard methods of water analysis given by Trivedy and Goel (1984), and APHA, AWWA, WPCF (1985) and Kondarkar M.S. 1995.

RESULTS AND DISCUSSIONS

The physico – Chemical parameters shows in the Table No. I and Aquatic insects in Table No. II.

Table No. II
List of Aquatic Insects

Sr. No	Name of the Aquatic (Insect)	
	Common Name	**Scientific Name**
1	Back Swimmer	Anisop
2	Dragon fly nymph	-
3	Water Stick Insect	Ranatra
4	Water Boatman	Corixa
5	Olive bettle	Cybister
6	Water bug	Water Scorpion

The air temp, shows lowest range in the month of December and highest in the month May. The humidity ranges between highest in the month of Feb and lowest in the month of Jan. The water temp. lowest recorded in the month of Dec 25 ^{0}C and highest in the month of June 35.1 ^{0}C, pH rangest between 7.1 to 8.3, DO recorded highest in the month of February and lowest in the month of April.

The free CO_2 recorded highest in month of March and lowest in Sept. the alkalinity ranges between 151 (Sept) to 266 (Aug). The Hardness shows higher in July and lower in Nov. the Chloride recorded higher in April and lower in Sept.

The mg shows range between 5.20 to 94.1 the phosphate recorded higher in June and lowest in Aug.

Near about seven species of Aquatic insects were found during the period of investigation i.e. Back swimmers, Dragon fly nymph, water stick insect, water boatman, olive beetle, water bug etc. these 3 are the harmful to the fishes.

ACKNOWLEDGEMENT

Authors are thankful to the Principal and Institution for providing necessary library and Laboratory facilities.

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Table No. I.
Physico – Chemical Parameter Shows in the Table No. I.
(January to December 2021)

Months / Parameter	Jan.	Feb.	March	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
Air Temp.	30	30	35	42	46	43	40	39	35	39	29	28
Water Temp.	27.2	28.1	31.2	34.3	34.4	35.1	26.2	27.1	28.3	27.0	26.1	25.0
Humidity	31	79	67	55	62	72	73	62	52	45	45	50
pH	7.6	7.5	7.1	7.4	7.4	7.3	7.8	7.9	8.0	8.2	8.0	8.3
D. O.	7.43	8.60	5.80	4.0	4.5	3.1	2.41	3.83	4.61	7.04	6.63	7.60
Free CO_2	AB	AB	31.0	19.2	12.3	14.2	15.1	17.1	11.0	AB	AB	AB
Alkalinity	245	196	200	156	220	222	221	266	151	196	220	221
Hardness	141	185	115	201	215	200	216	200	125	120	111	135
Chlorides	42.4	32.61	34.00	137.70	42.5	41.1	42.4	53.90	32.60	42.6	48.20	39.70
Magnesium	35.61	14.51	10.50	77.80	90.60	91.09	93.60	94.01	13.15	35.61	5.20	35.56
Phosphate	0.5	0.4	2.1	2.2	2.0	2.3	0.08	0.02	1.1	0.2	0.03	1.0

Preliminary phytochemical screening of five medicinal plants used in traditional medicine

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Keywords:
Medicinal plants, Phytochemical Screening, Bark Extract.

Abstract:
Plants have served human beings as a natural source for treatments and therapies from ancient times, amongst them medicinal herbs have gain attention because of its wide use and less side effects. In the recent years plant research has increased throughout the world and a huge amount of evidences have been collected to show immense potential of medicinal plants used in various traditional systems, thus in the present investigation the phytochemical analysis of five different medicinal plants plants i.e., *Neolamarckia cadamba* (Roxb.) Bosser., *Bombax ceiba* L., *Boswellia serrata* Roxb. ex Coleb., *Ceiba pentandra* (L.) Gaertn. and *Cordia dichotoma* G.Forst carried out as these plants have been proved to be one of the important medicine to treatment for human beings.

INTRODUCTION:

Medicinal plants play a major role in meeting the medical and health needs of about 80% of populations in developed and developing countries, which serve as an important resource for the treatment of various maladies and illnesses (Ngari, et al. 2010) (Florence, et al. 2015). In developing countries, there is an increasing attempt to incorporate the traditional medicines, especially herbal preparations in the local healthcare systems and modernized people are increasingly turning to herbal medicine (Njoroge and Bussmann 2007) (Florence, et al. 2015). Globally, about 85% of the traditional medicines used by different ethnic groups inhabiting various terrains for primary healthcare are derived from plants, especially in India; medicinal plants are widely used by all sections of the population with an estimated 7500 species of plants used by several ethnic communities (Farnsworth 1988) (Farnsworth 1988). There is ample literature on preliminary phytochemical surveys (Sazada, et al. 2009) (Mojab, et al. 2002) (N. Farnsworth 1966) (Placeholder1) (Venkata S, Nagendra and V 2010) and the knowledge of the chemical constituents of plants is desirable to understand herbal drugs and their preparations (Venkata S, Nagendra and V 2010).Phytoconstituents are the natural bioactive compounds found in plants. These phytoconstituents work with nutrients and fibers to form an integrated part of defence system against various diseases and stress conditions. Phytochemicals are basically divided into two groups, i.e. primary and secondary constituents; according to their functions in plant metabolism. Primary constituents comprises common sugars, amino acid, proteins and

chlorophyll while secondary constituents consists of alkaloids, terpenoids, steroids and flavonoids, so on (Dhawale 2013).

The present study has been performed on some medicinal plants, *Neolamarckia cadamba* is large tree with a broad umbrella-shaped crown and straight cylindrical bole. The branches are characteristically arranged in tiers. The tree may reach a height of 45 m with a stem diameter of 100–160 cm and sometimes it has a small buttress up to 2 m high. The bark is grey, smooth and very light in young trees, but rough and longitudinally fissured in old trees. The branches spread horizontally and drop at the tip. The leaves are glossy green, opposite, simple sessile to petiolate, ovate to elliptical (15–50 cm long by 8–25 cm wide). In young fertilised trees, the leaves are much larger, subordinate at base and acuminate at apex; the stipules are interpetiolar, narrowly triangular and deciduous. The fruitlets are numerous, somewhat fleshy, with their upper parts containing 4 hollow or solid structures. The fruit occurs in small, fleshy capsules packed closely together to form a fleshy yellow-orange infructescence containing approximately 8000 seeds. The seeds somewhat are trigonal or irregular shaped, not winged (Kanninen 2011).

Bombax ceiba is deciduous trees 10-30 m tall; bark grey, glabrous; prickles conical, black. Leaves crowded at the ends of branches, digitate; leaflets 5-7, ovate-lanceolate or elliptic-lanceolate, 5-23 x 1.5-9 cm, glabrous; petioles 12-20 cm long; petiolules 1-2.5 cm long; stipules small, triangular, caducous. Flowers appearing before the new leaves, 8-14 cm across, sessile, crowded at the ends of branches. Calyx often 3-lobed, 1.5-2 cm long; lobes roundee, glabrous outside, densely silky within. Corolla bright red, tomentose outside; petals elliptic-oblong, 5-7 cm long. Sta¬mens about half as long as the corolla; filaments flattened. Ovary conical, glabrous; style longer than the stamens; stigmas 5, linear. Capsules woody, ellipsoid, 10-12 cm long, 5-valved, thinly white silky. Seeds ovoid, 10-12 mm long, embedded in white silky cotton (Naik 1998).

Boswellia serrata is middle sized trees with thin papery ash - coloured bark. Leaves odd pinnate, 10-30 cm long; leaflets 8-15 pairs; opposite, subsessile, ovate - oblong, 2-6 X 1-3 cm, oblique at base, crenate - serrate, obtuse, pubescent. Flowers 8-10 mm across, in axillary racemes shorter than the leaves. Calyx pubescent outside; lobes broadly triangular - ovate, 2-3 mm long. Petals dull or greenish - white, ovate, 5 mm long, pubescent outside. Stamens inserted at base of red, annular crenate disk. Ovary ovoid, sunk into the disk; style long. Drupes trigonous - ovoid, 1.5 - 2 cm long, pale green. Pyrenes compressed, heart – shaped (Naik 1998).

Ceiba pentandra is tall trees with smooth, green stem and horizontally spreading whorled branches. Leaves digitate; leaflets 5-9, lanceolate, 5-12 cm long, cuspidate, green above, glaucous beneath; petioles 10-15 cm long; petiolules 2-3 mm long. Flowers 3-5 cm across, clustered in the leaf axils; pedicels 2-4 cm long. Calyx 1.2-2 cm long,5-lobed, glabrous outside, hairy.within; lobes triangular-ovate. Corolla dull white or pinkish; petals obovate-oblong, 3.5-4 cm long, woolly outside. Anthers sinuous. Ovary conical, 1.5-2 cm long, glabrous. Capsules fusiform, 3.5-4 cm long, hairy. Seeds pyriform, 5-6 mm long, black, embedded in silky wool (Naik 1998).

Cordia dichotoma moderate-sized, deciduous trees up to 10 m tall; bark dark-coloured, rough, fissured; young parts pubescent. Leaves alternate, ovate, elliptic or orbicular, 4-9 x 3-5.5 cm. rounded, cordate or shortly cuneate at base, entire or sinuate dentate, obtuse, scabrous above, glabrous beneath. Flowers polygamous (male and bisexual), in large, lax, terminal and axillary, pedunculate cy- mose panicles; peduncles 2-5 cm long; pedicels short. Calyx 4-6 mm long, glabrous outside, pubescent within; teeth 5, shallow. Corolla white; tube as long as the calyx; lobes oblong, obtuse or emarginate, recurved, as long as the tube. Drupes globose or ovoid, 1-2.5

cm diam. pale orange coloured, supported by saucer-shaped, irregularly 5-10-lobed, longitudinally striate, glabrous calyx; pulp sticky. Seeds 2-4 in bony stone (Naik 1998).

So far there are only a few studies regarding phytochemistry, hence the present study was aimed to determine the phytoconstituents present in the selected medicinal plants.

MATERIAL AND METHOD:

Plant Material: The stem bark of *Neolamarckia cadamba (Roxb.) Bosser* was collected from Shivaji park Paithan, *Bombax ceiba* L., *Boswellia serrata* Roxb. ex Coleb. were collected from Dr. BAMU Aurangabad, *Ceiba pentandra* (L.) Gaertn. was collected from PMP college Paithan and *Cordia dichotoma* G.Forst was collected from Jayakwadi, Paithan. The plant materials were identified and Authenticated by Dr. M. A. Kare, Department of Botany, Pratishthan Mahavidyalaya, Paithan.

Preparation of Extract: The stem bark of selected medicinal plants shaded dried, and then these are made into coarsely powdered form using dry grinder. The powdered bark of the plants (180gm.) were packed in soxhlet apparatus and continuously extracted with petroleum ether (40-600C) till complete extraction, after completion of extraction the solvent were removed by distillation and then concentrated extract obtained were dried under reduced pressure using rotator evaporator at temperature not exceeding 400C and then give moderate heating on water bath. A yellowish extract approximate 1 gm. was obtained. From the drug petroleum ether were removed and the defatted drugs were extracted with methanol (95%) till complete extraction, after completion of extraction the solvent was removed by distillation and then concentrated extract obtained dried under reduced pressure at temperature not exceeding 400C and then give moderate heating on water bath. The methanolic extract obtained was dark yellow in color, weighed about 42.8 gm. The methanolic extract was kept in Petridis and it was stored in desiccators at cool place (Mukherjee 2001).

RESULTS AND DISCUSSION:

Phytochemical screenings were done in various bark extracts of *Neolamarckia cadamba (Roxb.) Bosser, Bombax ceiba* L., *Boswellia serrata* Roxb. ex Coleb., *Ceiba pentandra* (L.) Gaertn. and *Cordia dichotoma* G.Forst. The value of medicinal plants lies in some chemical substances that produce a definite physiological action on the human body and the most important phytochemicals are alkaloids, flavonoids, tannins and phenolic compounds (Hill 1952) (Florence, et al. 2015). The present investigations were carried out on five plants to study the presence of medicinally active phytochemicals test, Tannins, Phenols, Alkaloids, Saponins, Iridoids, Quercetin, Kaempferol, Catechin, Coumarin, 6,7-Dimethoxy coumarin, 5-Methoxy genistein, Anthocyanin, Proanthocyanin, Carbohydrates, Flavonoids, Glycosides and Proteins in the medicinal plants. The results are summarized in table.

Alkaloids have been used as both antibacterial and antidiabetic properties and useful for such activities. Phenols and phenolic compounds have been extensively used in disinfections and remain the standard with which other bacterisides are compared (Akinyeye, Solanke and Adebiyi 2014) (Santhi and Sengottuvel 2016). In plants phytochemicals are naturally present. They give colour, flavor, smell and texture. A part from that, phytochemicals could prevent diseases including cancer and cardiovascular diseases and inhibit pathogenic microorganisms. Nowadays the use of medicinal plants rapidly increases in medicine (Renu 2005).

Photochemistry of Five Selected Medicinal Plants

Sr. No.	Name of the Species	Tannins	Phenols	Alkaloids	Saponins	Iridoids	Quercetin	Kaempferol	Catechin	Coumarin	6,7-Dimethoxy coumarin	5-Methoxy genistein	Anthocyanin	Proanthocyanin
1.	*Neolamarckia cadamba*	+	+	+	+	-	-	-	-	-	-	-	-	-
2.	*Bombax ceiba*	+	+	+	-	-	-	+	-	-	-	-	-	-
3.	*Boswellia serrata*	+	+	+	+	-	-	-	-	-	+	-	-	-
4.	*Ceiba pantandra*	+	+	+	+	-	-	-	-	-	+	-	-	-
5.	*Cordia dichotoma*	-	-	+	+	-	+	-	-	+	-	-	-	-

Phytochemical screening of the bark showed some differences in the presence of phytoconstituents which are known to have importance in medicine (Sukumaran S 2011) (Kiruba S 2011) (Jeeva S 2011) (J. M. Jeeva S 2012) (Johnson M 2012) (AR, et al. 2014). The preliminary screening tests may be useful in the detection of the bioactive principles and subsequently may lead to the drug discovery and development (Joselin J 2013) (S. B. Joselin J 2012) (Florence AR 2012) (J. S. Joselin J 2014) (AR, et al. 2014).

Tannins and Phenols are found in *Neolamarckia cadamba, Bombax ceiba, Boswellia serrata, Ceiba pantandra* not in *Cordia dichotoma*. Iridoids, Catechin, 5-Methoxy genistein, Anthocyanin, Proanthocyanin are not found in all plants. Alkaloids are found in all selected medicinal plants. Saponins are found in *Neolamarckia cadamba, Boswellia serrata, Ceiba pantandra* not in *Cordia dichotoma* but not in *Bombax ceiba*. Quercetin and Coumarin are found in *Cordia dichotoma,* not in *Neolamarckia cadamba, Bombax ceiba, Boswellia serrata, Ceiba pantandra*. Kaempferol found in *Bombax ceiba* but not in *Neolamarckia cadamba, Boswellia serrata, Ceiba pantandra, Cordia dichotoma*. 6,7-Dimethoxy coumarin found in *Boswellia serrata* and *Ceiba pantandra* but not in *Neolamarckia cadamba, Bombax ceiba* and *Cordia dichotoma*. Flavonoids are also reported to have inhibitory action on growth and proliferation of different types of tumors (Netto 2007).

CONCLUSION:

From these analyses, the researcher observed that most phytochemical contents are higher in the barks; and therefore recommend the bark as the major source of these phytochemicals. Phytochemical evaluation extract of selected medicinal plants showed the presence of Tannins, Phenols, Alkaloids, Saponins, Iridoids, Quercetin, Kaempferol, Catechin, Coumarin, 6,7-Dimethoxy coumarin, 5-Methoxy genistein, Anthocyanin, Proanthocyanin, Carbohydrates, Flavonoids, Glycosides and Proteins. However, the higher content of most of these phytochemical

components; therefore, it is suitable for industrial purposes like; in pharmaceutical industries and cosmetic industries.

ACKNOWLEDGEMENT:

The authors are grateful to Principal, Pratishthan Mahavidyalaya, Paithan, for providing the necessary laboratory facilities and we are also thankful with our deepest core of heart to Dr. M. A. Kare for his valuable guidance.

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PLATE 01

A Flowering Twig

Outer Surface of Bark

Inner Surface of Bark

Neolamarckia cadamba (Roxb.) Bosser

PLATE 02

A Flowering Twig

Outer Surface of Bark

Inner Surface of Bark

Bombax ceiba L

PLATE 03

A Flowering Twig

Outer Surface of Bark

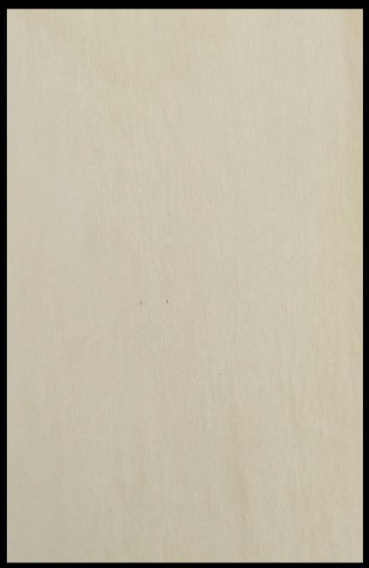
Inner Surface of Bark

Boswellia serrata Roxb. ex Coleb.

PLATE 04

A Flowering Twig

Outer Surface of Bark



Inner Surface of Bark

Ceiba pentendra (L.) Gaertn.

PLATE 05

A Flowering Twig

Outer Surface of Bark

Inner Surface of Bark

Cordia dichotoma G. Forst.

The cross-sectional study of ABO blood group system and Rh factor from Sillod tehsil Marathwada region (MS) India.

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Keywords:
ABO blood group, Anti ABD, Sillod.

Abstract:
Red blood cells (RBC) consist specific type of antigen on their surface. Discovery shows numerous type of antigen present on the RBC which determine type of blood group system such as ABO, P, MNS, LEVIS KIDS, Rh, DUFFY etc. The gene I on the 9th number of chromosomes shows multiple allele intragenic gene interaction for ABO blood group. Two important antigen A and B is the responsible for ABO blood group in human which are very common type. One gene (I), three alleles (IA, IB, I0), six genotypes and four phenotypes A, B, AB, O) determination as ABO system. ABO blood group system was first discovered by Karl Landsteiner in 1901. Another antigen D indicate Rh factor presence on the surface of same RBC. Thus ABO blood group system most essential in human being show allelic distribution and information about this system helps in blood transfusion accurately. The present cross-sectional study of ABO blood group system and Rh factor has been conducted from Sillod tehsil during June to December 2022. For this study involvement of authorized person of MAHA lab PHC Sillod Government, Private authorized clinical laboratory, and trained students. There were 2600 total blood samples collected for this study from Sillod tehsil. Out of them 1150 female while 1450 male participated in the camp as well filed visit to village. Anti ABD kit is used to determine type of blood group and Rh factor has been done. Out of total examination results shows highest number was O (846N, 32.53%), B (778N, 29.92%), A (696N, 26.76%) and AB (N=280N, 10.76%). Among Rh factor Rh -ve (184N, 7.07%), Rh +ve (2416N, 92.30%) and rare blood group O -ve were (57N, 2.19%) while two persons blood group not matched with Anti ABD system. Blood detection method is skilful and patience level process, minor misleading steps may cause variation in accuracy. Over a population and urbanization cause accidental cases and immediate blood transfusion, so there was an awareness about blood group detection camp organized in rural area. All participants was satisfied and note down there blood group type.

Introduction:

Among the human kind ABO blood group system is most widely worldwide accepted (ISBT, 2008). Red blood cells (RBC) consist of specific type of antigen on their surface. Discovery shows numerous type of antigen present on the RBC which determine type of blood group system such as ABO, P, MNS, LEVIS KIDS, Rh, DUFFY etc. The gene I on the 9th number of chromosomes shows multiple allele intragenic gene interaction for ABO blood group. Two important antigen A and B is the responsible for ABO blood group in human which is very common type. One gene (I), three alleles (IA, IB, I0), six genotypes and four phenotypes A, B, AB, O) determination as ABO system. ABO blood group system was first discovered by Karl Landsteiner in 1901 (Garraty G, 2000). Another antigen D indicate Rh factor presence on the surface of same RBC. Thus, ABO blood group system most essential in human being show allelic distribution and information about this system helps in blood transfusion accurately.

Molecular study reveals the human RBSs Antigen as glycoproteins and glycolipids represents type of blood group and controlled as genetically Mendelian manner whole life (Firkin F, 1989). H antigens are the main precursors for both A and B antigen while H antigen synthesis by the addition of fucose to the glycolipid or glycoprotein backbone and addition of N-acetyl galactosamine indicate Antigen A as well as galactose addition for B antigen (Jeffery S Anderson, Fauci A S, 2008). In order to avoid danger of mismatched blood transfusion, it is important to determine the blood group of those involved prior to a transfusion. These days, to eliminate the risk of transfusion reaction, the practice of autologous transfusion is followed by most of the physician. From ancient times human races were more been fascinated by blood, Aristocrats drank blood while Egyptians bathed in the blood (Ghori M R, 2003) and modern human transfuse and risk of blood transfusion (Ganong, 1997). Such transfusion occurs by ABO antibodies in plasma so this transfusion is highly fatal (Sazama K, 1990). The importance in organ transplantation, genetic research, Gene interaction legal medicine, and anthropology (Storry, 2003). Detection of blood group is most important for detection certain diseases (Qureshi M A, 2003). Different types of diseases also associated with different types of blood group (Aird I, 1953 and Mollison P L, 1993). The erythroblastosis foetal is the fatal new born death caused by Rh -ve type blood type in female (Lo Y M, Hjelm N M ,1998).

Materials and method:

The present cross-sectional study of ABO blood group system and Rh factor has been from Sillod tehsil during June to December 2022. For this study involvement of authorized person of MAHA lab PHC Sillod Government, Private authorized clinical laboratory, and trained students. There were 2600 total blood samples collected for this study from Sillod tehsil. Out of them 1150 female while 1450 male participated in the camp as well filed visit to village. Anti ABD kit is used to determine type of blood group and Rh factor has been done.

Fist fingertips were clean by 70% alcohol swab then pricked sterilized needle and carefully direct transport 3 drop separately on grease free slide. Anti ABD is used to 1-2 drops on blood sample separately. Mixed well them and wait 3 to 5 minutes to observation clotting in which blood type. The clotting part indicate type of blood group while anti D sera blood clots shows positive and does not clot shows negative type of blood group. Such type is recorded according to age, sex, religion wise. All the data input in Windows Microsoft excel; (win11) tabulation, percentage and frequency has been done.

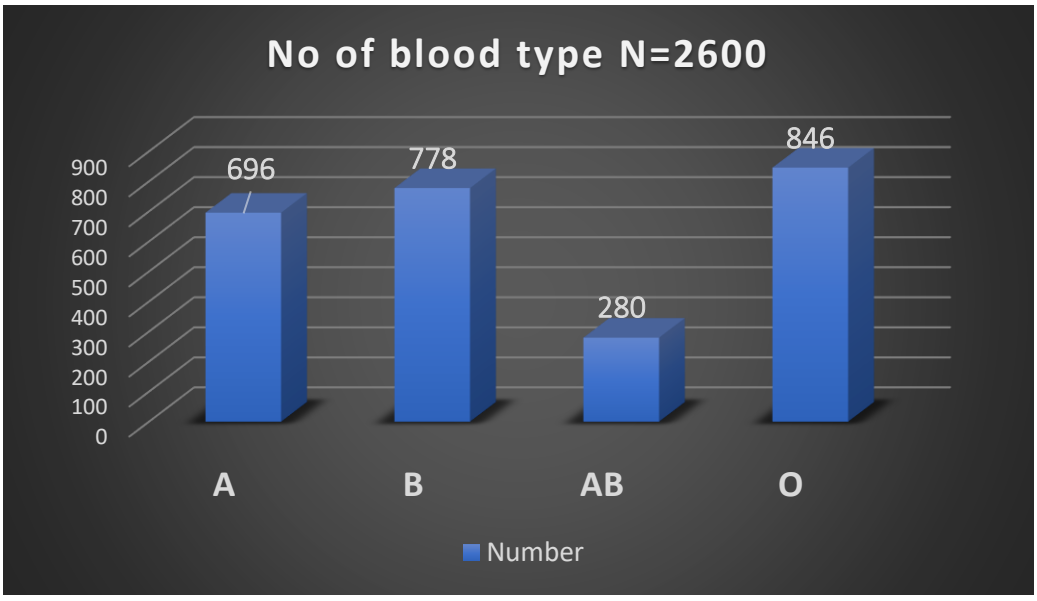
Results and discussion:

Blood group detection method is skilful and patience level process, minor misleading steps may cause variation in accuracy. Over a population and urbanization cause accidental cases and immediate blood transfusion, so there was an awareness and need to blood group detection camp organized in rural area. All participants satisfied and note down their blood group type. Blood group system mostly helpful in human population genetics, blood donation, blood disorder, haemolytic diseases etc.

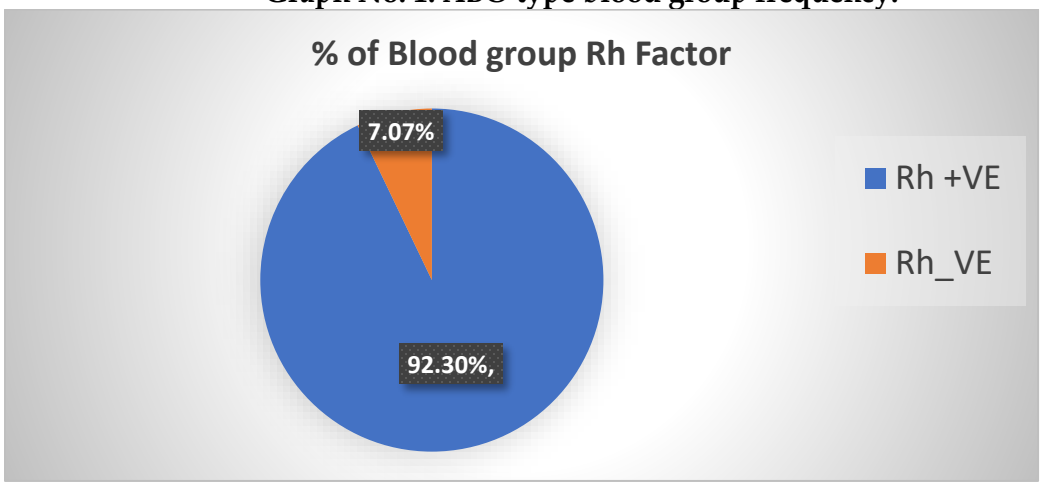
There were 2602 total blood samples collected for this study from Sillod tehsil. Out of them 1150 female while 1452 male participated in the camp as well filed visit to village, Graph No. 3. Anti ABD kit is used to determine type of blood group and Rh factor has been done. Out of total examination results shows highest number was O (846 N, 32.51%), B (778 N, 29.90%), A (696 N, 26.74%) and AB (N=280N, 10.76%), in Table No.1 and graph No. 1. Among Rh factor Rh-ve (184 N, 7.07%), Rh +ve (2416 N, 92.30%) in Graph No. 2. and rare blood group O-ve were (57 N, 2.19%) while 2 persons blood group not matched with Anti ABD system. Two samples of the participated person's blood type does not matched with ABO system so this might be new type blood group system and must need in detailed molecular study. Similar type of work also reported by Shaikh Y. A., 2007 from Gaza, Yousaf M, 1988 from Bahawalpur Division, Giri P. A., 2011 from Rural Tertiary Care Hospital in India, Warghat N. E., 2011 Amravati district (Maharashtra), Tulika Chandra, 2012 from north India and Tariq Kamal Jafri, 2014 from Karachi.

Type of blood groups	Number of samples	Percentage (%)
A	**696**	**26.74%**
B	**778**	**29.90%**
AB	**280**	**10.76%**
O	**846**	**32.51%**
Not matched	**002**	**0.07%**
ABO	**2602**	**100%**

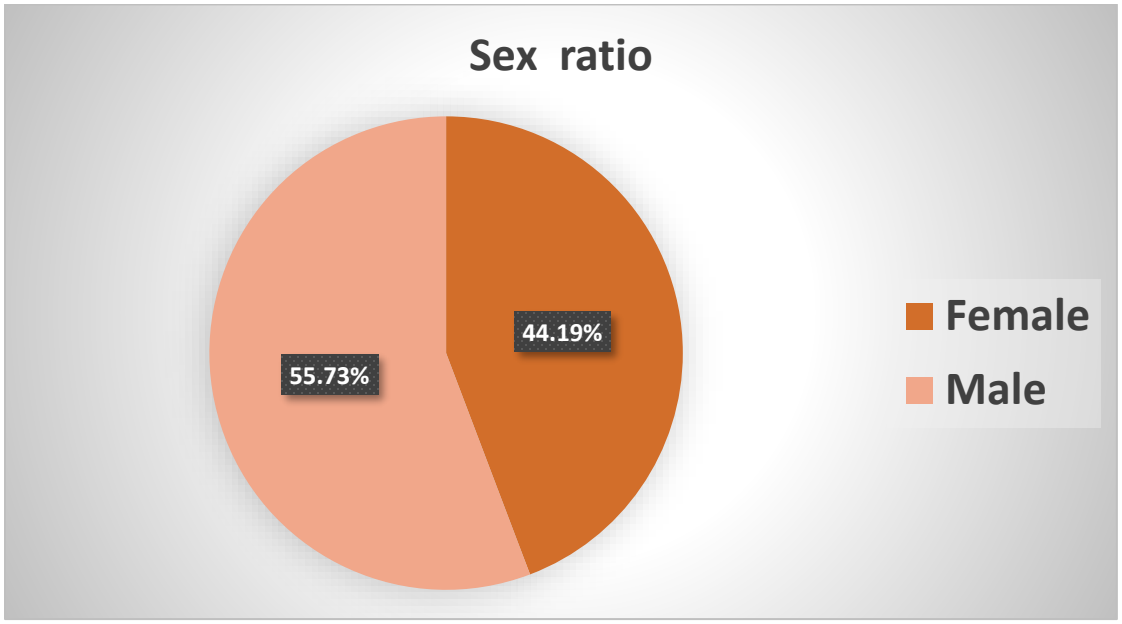
Table No.1: Table shows blood group frequency and percentage.



Graph No. 1: ABO type blood group frequency.



Graph No. 2: Blood group Rh Frequency.



Graph No. 3: Blood group Frequency Sex ratio.

Acknowledgement: Authors are thankful to the staff and clinical laboratory technician of Primary Health Centre, Sillod for their support and help during the camp, also thankful to the peoples those are participated in this camp. Authors are also thankful to the Principal, Yeshwantrao Chavan College, Sillod for using laboratory and library facilities.

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General Characteristics and Mode of Action of Cypermethrin

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Keywords:
Aquaculture technology, pyrethroid causes

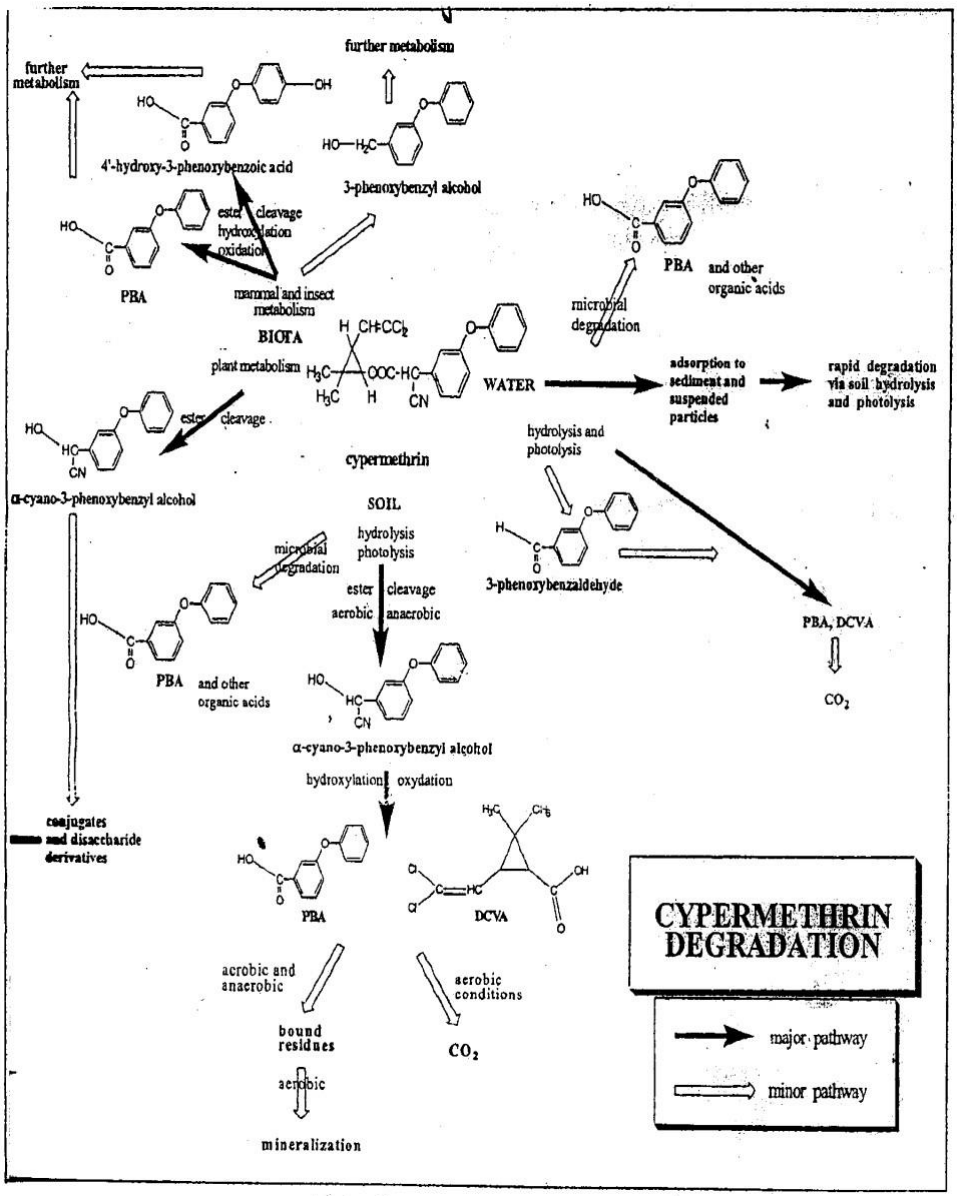
Abstract:

Now a days aquaculture has increased its potential by using powerful tools and systematic technology. Of course these technologies develop Aquaculture technology but on other hand intense activity in Industrial and agricultural sectors had inaviability increased the level of heavy metals, pesticides in natural waters among which pesticides play a major role among pollutants of aquatic environment. When we take spotlight on the impact of pesticides some pyrethroid causes hazardous impact on fishes, many aquaculture researchers had made research on impact of pyrthoid in various terms. Most of the work had been carried out outside India and least attempts had been carried out in India. Pyrethroid insecticides are used on a worldwide scale. It has been calculated that pyrethroids accounted for 25% of the insecticide market in the industrial countries in the 90s. Pyrethroids are the most common insecticides for both indoor and agricultural purposes. Their chief advantages are high insecticidal potency and low mammalian toxicity with rapid metabolism and lack of terrestrial accumulation.

Introduction

Cypermethrin is a synthetic, pyrethroid insecticide that has high insecticidal activity, low avian and mammalian toxicity, and adequate stability in air and light (Kaufman et al., 1981). It is used to control many pests including lepidopterous pests of cotton, fruit and vegetable crops and is available as an emulsifiable concentrate or wettable powder. According to the label for Ammo®2.5 EC insecticide, which contains 2.5 pounds of cypermethrin per gallon, the product should not be applied directly to water or to areas where surface water is present. Also, cypermethrin should not be applied when wind may cause drift beyond the intended treatment area. Due to its extreme toxicity to fish and aquatic organisms, Ammo®2.5 EC is registered as a "restricted use pesticide", and is for sale to, and to be used only by, Certified Applicators.

The environmental fate of cypermethrin [(±)-Alpha-Cyano-(3- phyenoxyphenyl) methyl (±) - cis/trans – 3 - (2, 2 - dichlorovinyl) -2, 2 dimethylcyclopropane carboxylate.] cypermethrin is a synthetic, pyrethroid insecticide that is available in several formulations as an emulsifiable concerntrate or wettable powder. The following diagram shows as per methrin degradation.



In vertebrates and invertebrates, cypermethrin acts mainly on the nervous system. Cypermethrin is both a stomach poison and a contact insecticide (Jin and Webster, 1998). In the peripheral nervous system of the frog, its primary action is to induce noticeably repetitive activity and produce trains of nerve impulses as a result of altering ion permeability of nerve membranes (Vijverberg and van den Bercken, 1990; and Siegfried, 1993). These long-lasting trains can cause hundreds to thousands of repetitive nerve impulses in the sense organs. This repetitive activity is induced by pyrethroid damage to the voltage-dependent sodium channel, causing sodium channels to stay open much longer than normal. (Vijverberg and van den Bercken, 1990).

Cypermethrin has been shown to inhibit ATPase enzymes involved in movement of ions against a concentration gradient which are regulated by active transport. This action is especially critical to fish and aquatic insects where ATPase enzymes provide the energy necessary to active transport, and very important at sites of oxygen exchange. ATPase inhibition and disruption of active transport, possibly affect ion movement and the ability to maintain ion balance, and disrupt

respiratory surfaces, indicating that cypermethrin is inherently more toxic to aquatic organisms (Siegfried, 1993).

Conclusion

Of course these technologies develop Aquaculture technology but on other hand intense activity in Industrial and agricultural sectors had inaviability increased the level of heavy metals, pesticides in natural waters among which pesticides play a major role among pollutants of aquatic environment.

When we take spotlight on the impact of pesticides some pyrethroid causes hazardous impact on fishes, many aquaculture researchers had made research on impact of pyrthoid in various terms.

Most of the work had been carried out outside India and least attempts had been carried out in India.

Cypermethrin is a synthetic, pyrethroid insecticide that has high insecticidal activity, low avian and mammalian toxicity, and adequate stability in air and light.

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The environmental fate of cypermethrin cypermethrin is a synthetic, pyrethroid insecticide that is available in several formulations as an emulsifiable concerntrate or wettable powder.

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Use of Silk cocoon: A boon in cosmetic Industry

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Keywords:
Silk cocoon, Fibroin, Sericin

Abstract:
It's said that beauty lies in the eyes of beholders, but now it's age to maintain the external and internal beauty of personality. Regarding the maintenance of beauty everyone is very protective regarding their skin. For instant better effects we use chemical-based beauty products. In this era of chemicals Silk cocoon facial is boon to human beings. Silk cocoon is mainly made up of two proteins. One is fiber protein that is fibroin and other is adhesive glue protein that is sericin. Sericin is an amazing protein containing 18 amino acids. Sericin acts as a thin film over the face sealing in moisture to keep skin hydrated. It contains highly hydrophobic amino acids which has antioxidants potential. Therefore, it has high demand in cosmetic industry. The moisturizing power allows indications as a therapeutic agent for wound healing, stimulating cell proliferation, protection against ultraviolet radiation and formulating creams and shampoos.

Introduction:
There is one worm from phylum Arthropoda, Class Insecta called *Bombyx mori,* which is reared for production of silk. In India four types of silk are reared i.e Mulberry Silk, Eri silk, tasar silk, muga silk. These silks are obtained from the silk cocoon. Cocoon is a material which is composed of thin and continuous strands of fibers. Cocoon is natural silk composite with non-woven structure made of continuous silk fiber. These fibers are nothing but the production of Proteins Of silk glands of silkworm. Silkworm's stage of pupa feeds on Mulberry leaves. Silkworm weaves a net around itself to keep itself alive in pupa stage. The secretions produce by the salivary glands ultimately turns into silk. Two major proteins are present in the Cocoon named fibroin and sericin. Fibroin is the central fibrous protein and sericin is the globular protein. Sericin is the sticky part that envelopes the fibers and coheres them together. Sericin has numerous properties like antitumor anti-ageing anti-wrinkle. Sericin is rich in four amino acids that are Serin, glycine, aspartic acid and threonine. These four amino acids have various function which results in healthy skin. serin and threonine functions as natural moisturizing factors which act as water binding molecules. These proteins also help in replenishing skins barrier function. Glycine improves the visible signs of ageing, increases production of collagen and strengthens the skin and promotes skin repair and regeneration. Aspartic acid naturally balances the pH of skin. Cocoon is package of all required beauty products. This sericin has excellent moisture absorbance and anti-microbial property The silk peptide has good therapy effects for several dermatosis. Such as skin cracks and skin burns. Sericin forms a substantive protective anti-wrinkle film on the skin surface.

Methodology- The silk cocoons were given to 20 peoples and asked them to apply it on face. Following steps were used as methodology for this research paper.

1) Take one cup of warm water and soak 4-6 cocoons in water.
2) Cut one end of cocoon through silkworm and insert it on a finger.
3) Gently rub it on your face and massage your facial skin.

Fig : Copied image showing steps of using cocoons

Results: Among the 20 peoples 8 were facing acne problems 5 were facing dry skin 2 has wrinkles problem and 5 were allergic to chemical-based cosmetics. Continuous use of this cocoon facial has shown following changes on facial skin.

1) Gaining moisturizer
2) Removing tan and dirt
3) Immediate imparting long lasting silk feeling to skin that can even persist after showering also.
4) Images shows the results of before and after use of silk cocoon facial.

Picture: showing use of cocoon facial before and after

Disscusion:

Sericin has properties like biocompatibility, biodegradability and wettability that allows development of cosmetic products for skin, nails and hair (Pawar & Padamwar, 2004; Voegeli et al., 1993; Yamada et al., 2001). Moisturizers made by using sericin protein has special development; these moisturizers are used to prevent and delay the dehydration of the top layer of the skin. The water loss from upper layer od skin epidermis can be restored by using these moisturizers. (GmbH Ziolkowsky, 1998).

Normal and healthy skin has wet, clean, soft, flexible, malleable look (Idson, 1987). The smoothness can be retain by applying silk sericin .(Blank, 1952).

Silk sericin is ideal ingredient for for cosmatic application in the formulation of specific products for skin care and hair care. It shows great potencial in reapiring (Chromatographic profiling of silk sericin for biomediical and cosmetic use by Sara Tengattini, Giulia orlandi 2020)

Conclusion : Chemical based products shows instant better effects but longer harmful effects. Silk cocoon facial is a eco friendly it's a natural fibre and biodegradable also. It has some very effective properties like antitumour anti tanning anti wrinkles anti oxidants and a sucessful effect of acne. Worn out cocoons are waste. So we can use it best from waste. As compaired other chemical based products this silk cocoon facial is affordable.

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A FLORISTIC SURVEY OF PLANTS DIVERSITY FROM JALNA EDUCATIONS SOCIETY'S COLLEGE CAMPUS, JALNA (MH) INDIA

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ABSTRACT

Keywords:
Diversity, Flora, Survey, Conservation.

Abstract:
The present study have been conducted to analyze the floristic survey to check species community of plants and enumeration of that species that will be helpful as a field guide to the beginners of having taxonomic interest. In this survey the total diversity of the college campus that includes all genus, species in plant community. Several studies deals with the survey of floristic diversity of the college campus in the former sense i.e. the number of individual species in an area, it will help to understand the significance if existing diversity because it's necessary to understand that its valued of the place and what its relevance for the future, so the purpose of this attempt to highlight the diversity of plants resources of a college campus in the conservation perspective. In this survey we have focused only flowering plants of the college campus.

INTRODUCTION

The angiosperms diversity is the biggest natural assets and economical important group of plants for the human kind in many ways. It is spread all over the globe not only angiosperms but the whole plants community on the earth such as bryophytes, pteridophytes and gymnosperms also provides the number of products, food material and act as medicinal and fuel source for human beings. The over utilization of such essential plants resources for development of nation and welfare of human being is being occurs daily. This indiscriminate utilization of plants resources cause the adverse effect on climate changes and ecological imbalance it leads to disturbance in biodiversity.

This scenario necessitates the urgent need for conservation of biodiversity and plants resources. To generate various strategies for this purpose first step which is important is to explore the inventories of flora. By keeping this perspective in point of view of conservation, the present's studies were conducted to explore and to understand the knowledge of the floristic composition of plant community in a given area. Floristic steadies acquire increasing importance in recent years in response to the need of developing and under developing countries to assess their plant wealth (Vediya and Kharadi, 2011).

The floristic survey which can be defined as the variety and variability of plants in a given region. Its includes the study of number of type of species in given region or group, it can measured at any level of ecological diversity (Nasir Aziz and V.R. Deshmukh 2015).

MATERIAL AND METHODS

The present study was carried out through extensive field visit from July 2019 to March 2020. During field survey the plants had been observed at flowering stages as far as possible from their natural habitat. They were identified with standard flora (Hooker, 1982; Naik, 1998).

Study area

Jalna is a district in the state of Maharashtra with its district headquarters situated at 19.8410°N 75.8864°E. It has an average elevation of 508 m (1,667 ft), on the banks of the Kundalika River. Jalna education society's college, Jalna is located near the police training center, Jalna. The Jalna Education Society, college Jalna is one of the institute of Marathwada region was formed in 1957 with an aim to provide higher education and research work to encourage the students of the town and nearby rural areas in various science disciplines. In fact, establishment of a full-fledged college was a long felt need of Jalna. It is sprawled over area of 16 acres in picturesque landmass with pollution free environment. An attempt was made to explore the flowering plant diversity of the college campus. The college has a set of beautiful old buildings along with play ground, gardens and teak plantation. The campus sprawled over an area of 16 acres in picturesque landmass.

RESULTS AND DISCUSSION

The present floristic survey deals with the documentation of eco friendly college campus which is associated with rich of flora of herbs, climbers, shrubs and trees. This taxonomic survey of campus was carried out from different localities of campus. There are many socio-economical, medicinal and ornamental plants grown in this campus. Most of plants are naturally grown and many of are planted obviously for beautifulness and to control the pollution.

All the visits were made during the flowering and fruiting seasons. During these visits, 146 species of plants belonging to 25 families were collected and identified from the college campus. The dominant families in the campus representing the maximum number of species were Fabaceae, Asteraceae, Malvaceae, Portulacaceae, Lythraceae, Rutaceae, Sapotaceae, Meliaceae, Myrtaceae and Apocynaceae. Whereas the rest of the families were represented by only one species in the study area Jalna education society's college campus had good biodiversity.

The campus is home to many trees, herbs and many species of shrubs growing naturally in campus. However in the recent past due to various activities such as construction and developmental works going on in the campus many species of trees have been cut down. This has caused huge damage to the floristic composition on the campus. Despite this many herbaceous plants have exited this campus owing to these anthropogenic activities. Many species of exotic ornamental plants have been also planted in the campus for decorative and beauty purposes which have also caused disturbance in the natural floristic diversity in the campus therefore efforts must be taken for ensuring ecological restoration and conservation of various species inhabiting the campus.

SR.NO.	BOTANICAL NAME	VERNACULAR NAME	LIFE FORM	FAMILY
1	*Annona aquaomosa L.*	Sitafal	T	Annonaceae
2	*Abitulon indicum (L) Sweet.*	Shikha	S	Malvaceae
3	*Abitulon pannosum (forst f.)*	Karandi	S	Malvaceae
4	*Acacia nilitica*	Babhul	T	Mimoceae
5	*Acalypha indica L.*	Khokali	H	Euphorbiaceae

6	*Acanthospermum hispidum DC.*	Landga	H	Asteraceae
7	*Achiranthus aspera L.*	Aghada	H	Amaranthaceae
8	*Alternanthera sessilis (L)*	…..	H	Amaranthaceae
9	*Aegle marmelos (L)*	Bel	T	Rutaceae
10	*Alstonia scholaris (L)*	Sapatparna	T	Apocynaceae
11	*Alysicarpous ovalifolius (Schumach) j. Leonard*	……	H	Fabaceae
12	*Alysicarpous vaginalis (L)*	……..	H	Fabaceae
13	*Alysicarpous bupleufolius (L)*	……..	H	Fabaceae
14	*Adhatoda vesica Nees*	Adulasa	S	Acanthaceae
15	*Aloe vera (L.)*	Korfad	H	Liliaceae
16	*Andrigraphis echioides Roxb*	Ranchmani	H	Acanthaceae
17	*Azadirachta indica A.Juss*	Kadu Nimb	T	Meliaceae
18	*Argemon maxicana*	Pivala Dhotara	S	Papavaraceae
19	*Bambusa vulgaris L.*	Bamboo	H	Poaceae
20	*Biophylum sensitivum L.*	Lajalu	H	Oxalidaceae
21	*Bauhunia resemosa Lamak.*	Apta	T	Caesalpinaceae
22	*Boerhavia diffusa L.*	Punarnava	H	Nyctanginiaceae
23	*Bougainvallea spectabilis Willd*	Bouganvalia	Tw	Nyctanginiaceae
24	*Bryophyllum calycinum*	Panfuti	H	Crassulaceae
25	*Butea monosperma (Lamak)*	Palas	T	Fabaceae
26	*Barleria prinitis L.*	Kate Koranti	S	Acanthaceae
27	*Bidens biternata (Lour.)*	……..	H	Asteraceae
28	*Caesalpinia pulcherima (L.)*	Shankarshwar	T	Caesalpinaceae
29	*Calotropis procera (ait)*	Rui	S	Asclepiadaceae
30	*Calotropis gigantean (L.) R. Br*	Mandar	S	Asclepiadaceae
31	*Cynadon dectyln L.*	Harali	H	Poaceae
32	*Cyonotis fasciculate (Heyne & Roth)*	…….	H	Commelinaceae
33	*Crotolaria medicagina Lamk.*	…….	H	Fabaceae
34	*Cordia diacotoma Forst.*	Bhokar	T	Boraginaceae
35	*Cocos nusifera*	Naral	T	Arecaceae
36	*Corchorus capsularis L.*	Harin Khuri	H	Tiliaceae
37	*Corchorus olitorius L.*	Harin Khuri	H	Malvaceae
38	*Commelina benghalansis L.*	Kena	H	Commelinaceae
39	*Canthium coromandelium (BURN.F.)*	Karabit	S	Acanthaceae
40	*Cardiospermum helicacabum L.*	Kapalfuti	Cl.	Sapindacaeae

41	*Caryota urens L.*	Surmaata	T	Areaceae
42	*Cassia absus L.*	……..	S	Caesalpinaceae
43	*Cassia fistula L.*	Bhahava	T	Caesalpinaceae
44	*Cassia auriculata L.*	Tarwad	S	Caesalpinaceae
45	*Cassia siamea L.*	Kashid	T	Caesalpinaceae
46	*Cassia tora L.*	Tarwad	H	Caesalpinaceae
47	*Cassia uniflora L.*	….	H	Caesalpinaceae
48	*Clorious montana Roxb.*	….	H	Poaceae
49	*Cissamum indicum L.*	Havari	H	Pediliaceae
50	*Cleom viscosa L.*	Piwali Tilwan	H	Cleomaceae
51	*Clitoria ternatea L.*	Gokarne	Tw	Fabaceae
52	*Cocculus hirsutus L.*	Wasan Wel	Cl.	Menispermaceae
53	*Covolvulus arevensis L*	Gland Wel	Tw	Convolvulaceae
54	*Crotolaria verrucosa L.*	…..	H	Fabaceae
55	*Dalbergia sissoo Roxb.*	Sheesham	T	Fabaceae
56	*Datura ferox L.*	Dhotra	H	Solanaceae
57	*Datura inoxa L.*	Dhotra	H	Solanaceae
58	*Datura metal L.*	Kala Dhotra	H	Solanaceae
59	*Datura stramonium L.*	Dhotra	H	Solanaceae
60	*Delonix regia (L.) Gambe*	Gulmohar	T	Caesalpinaceae
61	*Diplocyclos palmatus (L.) Jeffrey*	Shivlingi	Cl.	Cucurbitaceae
62	*Duranta erecta L.*	……..	S	Verbanaceae
63	*Eclpta alba L.*	Maka	S	Asteraceae
64	*Enicostema oxillare Lam.*	Nai.	H	Gentianaceae
65	*Emblica sonchifolia L.*	Sadmandi	H	Asteraceae
66	*Ecbolium viride L.*	Harawi Aboli	S	Acantaceae
67	*Eucalyptus globulus L.*	Nilgiri	T	Myrtaceae
68	*Eucalyptus tereticornis Sm.*	Nilgiri	T	Myrtaceae
69	*Euphorbia glandifolia L.*	Dudhi	H	Euphorbiaceae
70	*Euphorbia thymifolia L.*	Dudhi	H	Euphorbiaceae
71	*Euphorbia hirita L.*	Dudhani	H	Euphorbiaceae
72	*Evolvulus alsinoides L.*	Vishanu Kranta	H	Convolvulaceae
73	*Fucus religiosa L.*	Pimpal	T	Moraceae
74	*Fucus benghal;ansis*	Vad	T	Moraceae
75	*Ficus recemosa*	Umber	T	Moraceae
76	*Goniogyna hirita Vahl.*	Godhadi	H	Fabaceae
77	*Gliricidia sepium (Jacq.)*	Undirmari	T	Fabaceae
78	*Hemilia patens Jacq.*	……..	S	Rubiaceae
79	*Hibiscus rosa- sinensis L*	Jaswand	S	Malvaceae

80	*Hyptis suaveolens L.*	Rantulas	H	Maliaceae
81	*Indigofera linifolia (L.) Retz*	……..	H	Fabaceae
82	*Indigofera glandulosa Wendl.*	Barbada	H	Fabaceae
83	*Indigofera cordifolia Roth.*	……..	H	Fabaceae
84	*Ipomea carenea Jacq.*	Besharam	Tw	Convolvulaceae
85	*Ipomea obscura L.*	…..	Tw	Convolvulaceae
86	*Ipomea pesti-gridis L.*	…..	Tw	Convolvulaceae
87	*Jatropha gossypifolia L.*	…..	S	Euphorbiaceae
88	*Jatrophaintegerrima L.*	…	S	Euphorbiaceae
89	*Justicia diffusa willd.*	….	H	Acanthaceae
90	*Kirgania reticulata (Pior.) Bill*	Datwan	S	Euphorbiaceae
91	*Lantana camera L.*	Ghaneri	S	Verbanaceae
92	*Launaea procumbence Roxb.*	Pathari	H	Asteraceae
93	*Leucaena leucocephala*	Subabool	T	Fabaceae
94	*Leonatis inermis L.*	Deepmal	H	Lamiaceae
95	*Lavandula bipinnata L.*	Jangali Lavender	H	Lamiaceae
96	*Leucosa aspera Willd.*	……	H	Lamiaceae
97	*Mangifera indica L.*	Amba	T	Anacardiaceae
98	*Martynia annua*	Wagh Nakhi	S	Martantaceae
99	*Mimosa pudica L.*	Lajalu	S	Mimoceae
100	*Mimosa hamata Willd.*	……	S	Mimoceae
101	*Melia azedarach L.*	Bkan Limb	T	Meliaceae
102	*Malva malvastrum L.*	…..	H	Malvaceae
103	*Malvastrum coromandelianum L.*	…..	H	Malvaceae
104	*Merremia emarginata Burm.*	Undir Kani	H	Convolvulaceae
105	*Nerium indicum Mill*	Kanher	S	Apocynaceae
106	*Nyctanthes ardor-tristis*	Din Ka Raja	S	Nyctanthaceae
107	*Oscimum basilacum L.*	Sabja	H	Lamiaceae
108	*Oscimum amaricanum L.*	Ajgandha	H	Lamiaceae
109	*Oscimum tenuiflorum L*	Tulas	H	Lamiaceae
110	*Oxalis corniculata*	Ambuti	H	Oxalidaceae
111	*Peltophorum pterocarpum (vogal) Wal.*	Sonmohar	T	Fabaceae
112	*Pennisetum pedicellatum Trin.*	……	H	Poaceae
113	*Parthenium hysterophorus L*	Gajar Gawat	H	Asteraceae
114	*Phyllanthus nirurri Acut.*	Bhui Awala	H	Euphorbiaceae
115	*Plumeria acutifolia L.*	Chapha	S	Apocynaceae
116	*Pongamia pinnata L.*	Karanj	T	Fabaceae
117	*Prosopis julifera (SW.)DC.*	Wilayti Babhul	T	Fabaceae
118	*Pithecellobium dulce L.*	Wilayti Chinch	T	Fabaceae

119	*Pisidium guayava L.*	Peru	T	Myrtaceae
120	*Polyalthia longifolia (Sonner.)*	Ashok	T	Annonaceae
121	*Pergularia daemia (forsk)*	Utran	S	Asclepiadaceae
122	*Pandanus odorifer*	Kewada	S	Pandanaceae
123	*Ricinus communis L.*	Earand	S	Euphorbiaceae
124	*Syzyjium cuminis*	Jamun	S	Myrtaceae
125	*Sesbania aculeata Willd.*	Ran Shivari	S	Fabaceae
126	*Santalum album L.*	Chandan	T	Santalaceae
127	*Tribulus terestri L.*	Sarata	H	Zygophyllaceae
128	*Tridex procumbence L.*	Ekdandi	H	Asteraceae
129	*Tinospora cordifolia (Willd)*	Gudwel	Tw	Menispermaceae
130	*Thunbergia fragrance L.*	…..	Tw	Apocynaceae
131	*Terminallia catappa*	Badam	T	Combretaceae
132	*Thevetia peruviana*	Piwala Kanher	T	Apocynaceae
133	*Tectona grandis*	Indian Sag	T	Verbenaceae
134	*Tamarindus indica*	Chinch		Caesalpinaceae
135	*Triumphetta rotundifolia Lamk*	…..	T	Tiliaceae
136	*Triumfetta rhomboidea Jacq.*	Chota Zingoti	H	Tiliaceae
137	*Verbascum chinesis L.*	Kutaki	H	Scrophuariceae
138	*Vigna radiata L.*	Moog	H	Fabaceae
139	*Vigna ungulata L.*	….	H	Fabaceae
140	*Vinca rosea L.*	Sada Fuli	H	Apocynaceae
141	*Vitex nugundo*	Nirgudi	S	Verbanaceae
142	*Withania somiofera L.*	Aswaghandh	H	Solanaceae
143	*Xanthium stramonium L.*	Landa	S	Asteraceae
144	*Zizypus jijuba lamk.*	Ber	T	Rhamnaceae
145	*Zornia diphyla L.*	….	H	Fabaceae
146	*Zizipus mauritiana*	Ber	T	Rhamnaceae

CONCLUSION

Every educational society and institutions should have right from school level to PG level colleges have to maintain the list of flora as well as fauna in that particular area. It will help to understand species, endemic species, ecological community and natural resources which are important for long time survival of human well being. Biodiversity which provides a valuable important environment services from its species and ecosystem that are very essential for global, regional and locally as well. The plants maintain the water balance, preserving ecological structure and function such as food web, fixing of nitrogen, soil formation, etc. Humans are responsible to encroachment of biodiversity and it results includes a conflict between man and wild life so, the human has to conserve the wild flora and natural resources by protecting them in a natural environment itself.

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Green Chemistry and it's uses in Life Sciences

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Keywords:
Green Chemistry, Sustainable Chemistry, Life Science, Bio-Catalyst, Sustainable Development, Environment,etc

Abstract:
Green chemistry, also known as sustainable chemistry, is a field that aims to reduce or eliminate the use and generation of hazardous substances in chemical production. In the life sciences, green chemistry principles can be applied to develop more sustainable and environmentally friendly methods for producing natural products, pharmaceuticals, and agricultural products. This can be achieved through the use of renewable resources, safer solvents and reaction conditions, and the reduction of waste and pollution. By designing products that can be easily recycled or reused, the life sciences can become more sustainable and environmentally friendly. Green chemistry has significant potential to improve the safety and sustainability of the life sciences, and will continue to play an increasingly important role in shaping the future of this field.

Introduction

Green chemistry, also known as sustainable chemistry, is a field that focuses on designing chemical products and processes that reduce or eliminate the use and generation of hazardous substances. The principles of green chemistry aim to minimize the negative impact of chemical production on the environment and human health. In recent years, green chemistry has become increasingly important in life sciences due to its potential to improve the safety and sustainability of pharmaceuticals, agriculture, and other areas of the life sciences.

One of the key principles of green chemistry is the use of renewable resources. In the life sciences, this means developing sustainable and environmentally friendly methods for producing natural products such as medicines, fragrances, and flavors. For example, the use of plant-based sources of active ingredients in medicines can reduce the reliance on synthetic chemical production and minimize the risk of toxic by-products.

Green chemistry also emphasizes the use of safer solvents and reaction conditions. This can be particularly important in the development of pharmaceuticals, where the use of toxic solvents can pose a significant risk to human health. By using safer solvents and reaction conditions, the development of pharmaceuticals can be made more sustainable and environmentally friendly. In addition, green chemistry can help to reduce waste and pollution in the life sciences. One example is the development of sustainable agriculture practices that minimize the use of harmful pesticides and fertilizers. This not only reduces the environmental impact of agriculture but also helps to improve food safety and security.

Another example is the use of biocatalysis, which involves using enzymes and other natural catalysts to facilitate chemical reactions. Biocatalysis can be used to produce a wide range of products, including pharmaceuticals, flavors, and fragrances, in a more environmentally friendly way than traditional chemical processes. Green chemistry also emphasizes the importance of

designing products that can be easily recycled or reused. This is particularly important in the development of packaging materials for pharmaceuticals and other life science products. By designing packaging materials that are easily recyclable or reusable, the environmental impact of these products can be minimized.

Overall, the principles of green chemistry have significant potential to improve the sustainability and safety of the life sciences. By reducing the use and generation of hazardous substances, minimizing waste and pollution, and designing products that can be easily recycled or reused, the life sciences can become more environmentally friendly and sustainable. As the importance of sustainability continues to grow in the life sciences, green chemistry will play an increasingly important role in shaping the future of this field.

The role of green chemistry in zoology

Green chemistry can be useful in zoology in several ways. One of the main applications of green chemistry in zoology is in the development of sustainable and environmentally friendly methods for animal research. This can include the use of non-toxic or low-toxicity chemicals and solvents in the laboratory, reducing waste generation and disposal, and implementing recycling and waste management practices. Another application of green chemistry in zoology is in the development of sustainable and safe methods for controlling pests and disease in animals. This can involve the use of natural pesticides and biological controls, such as the use of predator species to control pests, rather than relying on synthetic chemicals that can have negative environmental impacts. Green chemistry can also be used to develop sustainable and safe methods for the preservation of animal specimens and tissues, which is important for research and teaching purposes. This can involve the use of non-toxic preservatives and fixatives that do not generate hazardous waste, as well as implementing waste management practices for disposal of these materials.

Overall, green chemistry offers an approach to animal research and management that is safer, more sustainable, and less harmful to the environment. By reducing the use and generation of hazardous substances, minimizing waste and pollution, and designing sustainable methods for animal research and management, zoology can become more environmentally friendly and sustainable.

Green Methods Used in Life Sciences

some methods of green chemistry that are used in life science research:

Microwaves: Microwave-assisted chemistry is a fast and efficient way to carry out chemical reactions in a sustainable way. In life sciences, microwave-assisted extraction is used to extract bioactive compounds from plant material for drug development.

Ionic liquids: Ionic liquids are a type of solvent that can be used in place of traditional organic solvents. They are non-toxic, non-volatile, and have low environmental impact. Ionic liquids have been used in life sciences research for protein extraction, enzyme immobilization, and as a solvent for drug development.

Supercritical fluids: Supercritical fluids are gases that are compressed to a liquid-like state. They have unique properties that make them useful for green chemistry, including high diffusivity and low viscosity. Supercritical CO2 is used in life sciences research for the extraction of natural compounds from plant material, and as a solvent for the production of nanoparticles for drug delivery.

Enzymes: Enzymes are biocatalysts that can carry out reactions in a sustainable way. They are renewable, biodegradable, and have low toxicity. Enzymes have been used in life sciences

research for the synthesis of bioactive compounds, the production of biofuels, and for bioremediation.

Green solvents: Green solvents are solvents that have low toxicity, low volatility, and are biodegradable. Examples include water, ethanol, and glycerol. Green solvents have been used in life sciences research for the extraction of bioactive compounds from plant material, and for the synthesis of bioactive compounds.

Flow chemistry: Flow chemistry is a technique where chemical reactions are carried out in a continuous flow system. It offers several advantages over traditional batch methods, including higher efficiency, better control over reaction conditions, and reduced waste. Flow chemistry has been used in life sciences research for the synthesis of bioactive compounds, and for the production of nanoparticles for drug delivery.

These are just a few examples of the many green chemistry methods that are used in life science research.

Pros of green Chemistry in life sciences

There are several benefits of using green chemistry principles in life science subjects, including:

Reduced environmental impact: Green chemistry reduces the use and generation of hazardous substances in chemical production, leading to a reduced environmental impact. This can help to minimize the negative effects of chemical production on the environment, including air and water pollution, and the production of hazardous waste.

Improved safety: By using safer solvents and reaction conditions, green chemistry can help to improve the safety of chemical production in the life sciences. This can reduce the risk of accidents and exposure to toxic chemicals for workers and researchers, as well as the general public.

Sustainable production: Green chemistry emphasizes the use of renewable resources, which can reduce reliance on non-renewable resources and promote sustainable production methods. This can lead to a more sustainable and environmentally friendly production of natural products, pharmaceuticals, and agricultural products.

Cost savings: Green chemistry can lead to cost savings in the long run, as the use of renewable resources, safer solvents and reaction conditions, and reduced waste and pollution can lead to a more efficient production process and lower costs of waste disposal.

Improved public perception: The use of green chemistry principles in life science subjects can improve the public perception of the field, as it demonstrates a commitment to sustainability and environmental protection.

Thus, the use of green chemistry principles in life science subjects can lead to a safer, more sustainable, and more environmentally friendly production process, while also reducing costs and improving public perception.

Conclusion

In conclusion, the application of green chemistry principles in life sciences can provide a range of benefits, including improved environmental sustainability, reduced hazardous waste generation, and improved safety for workers and the public. By using safer solvents and reaction conditions, reducing waste and pollution, and promoting the use of renewable resources, the life sciences can become more environmentally friendly and sustainable. Furthermore, the implementation of green chemistry practices can lead to cost savings in the long term, as well as improved public perception of the field. This can help to build trust and support for the life

sciences, which is crucial for advancing scientific progress and addressing important global challenges, such as food security, climate change, and public health.

Therefore, the integration of green chemistry principles in life sciences is essential for the development of more sustainable and environmentally friendly methods for producing natural products, pharmaceuticals, and agricultural products, as well as for animal research and management. By adopting a more sustainable and responsible approach to chemical production, the life sciences can contribute to a healthier and more sustainable future for all.

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Renal calculi a urological disorder in relation to dietary habit, drinking water sources and hereditary factor, A case Study

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Keywords:
Renal, Urology, Hereditary, Struvite and Cystine.

Abstract:
Kidney stone disorder affecting about 12% of the human population on this earth planet. It is deposition of minerals, crystals of chemicals in excretory system. It classified in five basic types on the basis of chemical composition of stones as- calcium, struvite, uric acid, cystine and drugs induced stones. Minerals and crystals can deposit in one part of or entire excretory system. Calcium stone patients were found most dominant as compared to other types of renal stone cases. The sign and symptoms of renal complains were varied from location of stone in excretory organ and its chemical composition. There are ten patients cases were suffered from renal calculi have been studied and concluded them on the basis of U.V. graphy, diagnostic, treatment methods and other medical reports obtained from them in paper evidences. Reports of kidney stone patients were collected, analysed and discussed with them to conclude the exact causes urological complains at dietary, drinking water sources and hereditary level. Cases were studied with the aid and in presence of urologist. During study of renal patient's cases study, it was found that most of patient are now relief from renal complains by taken allopathy, surgery treatment. While in some cases homeopathic and ayurvedic treatment are affected to clear kidney stone.

Introduction:

Renal calculus has worldwide complain found in human being as well as in species and members of mammalian class. But particularly common in some geographical location such as United states, South Africa India and other east from Asian continent. calcium salts, uric acid, cystine and struvite are basic constituents of most of urinary complains. Out of that calcium stones complains are more common (75-80%) among the all types of kidney stones before stone type of uric acids found in human being [1]. In this era renal calculus complains are increasing with relation to dietary habits, drinking water sources and hereditary factor line. During the research work we have been observed that, kidney complains patients were freely interacted with our investigating team and discussed related to their complains, sign, symptoms and place where they have done diagnosis, treatment methods to cure such complains. But out of all cases studied only 50% patients had been given the permission to express their kidney stones complains data in our research work. There are only few reported evidences based works in homeopathy on removal of calculi to expel with homeopathic medicine has notified in some study work [1]. Current research paper have designed and presented

on the basis of data given by patients with entire information about their dietary habits, hereditary, past history and methods of diagnosis, treatments taken by them. In present research work we are also focused on the status of hormonal coordination, especially the hormones of thyroid and parathyroid glands. Those patients are now relief from urological complains are reported in current paper. And which medicinal treatment methods were suitable to cure the kidney complain have also reported in this research work.

Study Area: In this present research work, we haven't kept the limit of study area to investigate the cases of kidney complains to analyse and conclude them. The data about patients was collected, analysed, concluded and reported as per availability of time of expert's urologists and physical condition of patients. Hence, we can't represent the exact time duration and study area for this research work. But intentionally we had focussed to investigate the cases regarding to this research which belongs to our nearby area.

Materials and Methods:

Data Collection: More than ten patient's data about the kidney stones were collected, analysed and concluded for this research work. Out of that data of some typical cases about the renal complains have been reported. During the present research work our aim was to find out, which treatment line is best to relief from kidneys complains and survive the healthy life. In present research work we are also focused on the patient's endocrinal physiological status and function in especially of thyroid and parathyroid gland.

Case study and reports:

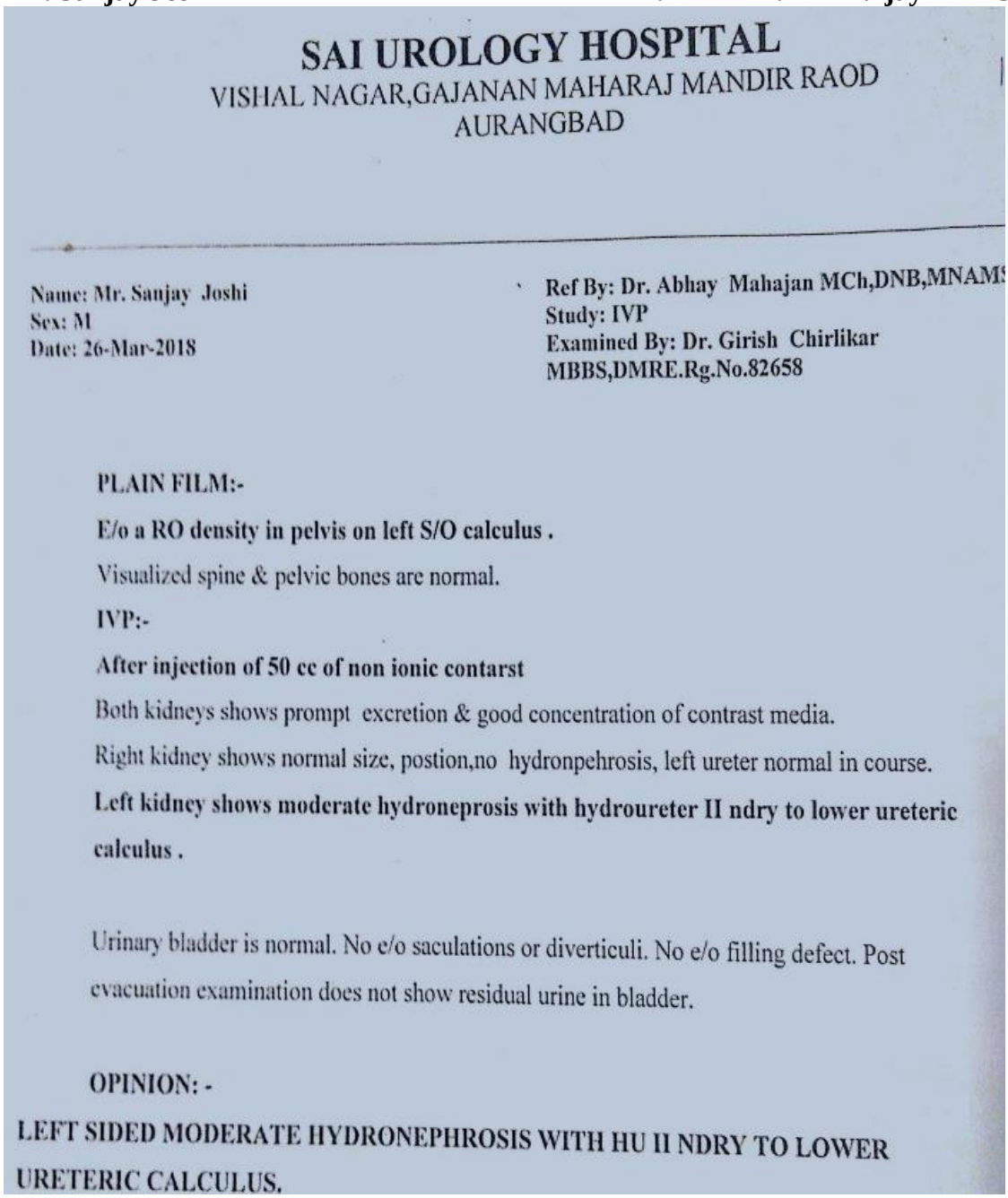
1. Patient details: Mr. Sanjay Joshi: A 50-year-old male came in OPD with symptoms like a sever abdomen pain specially in pelvis region, difficulty in micturition and high fever. Sufferer is married person, good educated and professionally lecturer. During discuss with them, we noticed that he is no smoker and alcohol addicted person. And there is no any hereditary history about kidney complains in his ancestral family members. After the USG report (26/03/2018) it was found that the left kidney shows **moderate hydronephrosis.**

2. Patient details: Mr. Vijay Sanap: A 35 year old male came in OPD with symptoms like an abdomen pain in lower region of dorsal side, difficulty in micturition and high fever. Sufferer is married person, good educated and professionally assistant professor. During discuss with them, we noticed that he is no smoker and alcohol addicted person. And there is no any hereditary history about kidney complains in his ancestral family members. After the USG report (26/07/2016) it was diagnosed that **11mm sized stone found in calyx region of left kidney**.

3. Patient details: **Mr. Narayan Gedam:** A 41year old male came in OPD with symptoms like a abdomen pain and no any fever, nausea. Sufferer is married person, good educated and professionally assistant professor. During discuss with them, we noticed that he is no smoker and alcohol addicted person and do physical exercise daily. there is no any hereditary history about kidney complains in his ancestral family members. After the urography report (10/11/2012) it was diagnosed that right mild hydronephrosis with hydroureter due to a **15.1mm VUJ calculus.**

Reports Evidence: Evidence of diagnostic reports of some typical cases studies are presented:

1. Mr. Sanjay Joshi **2. Mr. Vijay Sanap**



SAI UROLOGY HOSPITAL
VISHAL NAGAR,GAJANAN MAHARAJ MANDIR RAOD
AURANGBAD

Name: Mr. Sanjay Joshi
Sex: M
Date: 26-Mar-2018

Ref By: Dr. Abhay Mahajan MCh,DNB,MNAMS
Study: IVP
Examined By: Dr. Girish Chirlikar
MBBS,DMRE.Rg.No.82658

PLAIN FILM:-

E/o a RO density in pelvis on left S/O calculus .

Visualized spine & pelvic bones are normal.

IVP:-

After injection of 50 cc of non ionic contarst

Both kidneys shows prompt excretion & good concentration of contrast media.

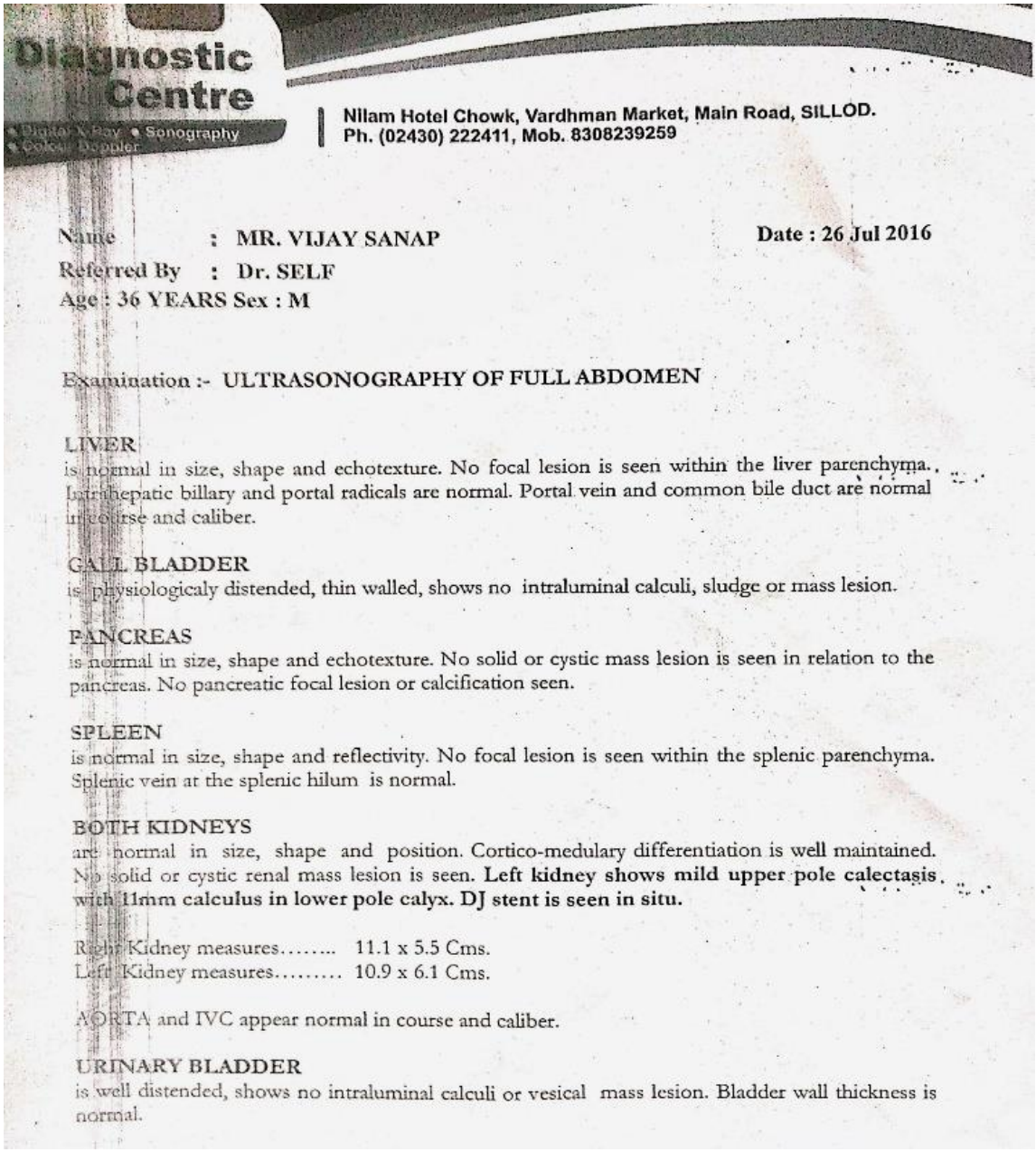
Right kidney shows normal size, postion,no hydronpehrosis, left ureter normal in course.

Left kidney shows moderate hydroneprosis with hydroureter II ndry to lower ureteric calculus .

Urinary bladder is normal. No e/o saculations or diverticuli. No e/o filling defect. Post evacuation examination does not show residual urine in bladder.

OPINION: -

LEFT SIDED MODERATE HYDRONEPHROSIS WITH HU II NDRY TO LOWER URETERIC CALCULUS.



Diagnostic Centre

Digital X-Ray • Sonography
Color Doppler

Nilam Hotel Chowk, Vardhman Market, Main Road, SILLOD.
Ph. (02430) 222411, Mob. 8308239259

Name : MR. VIJAY SANAP Date : 26 Jul 2016

Referred By : Dr. SELF

Age : 36 YEARS Sex : M

Examination :- ULTRASONOGRAPHY OF FULL ABDOMEN

LIVER
is normal in size, shape and echotexture. No focal lesion is seen within the liver parenchyma. Intrahepatic billary and portal radicals are normal. Portal vein and common bile duct are normal in course and caliber.

GALL BLADDER
is physiologicaly distended, thin walled, shows no intraluminal calculi, sludge or mass lesion.

PANCREAS
is normal in size, shape and echotexture. No solid or cystic mass lesion is seen in relation to the pancreas. No pancreatic focal lesion or calcification seen.

SPLEEN
is normal in size, shape and reflectivity. No focal lesion is seen within the splenic parenchyma. Splenic vein at the splenic hilum is normal.

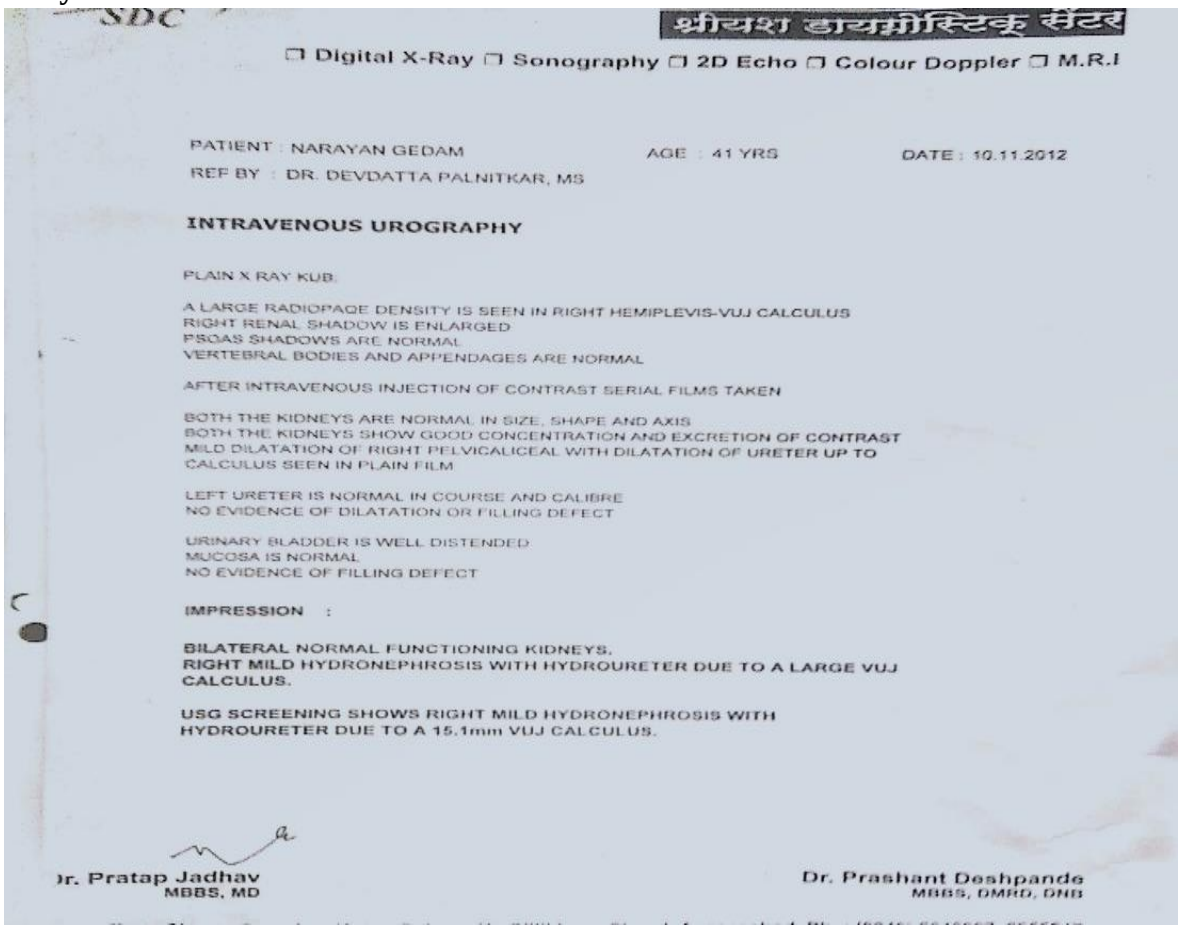
BOTH KIDNEYS
are normal in size, shape and position. Cortico-medulary differentiation is well maintained. No solid or cystic renal mass lesion is seen. **Left kidney shows mild upper pole calectasis with 11mm calculus in lower pole calyx. DJ stent is seen in situ.**

Right Kidney measures..... 11.1 x 5.5 Cms.
Left Kidney measures..... 10.9 x 6.1 Cms.

AORTA and IVC appear normal in course and caliber.

URINARY BLADDER
is well distended, shows no intraluminal calculi or vesical mass lesion. Bladder wall thickness is normal.

3. Mr. Narayan Gedam



श्रीयश डायग्नोस्टिक सेंटर

❒ Digital X-Ray ❒ Sonography ❒ 2D Echo ❒ Colour Doppler ❒ M.R.I

PATIENT : NARAYAN GEDAM AGE : 41 YRS DATE : 10.11.2012
REF BY : DR. DEVDATTA PALNITKAR, MS

INTRAVENOUS UROGRAPHY

PLAIN X RAY KUB.

A LARGE RADIOPAQE DENSITY IS SEEN IN RIGHT HEMIPLEVIS-VUJ CALCULUS
RIGHT RENAL SHADOW IS ENLARGED
PSOAS SHADOWS ARE NORMAL
VERTEBRAL BODIES AND APPENDAGES ARE NORMAL

AFTER INTRAVENOUS INJECTION OF CONTRAST SERIAL FILMS TAKEN

BOTH THE KIDNEYS ARE NORMAL IN SIZE, SHAPE AND AXIS
BOTH THE KIDNEYS SHOW GOOD CONCENTRATION AND EXCRETION OF CONTRAST
MILD DILATATION OF RIGHT PELVICALICEAL WITH DILATATION OF URETER UP TO CALCULUS SEEN IN PLAIN FILM

LEFT URETER IS NORMAL IN COURSE AND CALIBRE
NO EVIDENCE OF DILATATION OR FILLING DEFECT

URINARY BLADDER IS WELL DISTENDED
MUCOSA IS NORMAL
NO EVIDENCE OF FILLING DEFECT

IMPRESSION :

BILATERAL NORMAL FUNCTIONING KIDNEYS.
RIGHT MILD HYDRONEPHROSIS WITH HYDROURETER DUE TO A LARGE VUJ CALCULUS.

USG SCREENING SHOWS RIGHT MILD HYDRONEPHROSIS WITH HYDROURETER DUE TO A 15.1mm VUJ CALCULUS.

Dr. Pratap Jadhav
MBBS, MD

Dr. Prashant Deshpande
MBBS, DMRD, DNB

Discussion: Renal calculus is concretion of crystals of chemicals in any one organ or entire excretory system. The crystals of chemicals once deposited that adhere to another crystal particle to causes calculus in excretory organs. Due to stone formation, there is impairing, altering the basic function of nephron, calyx, pelvis, ureter, bladder or urethra in excretory system. Renal calculi as an important one challenging clinical problem [1] The patients details parameters like age, sex, dietary habit, hereditary factor, past history, diagnostic and treatment methods are basic aid and keys are studied to analyses the aim of present research work. The collected data is helpful to find out the treatment line is best to relief from stone complains permanently. During the investigation we find out that the patient treated with allopathy and surgery methods can't avoid recurrence and its effects permanently. And they have desired to take treatment other than allopathy for to avoid its regeneration and relief from complain permanently. The kidney stone (Ashmari) can be cure by Ayurvedic treatment and medicine without going to surgical invention and avoid to recurrences of stone [2] The methanolic extract of plant *Biophytum sensivitum* (MSB) is help full to prevent and recurrence of stone in rats [3]. Calcium oxalate is most common types of urinary stone and stone under 5mm size passed spontaneously found in 90% kidney stone cases studied [4]. Caffeine contains beverages like coffee, tea contain high level of oxalate a common component of kidney stone. High sodium intake is responsible to increase the calcium reabsorption by uriniferous tubules. Diet plays an important role to cause the kidneys complains especially in patients who are suffering from excretory complains. According to World Health Organisation (WHO) the daily the consumption of salt should not exceed 5 grams or one tea spoon .but average Indian salt

consumption is 9 to 10 grams per day [5]. Long term pharmacological therapy and its potential side effects often lead to subsequent failure. Hence nutritional management is the prevention against urolithiasis [6]. During comparative case study work we find out that some kidneys complain sufferer was in search of ayurvedic, homeopathic and other local treatment to cure. We noticed that some patients haven't the desire to take treatment belongs to ayurveda and homeopathy, it might be due to their lack of awareness or faithful vision on these treatment line. All the cases of kidney patient studied have good physiological and functional status of thyroid and parathyroid gland but some are arrested with diabetic mellitus complain. It is our hypothesis that urinary abnormalities are the result of cellular dysfunction, some inherited, endogenous, extrinsic and environmental factors [7]. Chronic patient of renal calculi has been discussed with our investigating team and given their opinion and experiences about their kidney complain as-ayurvedic and homeopathic treatment is most suitable to expel out renal stone permanently with its recurrences. Homeopathy may be used as safe alternative to surgical intervention when stone isn't in large size have been reported in some research papers.

Discussion out Put: The cases of chronic renal patients was interacted with us and given their opinion and experience of treatment taken by allopathy, surgery and thereafter ayurveda, homeopathy and other local treatment line. Mr. Sanjay Joshi, Mr. Vijay Sanap and Mr. Narayan Gedam are completely relief from any renal complains and its recurrences since last three years by ayurvedic, homeopathy and local treatment methods. These are only few cases reported evidences-based works for to find out exact line of treatment to cure stone permanently, without any medicinal side effects. however, in future we need to study the more comparative data about the renal patients. Which would be the helpful to conclude, find out exact treatment line for kidney complains, its recurrence in life span. However medical evaluation and prophylaxis may not be cost effective in patients who from stones less than one every three year [4]. Steroids are also found to be useful as medicinal expulsive agents in distal uretic stones, steroids reduces the inflammation and neutrophils induced damaged [8]. A recommendation of more than 2 l water per day is good for stone former and should have maintain urine output at least 2 l per day [9].

In present study work it was found that in some hard and labour working kidney patient may not maintain their medicinal, dietary management advised to them and unable to avoid the recurrence of renal calculus. In fact, these listed and reported studied cases had primary treated, operated by allopathy method. unfortunately, they can't relief and avoid the recurrence of stone complain permanently as compared to ayurveda, homeopathy and other local treatment methods. Homeopathy may be used as a safe alternative to surgical intervention especially when the calculi are not of very large size or staghorn variety [1]. In pets animals like dogs and cats the struvite stone is most common as compare to follow by urate, calcium oxalate stones, hence in future our target is that to research on herbal plants with anti-urolithiasis properties [10].

Conclusion: The exact preventive measures for calcium stone formation still not available to increase the awareness in man, however increased fluid intake to maintain urine output of about 2-3 l per day, avoiding to intaking of protein content enriched in animal meat, nuts, dairy products and other sources like induced drugs may be improve the decrease in degree of amino acids, uric acids in blood plasma. Patients with hyperglycaemia and hypertension have more chances for causing of renal complains. Oxalate is found in nuts, beans, tomatoes, chocolates, Coldrinks and tea may increases the risk of kidney stone. Concretion of stone in excretory system might be associated with imbalanced secretion of endocrine glands. The level of hormones secreted by thyroid and parathyroid gland strongly related with excretion, deposition of calcium and other salts in kidneys and assisted organ in excretory system. The successful outcome of our study is that very few

patients followed and maintain the medicinal, dietary management advised by their urologist. We find out that sufferer those doing labour and hard work daily may not maintain medicinal, dietary management advised to them and unable to avoid the recurrence of renal calculus. Hence in some studied cases, we can't overestimate the exact treatment for renal calculus.

Acknowledgement: The authors are thankful to the Principal, Yeshwantrao Chavan college of Arts, commerce and science for providing research facilities to carry out study work. Authors are also thankful to the urologist and patients Mr. Sanjay Joshi, Mr. Vijay Sanap and Mr. Narayan Gedam for providing necessary data. Special thanks to Dr. Datta kakde for providing necessary honest cooperation.

Conflict of Interest: The authors declared that they have no conflict of interest.

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Impact of Heavy Metals (mercuric chloride, copper sulphate and zinc sulphate) on Survivability of Earthworms

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Keywords:
Perionyx excavatus, Heavy Metals, toxicity

Abstract:
Earthworms play a major role in enhancing the physical, chemical, and biological properties of soil. They are considered as keystone species within ecosystem and are used extensively as bio indicators of environmental contamination. The survival rate of earthworm gradually decreased in all media with an increase in concentration of heavy metals. The observed lethal dose (LD50) for 24, 48, 72 and 96 h were ascertained to be 9, 6, 4 and 3 mg/100 g soil respectively of mercuric chloride. 59, 35, 18 and 10 mg / 100 g of soil respectively of copper sulphate and 140, 70, 35 and 17 mg per 100 g of soil respectively of zinc sulphate. The graphically ascertained lethal dose (LD50) values with the help of regression analysis for 24, 48, 72 and 96 h were found to be 9.509, 6.13, 4.798 and 3.017-mg/100 g soil for mercuric chloride, 59.533, 35.343, 18, 0904 and 10.339 mg/100g of soil for copper sulphate and 140.012, 70.3217, 35.0017 and 17.8053-mg/100 g of soil for zinc sulphate. The present study revealed that survival of worms *Perionyx excavatus* declined with an increase in the proportion of concentration added and with exposure time. Heavy metals impose negative impact on growth of earthworms.

Introduction

Earthworms are ubiquitous animals living in soils, enhancing chemical and physical properties and the distribution and activity of microbes and soil animals. In addition, it has been reported that after earthworm activity, the fraction distribution of heavy metals Cu, Zn, Cr, Cd and Pb is changed significantly, thus affecting the bioavailability of these metals Wen *et al.,* (2004). Over 80 % of the biomass of terrestrial composed of earthworms. They play a vital role in the decomposition of organic matter in soil and form an important link in the food chain by channelizing waste energy from dead and decaying organic matter to higher tropic levels by being a prey for terrestrial vertebrates and birds. Through their feeding, burrowing and casting activities, they also improve soil quality nutrient uptake plant growth, and plant yield. Earthworms have a particularly intimate contact with the soil, consuming large quantities of soil and having few external barriers to the soil solution. For these and other reasons, earthworms have been used extensively in ecotoxicological soil studies Sivakumar (2015). Earthworms are one of the important soil macro invertebrates and they function as consumers, decomposers, soil modulators, and food resources for other animals.

Earthworms ingest the metals through feed or by dermal uptake which are known to negatively affect the physiological functions. Heavy metal is a widely distributed metalloid

having significant risk factors for cancer when exposed to contaminated environment Shefali and Yadav (2018). Earthworms are an essential part of the soil fauna in most global soils, represent a significant proportion of the soil biomass and are regarded as a useful indicator of soil health and quality. It is their important role in the decomposition of organic matter and subsequent cycling of nutrients that has led to their use as an indicator organism for the biological impact of soil pollutants and this in turn has lead to a large body of work on earthworm ecotoxicology. Earthworms are also often the subject of inoculation programmes during the restoration of degraded lands and inoculation of earthworms to metal-contaminated soils has been suggested largely due to the role earthworms are known to play in soil formation at such sites Sizmur and Hodson (2009). Heavy metals are present in soils as natural components or as a result of human activities. Industrial, mining and agricultural activities have in some cases produced considerable metal pollution of soils, which is increasingly becoming a serious environmental problem.

Previous studies have shown that the presence of excess heavy metals in the soil, is leading to increased mortality of the worms Mostafaii *et al.,* (2016). Mortality is a parameter used to evaluate the LC50, i.e. the concentration which proves lethal for 50% of exposed individuals. In the literature review, the median lethal concentration was assessed for various metals using various earthworm species and substrates at different time periods from 24 h to 56 days Sivakumar (2015). The effects of metals on the growth and reproduction of earthworms, as well as their behavioural and morphological symptoms and their avoidance behaviour due to metal exposure. Statistical analysis is an essential tool for the interpretation of ecotoxicity test results. Estimations of the ecotoxicological impact of pollutants are usually expressed either as a concentration causing 50 % mortality (lethal concentration, LC50) and effects on the growth and reproduction of the tested organisms (effective concentration, EC50) or as the highest concentration tested not showing any effects on growth and reproduction compared to controls (no observed effective concentration, NOEC).

The earthworm *Perionyx excavatus* is an indigenous, epigeic earthworm widely recognized as a model test organism for ecotoxicological risk assessment of polluted soil. The term epigeic in Greek stands for "upon the earth." These compost worms don't create permanent burrows underground and spend most of the time above at upper soil surface. They are phytophagous worms and are efficient bio-degraders.

The LC50 test using the earthworm *Perionyx excavatus* has been important for risk assessment and regulation of new and existing chemicals (Becker et a/., 1992). The end point of the 'earthworm acute toxicity test' is mortality. However, mortality is unlikely to be either the most sensitive or ecologically relevant parameter for predicting effects on field populations. The standard earthworm acute toxicity test (OECD, 1984) has been used to determine the concentrations of *mercuric chloride, copper sulphate* and *zinc sulphate* that cause specific lethal and sub-lethal effects in the earthworm *Perionyx excavatus*.

Material and methods:

The megascolecid earthworm *Perionyx excavatus* having approximately equal size (10 cm in length) and weight (3 g) were exposed for 5 days separately to lower and higher sublethal concentrations of heavy metals. Soil was collected from an upland non-irrigated field, which had no record of input of agrochemicals (fertilizers and pesticides). The soil had the following characteristics: Laterite type, sandy loam texture, pH-6.8, organic matter 2.7 g%, nitrogen 0.22 g% and a C/N ratio of 12.27. The soil was air dried and sieved before use. The earthworms were also collected from the above characterized field. They were cultured in their habitat soil and acclimated for one month with adequate provision of food (10% organic matter, cow dung+leaf

litter), moisture (20g %) and temperature (25+2°C). Earthworms were removed from culture pots and kept half immersed in glass petriplates containing 30ml of tap water in 25+2°C temperature for 24 hours to evacuate their guts (Dash and Patra, 1977). The study was carried out in plastic culture pots under laboratory conditions following the protocol of Panda and Sahu (2002). In brief, three heavy metals namely mercuric chloride ($HgCl_2$), Copper sulphate ($CuSO_4$) and Zinc sulphate ($ZnSO_4$) respectively used as the test chemicals were obtained from Ranbaxy Chemicals Ltd. The pesticides were chosen on the basis of their extensive use in this area.

Toxicity tests using of adult (10cm length and 3 gm in weight) *Perionyx excavatus* were conducted for 24 to 96 hours with different concentrations of each heavy metal (mg/kg dry soil equivalent) applied to the soil as per the method used by Dash and Patra (1977). The concentration were prepared in dilution of acetone and sprayed on the soil surface. After evaporation of the solvent, the treated soil was thoroughly mixed to distribute the pesticide evenly and enough water was added to bring the moisture content up to the field capacity. The same procedure with pure acetone was applied to prepare the controls. Ten healthy gut cleared earthworm were added to each pot. The experiment was maintained at 20 % soil moisture at 250 +2°C soil temperatures. Earthworm deaths were recorded and probit method of Finney (1971) was followed to calculate LD50 value for adult earthworm, Panda and Sahu (2002). Calculation of the Regression line: Assessment of median lethal dose LD50

The experiment was carried out for finding the range of concentrations for confirmatory evaluation. The mortality was recorded for earthworm *Perionyx excavatus* at 24, 48, 72 and 96 h exposure to mercuric chloride, copper sulphate and zinc sulphate, were corrected for natural response by Abbott's formula (Abbott, 1925).

Abbott's formula:

$$\text{Corrected mortality\%} = \frac{\text{Percentage living in control-Percentage living in treatment}}{\text{Percentage living in control}} \times 100$$

The corrected mortality data was analyzed following the method of Finney (1971) to determine the LD50 values. The LD50 values were obtained by probit regression line, taking test concentration and corresponding percent mortalities on log value and probit scales respectively. Straight line (regression line) was drawn between the points, which represent the survival percentage versus concentration (APHA, 1989). From the points at which this line intersects the 50 percent survival line, a perpendicular line drawn to the concentration ordinate, indicates the LD50 dose of that particular period. By graphical interpolation LD50 values were fixed and their fiducial limits 95% upper and lower confidence limits were also calculated.

Results:

Results obtained from the present investigation clearly shows at the earthworm *Perionyx excavatus* survived well from 1 to 6 mg /100 gm soil for 24 h, 1 to 4 mg/100 gm soil for 48h, 1 to 3 mg /100mg soil for 72 h, 1 to 1.5 mg /100 gm soil for 96 h for mercuric chloride. They also survived well from 20 to 55 mg/100 mg of soil for 24 h, 20 to 30 mg /100 gm of soil for 48 h, 10 to 15 mg/100 gm of soil for 72, 5 to 10 mg /100gm of soil for 96 h for copper sulphate and 100 to 180 mg /100 gm of soil for 24 h, 50 to 60 mg /100 gm of soil for 48 h, 20 to 30 gm of soil for 72h and 10 to 15 mg/100 gm of soil for 96 h respectively for zinc sulphate.

The survival rate of earthworm gradually decreased in all media with an increase in concentration of heavy metals. The observed lethal dose (LD50) for 24, 48, 72 and 96 h were

ascertained to be 9, 6, 4 and 3 mg/100 g soil respectively of mercuric chloride. 59, 35, 18 and 10 mg / 100 g of soil respectively of copper sulphate and 140, 70, 35 and 17 mg per 100 g of soil respectively of zinc sulphate. The graphically ascertained lethal dose (LD50) values with the help of regression analysis for 24, 48, 72 and 96 h were found to be 9.509, 6.13, 4.798 and 3.017-mg/100 g soil for mercuric chloride, 59.533, 35.343, 18, 0904 and 10.339 mg/100g of soil for copper sulphate and 140.012, 70.3217, 35.0017 and 17.8053-mg/100 g of soil for zinc sulphate.

Discussion:

Exposure to pollutants, even in exceedingly low concentrations, can elicit behavioural responses in earthworms. Furthermore, because of their visibility, morphological abnormalities can readily serve as definitive evidence of adverse effects. The term metal toxicity or metal poisoning refers to the toxic effects of certain heavy metals in certain forms and doses on living organisms. Thus, ranges of toxicity test have been proposed to assess the potential hazards of pollutants to earthworms. In the present study as per OECD (1981), statistically calculated values for the lethal concentration causing 50% mortality (LC50) of earthworm *Perionyx excavatus* by mercuric chloride were found to be 9mg, 4mg, 3mg and 2 mg/100gm of soil for 24, 48, 72 and 96 h. For copper sulphate LC50 values were found to be 67mg, 36mg, 18mg and 10 mg / 100 gm of soil for 24, 48, 72 and 96 h. LC50 values of zinc sulphate were computed as 166mg, 87mg, 49mg and 23 mg /100 gm of soil for 24, 48, 72 and 96 h.

The present study revealed that survival of worms *Perionyx excavatus* declined with an increase in the proportion of concentration added and with exposure time. Heavy metals impose negative impact on growth of earthworms. Earthworms are the potential bio-indicators of heavy metal contamination. Earthworms can act as bio-indicators for heavy metal pollution as the survivability of earthworms indicated declining trends with the increased concentrations of heavy metals Parihar *et al.,* (2019). However, by minimizing the heavy metal contamination in soil the earthworm population density may be enhanced. The toxicity values indicate that mercuric chloride is more toxic than copper sulphate and zinc sulphate.

The study were investigating the impact of heavy metals (mercuric chloride, copper sulphate, and zinc sulphate) on the survivability of earthworms, the significance of the findings would likely relate to understanding the toxicity of heavy metals and how they affect the physiology and survival of earthworms. One of the main significance of this research would be to understand the mechanism of heavy metal toxicity and how it affects the survival rate of earthworms. Earthworms play a crucial role in soil ecology and are considered bioindicator species, meaning changes in their survival rate can indicate changes in the overall health of the soil. Therefore, understanding the effects of heavy metal contamination on the survival rate of earthworms can provide insights into the impact of heavy metal pollution on soil health.

Additionally, the study may find that heavy metal exposure leads to changes in the survival rate of earthworms, and this information can be used as a guide for developing strategies for mitigating the effects of heavy metal pollution on soil ecosystems. It may also give an insight into which heavy metals are more toxic to the earthworm species used in the study. The study may also find that different heavy metals have different effects on the earthworm survival, which can help to understand which heavy metals are more toxic and which have a lesser effect on the earthworm survival. Finally, earthworm survival rate is a well-known indicator of the overall soil health, so if the study found that earthworm survival rate changes with heavy metal exposure, it may be possible to use it as a bio indicator of heavy metal pollution in soil.

Behavioural responses and morphological symptoms have been recorded so far are as follows: failure to burrow, curling of the body, violent coiling, slow and swift movement,

shortening of the body, elongation of the body, oozing out of coelomic fluid, preclitellar swelling, body constriction and segmental bulging Sivakumar (2015). In general, earthworms are effective accumulators of metals from the soils leading to compartmentalization, storage, or excretion of metal ions from the most sensitive tissues.

They apparently have well developed, specific trafficking and storage pathways and redistribution capacity to regulate heavy metals, especially essential trace metals, in their bodies that may lead to balance between uptake and excretion. The regulatory capacity of metals can partly explain the ability of some earthworm species to live even in highly metal contaminated areas. Moreover, metal body did not increase with increasing soil metal concentrations, not even with these timated bioavailable fractions of the metals. Thus, the metal regulation may also have contributed to the development of metal resistance observed in some earthworm populations Lukkari *et al.,* (2004). Harmfulness of certain metal concentrations measured from the earthworms may be difficult to evaluate. Especially in the field, body burdens affecting survival, growth, and reproduction of earthworms seem to be site and species-specific. In addition, responses of earthworms to metal contamination are modified by several other environmental factors Latha & Mahaboob (2016).

Conclusion

The conclusion of a study on the impact of heavy metals (mercuric chloride, copper sulphate, and zinc sulphate) on the survivability of earthworms would summarize the main findings of the research, discussing how exposure to these heavy metals affected the survival of the earthworms. It would also highlight any notable trends or patterns that emerged from the study, such as whether one heavy metal had a more pronounced effect on earthworm survival than the others, or whether there was any dose-response relationships observed. Additionally, the conclusion would relate the results to other existing studies on the topic, and discuss the ecological and environmental implications of the findings.

It could be like: "The results of this study demonstrate that exposure to mercuric chloride, copper sulphate, and zinc sulphate heavy metals have negative effects on the survival rate of earthworms. The study reveals that mercuric chloride has the highest toxic effect among the heavy metals used in this study. The findings of this research have ecological and environmental implications as earthworms play an important role in ecosystem functioning. Further research should be conducted to better understand the mechanisms of heavy metal toxicity in earthworms, as well as to evaluate the potential long-term impacts of heavy metal pollution on earthworm populations."

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REVIEW ARTICLE

Fungal Laccase: Applications, Screening and Optimization of cultural condition for laccase production.

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Fungal Laccase: Applications, Screening and Optimization

Abstract:

In the recent years, enzymes have gained great importance in industries. Laccases are one of them which are widely present in nature. Laccases are found in plants, insects and bacteria, but the most important sources of these enzymes are fungi. Most of the laccases studied are of fungal origin especially from the white-rot fungi class. Fungal laccases play an important role in plant pathogenesis, pigment production and degradation of lignocellulosic materials. Application of laccase includes Textile dye decolourization, Bioremediation, Food industry, Pulp and paper industry, Pharmaceutical industry and Cosmetic Industry. Laccase production may be affected by fermentation factors such as, medium composition, nutrient sources, pH and temperature. This paper reviews the application, function, screening and optimization of cultural condition for laccase production.

1.Intoduction:

Laccase:

Laccases are the oldest and most studied enzyamatic systems (benzenediol:oxygen oxidoredutases, EC 1.10.3.2). Yoshida in 1883 first described laccase from the exudates of the Japanese lacquer tree, *Rhus vernicifera* and in 1896 laccase was demonstrated to be present in fungi for the first time by Bertrand and Laborde **(**Thurston, 1994).

Laccase is an enzyme that has potential ability of oxidation. These enzyme catalyzes one electron oxidation of wide variety of organic and inorganic substrates including mono-, di- and polyphenols, aminophenols, methoxyphenols, aromatic amines and ascorbate with concomitant four electron reduction of oxygen to water (Vernekar and Lele, 2009). There are several compounds that have been used as substrates for laccase *viz*. 2,2'-azinobis-(3-ethylbenzthiazoline-6-sulfonate) (ABTS), tannic acid, syringaldazine, o-dianisidine, guaiacol, dimethoxyphenol and naphthol.

2. Sources of laccase:

There are diverse sources of laccase producing organism like fungi, plants, bacteria and insects. Laccases were reported from plants include Lacquer, Mango, Mung bean, Peach, Pine,

Prune and Sycamore (Arora and shamra 2010). In fungi it is present in Ascomycetes, Deuteromycetes and Basidiomycetes and abundant in lignin-degrading white-rot fungi. It is found very commonly in white rot fungus (Shraddha *et al.,* 2011). Laccase has been reported only in few bacteria including *Azospirillum lipoferum, Marinomonas mediterranea, Streptomyces griseus, Bacillus subtilis* and *Streptomyces lavendule* (Suzuki *et al.,* 2003). Laccase is also found to be present in dozen of insects genera that include *Bombyx, Calliphora, Diploptera, Drosophilla, Lucilia, Monduca, Musca, Orytes Papilio, Phormia, Rhodnius, Sarcophaga, Schistocera* and *Tenebrio* (Xu 1999; Arora and Sharma 2010).

3. Functions of lacccase:

Laccases carry out vital role during fungal life cycle including morphogenesis, fungal plant pathogen/ host interaction, stress defence and lignin degradation, delignification, pigmentation, fruiting body formation, pathogenesis and protection from toxic phenolic monomers of polyphenol (Thruston 1994; Fatima *et al.,* 2015). In plants, laccases have been found in the wood and cellular walls and carry out lignin biosynthesis (Giardina *et al.,* 2010). Bacterial laccases appear to have a role in morphogenesis, in the biosynthesis of the brown spore pigment and in the protection afforded by the spore-coat against UV light and hydrogen peroxide, and in copper homeostasis. The main function of the laccase-type proteins in insects is believed to be sclerotization of the cuticle in the epidermis (Giardina *et al.,* 2010). Fungal laccases have higher redox potential than bacterial or plant laccases.

4. Applications of Laccase:

4.1 Food Industry:

Laccase used in the food industry based on polymerization ability. They can be applied to certain processes that enhance or modify the colour appearance of food or beverage for the elimination of undesirable phenolics, responsible for the browning, haze formation and turbidity in clear fruit juice, beer and wine (Shraddha *et al.,* 2011). They are also employed to ascorbic acid determination, sugar beet pectin gelation, baking and in the treatment of olive mill wastewater.

4.2 Pulp and paper Industry:

Laccases are able to depolymerize lignin and delignify wood pulps, kraft pulp fibers and chlorine-free in the biopolpation process. Laccases can be used for binding fiber-, particle- and paper-boards Brijwani *et al.* (2010).

4.3 Textile Industry:

Different chemicals are used and most of them are difficult to decolourise due to their synthetic origin. Conventional processes to treat dye wastewater are ineffective and not economical. Therefore, the development of processes based on laccases seems to be an attractive solution due to their potential in degrading dyes of diverse chemical structure, including synthetic dyes currently employed in the industry Brijwani *et al.* (2010). It is reported that laccase-mediator system finds potential application in enzymatic modification of dye bleaching in the textile and dyes industries.

4.4 Bioremediation:

Laccases are involved in green biodegradation due to its catalytic properties. They could be used for decolorizing dye house effluents that are hardly decolorized by conventional sewage treatment plants. Immobilized laccase was found to be useful to remove phenolic and chlorinated phenolic pollutants. Laccase mediator systems have been also used to oxidize alkenes, carbazole, N-ethylcarbazole, fluorene, and dibenzothiophene (Brijwani, *et al.,* 2010).

4.5 Pharmaceutical Industry:

Laccases are very specific and bio-based in nature having potential applications in the pharmaceutical sector. These enzymes had been used by pharma companies for the synthesis of

complex medical compounds such as anesthetics, anti-inflammatory, antibiotics and sedatives. One potential application is laccase-based *in situ* generation of iodine, a reagent widely used as disinfectant. Recently laccases also reported to possess significant HIV-1 reverse transcriptase inhibitory activity (Alpeshkumar and Shiroya 2004; Wang and Ng 2004).

4.6 Cosmetic Industry:

More recently laccases have been exploited for preparation of cosmetics. Cosmetic and dermatological preparations containing proteins for skin lightening have been developed. Recently developed laccase-based hair dyes could be less irritant and easier to handle than current hair dyes. Laccases may find use as deodorants for personal-hygiene products, including toothpaste, mouthwash, detergent, soap, and diapers. Protein engineered laccase may be used to reduce allergenicity (Wang and Ng 2004).

5. Screening of laccase producing fungi:

There are several substrates used for laccase screening *viz*. 2,2'-azinobis-(3-ethylbenzthiazoline-6-sulfonate) (ABTS), tannic acid, syringaldazine, o-dianisidine, guaiacol, dimethoxyphenol and naphthol.

Arora and Sandhu (1984) screened different fungi for their laccase producing ability by using different indicator compound and reported that tannic acid was best substrate for laccase production from *Trametes hirsuta* as compare to lignins, phenolic compounds and sugar tested as substrates and concluded that tannic acid was efficient laccase producing substrate. Kiiskinen *et al*., (2004) carried out plate test screening based on polymeric dye compounds and reported that guaiacol and tannic acid is the efficient substrates for laccase production. Buddolla *et al*., (2008) tested 12 different fungal cultures using guaiacol indicator compound and reported that six cultures were found to be laccase positive with *Stereum ostrea* and *Phanerochaete chrysosporium*.

Jhadav *et al*., (2009) used guaiacol for quantitative screening and reported that *Phanerochaete chrysosporium* is efficient laccase producing fungi. Sathiyavathi and Parvatham (2011) screened 12 fungal strains for their ability to produce laccase and xylanase by plate screening method using the indicators guaiacol and congo red respectively and reported that among the 12 isolates, *Trichoderma* sp. was predicted to be the only strain to produce both laccase and xylanase.

Desai *et al*., (2011) isolated 9 different fungal strain from soil and tree bark and screened this fungal strain on PDA medium using guaiacol and tannic acid as an indicator and reported that among this one strain showed potency to produce laccase and it belongs to genus *Trichoderma*. More *et al*., (2011) determine laccase activity using ABTS as a substrate and purified laccase by ion exchange and gel filtration chromatography. Vaidyanathan *et al*., (2011) studied screening and induction of laccase activity in fungal species and its application in dye decolorization. Ten fungal species were screened for laccase activity by indicator plate method (0.02% guaiacol and 0.1% syringaldazine) out of ten only five species were found to be laccase-positive. *Pleurotus ostreatus* gave the highest laccase activities followed by *Agaricus bispora* and other strains.

Christie and Shanmugam (2012) studied productions of laccase enzyme from four Ascomycetes species by in silico and in vitro analyses and reported that crude enzyme showed complete oxidation of ABTS and Guaiacol after 7 days of incubation. *Alternaria arborescence* showed maximum production of enzyme followed by *Fusarium oxysporium*. Aslam *et al*., (2012) studied screening of laccase from *Cladosporium cladosporioides* and reported that *Cladosporium cladosporioides* was efficient laccase producer when grown on malt extract media supplemented with 0.02% ABTS and 0.02% guaiacol. Singh and Abraham (2013) isolated laccase producing fungi

from compost soil and characterize them. 11 fungal isolates were isolated by serial dilution and they were cultivated on potato dextrose agar plate with indicator compound ABTS to screen for the laccase production ability. Out of 11 isolates, one was presumed to be potent, another showed medium potency, and three showed weak laccase producing ability.

Daphne and Gnanadoss (2013) used two substrates viz. Guaiacol and ABTS in media to isolate laccase producing fungi. Twenty six fungi were isolated and screened for their ability to produce laccase on solid medium containing guaiacol and ABTS. Six isolates showed positive laccase activity out of that two fungi were quantitatively screened and identified as *Psathyrella condolleana* LCJ 178 and *Myrothecium gramineum* LCJ 177. Kaur and Nigam (2014) in search of active producer of laccase enzyme used different culture media out of tested media, malt extract medium supplemented with substrate ABTS showed maximum laccase production out of other tested media and substrates or indicator compounds. Abeer *et al.,* (2015) carried out qualitative and quantitative assay of marine fungal isolate *Alternaria tenuissima* and reported that it showed highest zone diameter and colony diameter in agar plate screening test with guaiacol as an indicator compound.

Vantamuri and Kaliwal (2015) isolated 150 fungal strain and screen for their ability to produce laccase enzyme using ABTS, tannic acid, guaiacol and syringaldazine as an indicator compounds and showed that among 150 fungi 8 fungal strain shown ability for laccase production. Fatima *et al.,* (2015) screened soil fungi for laccase production by using guaiacol as a substrate and found that *Verticillum sp*. and *Helminthosporium sp.* showed large reddish-brown zone in media due to oxidative polymerization of guaiacol. Sidhu *et al.,* (2017) isolated twenty five lignolytic fungi from decaying wood samples and screened for the laccase production using guaiacol and ABTS as indicator compounds. The positive isolates were confirmed for the presence of laccase using indicators and quantitative enzyme assay in presence of catalase using ABTS as substrate.

6. Optimization of culture condition for laccase production:

6.1. Optimization of proper culture media for laccase production:

Researcher used different medium for laccase production such as; Potato Dextrose Broth, Malt Extract Broth, Glucose Peptone Broth, Glucose Nitrate Broth, Yeast Extract Peptone Dextrose Broth and Czapek Dox Broth.

Manimozhi and Kaviyarasan (2012) concluded that malt dextrose broth and potato dextrose broth was optimal medium for laccase activity and biomass production of *Agaricus heterocystis*. Amutha and Abhijit (2015) studied different media for growth and laccase production from *Trametes versicolor* and observed that yeast glucose broth is suitable for the growth of *Trametes versicolor* at pH 5.5 and temperature 28^0C and Modified Kirk and Ferrell media showed maximum enzyme production in presence of pH 5.5 and temperature at 28^0C. Prasher and Chauhan (2015) carried out an experiment, effect of twelve different basal media for the growth and ligninolytic enzymes activity of *Dictyoarthrinium synnematicum* and reported that the Glucose-peptone medium was found to be an optimum medium for the growth and laccase production of *Dictyoarthrinium synnematicum*. Sidhu *et al.,* (2017) studied laccase producing capacity of fungus estimated in seven different media and reported that Czapek dox medium showed highest enzymatic activity.

6.2. Effect of different nutrient sources on laccase production:

Some researchers observed that the elevated laccase activity was achieved by using low carbon to nitrogen ratio while others observed that it was achieved at high carbon to nitrogen ratio.

Pointing *et al.,* (2000) reported that laccase enzyme production was maximum with high carbon low nitrogen ratio medium. Slight repression of enzyme production was observed in high nitrogen culture medium. Other potential inducers were less effective (Tween 80, wood fibres) and reduced the stimulation observed by xylidine alone when included in the same culture medium. Soden and Dobson (2001) reported that laccase activity in *Pleurotus sajor-caju* is affected by nitrogen and carbon and by the addition of copper and manganese to the growth medium. In addition, 2,5-xylidine, ferulic acid, veratric acid and 1-hydroxybenzotriazole induced laccase activity in the same. Chawachart *et al.,* (2004) tested different substrates with different carbon sources for laccase production using thermo tolerant Basidiomycete *Coriolus versicolor* strain RC3 and concluded that 1% rice bran as a carbon source was found to be the most efficient substrate for laccase production compared to 1% glucose, wheat bran and rice straw meal.

Mikiashvili *et al.,* (2006) studied effects of carbon and nitrogen sources on two strain of *Pleurotus ostreatus* for laccase production and concluded that the maximal laccase yield of *P. ostreatus* 98 and *P. ostreatus* 108 varied depending upon the carbon source from (5 to 62 U l) and from (55 to 390 U l), respectively. Laccase activity of mushrooms decreased with supplementation of defined medium by inorganic nitrogen sources whereas peptone followed by casein hydrolysate appeared to be the best nitrogen sources for laccase accumulation by both fungi. Xavier *et al.,* (2007) studied effects of different inducers for laccase production with *Trametes versicolor* and found that the best result for laccase induction was obtained with solid lignin, a by-product of pulp and paper industry and the higher laccase activity attained was obtained with the combined effect of xylidine addition and glucose suppression.

Agostini *et al.,* (2011) studied effects of carbon and nitrogen ratio on laccase production by *Pleurotus ostreatus*, *Lentinula edodes* and *Agaricus blazei* and reported that higher C/N ratios increased mycelial growth and decreased laccase production. The highest activities were obtained with a C/N ratio of 5. *P. ostreatus*, *L. edodes* and *A. blazei* produced more laccase when ammonium sulphate, combination of ammonium sulphate and urea and only urea respectively. Rajendran *et al.,* (2011) screened different nutrient levels for optimization of laccase production and reported that D (+)-glucose, $(NH_4)\ 2SO_4$ and $MgSO_4.7H_2O$ concentrations did not exert any significant effect on the fungal laccase production whereas in terms of higher order effects, the interaction between D (+)-glucose and $MgSO_4.7H_2O$ plus D (+)-glucose and $CuSO_4.5H_2O$ were found to be significant. $CuSO_4.5H_2O$ supplementation at 0.1 mg L-1 was effective in improving the laccase production by *P. sanguineus* in submerged fermentation.

Periasamy *et al.,* (2011) studied the effect of 6 different nitrogen sources (5 g/L) on laccase production from fungus *Pleurotus ostreatus* IMI 395544. The highest enzyme production (0.36 U/ml) was achieved when using malt extract as the N-source. There was a slight reduction in the production of laccase (0.34 U/ml) was obtained when malt extract was replaced by yeast extract. Less than 0.20 U/ml of laccase activity were obtained when using beef extract and meat extract as N-sources. Sivakami *et al.,* (2012) reported that the addition of carbon source, such as Dextrose, Mannose, Fructose, Sucrose, Glucose and Lactose to the basal medium yielded optimum production. Out of 5 sources dextrose was found to be an effective carbon source for the production of laccase. Increasing the concentration of glucose will leads to the decrease in the yield of laccase from *Pleurotus ostreatus.*

Abdel-Azeem and Salem (2012) tested 60 taxa for laccase production and standardized optimum cultural conditions such as temperatures, pH, carbon sources and nitrogen sources for the production of high extracellular laccase activity with guaiacol as colour indicator. Sixteen isolates showed positive reaction indicating a lignin-degrading potentiality and out of them eight

measured the highest zone diameter with high oxidation scale. The most promising taxa were endophytic namely *Chaetomium globosum*, *Phoma exigua*, *Thanatephorus cucumeris* and *Sordaria fimicola*. The pH 7, incubation temperature 30^0C, 1% maltose and 0.3% peptone supported the highest biomass and laccase production for *Chaetomium globosum*.

Kenkebashvili *et al.,* (2012) studied the effect of a wide range of culture conditions on production of laccase and manganese peroxidase by *Coriolopsis gallica* and reported that copper, carbon and nitrogen sources, and their concentration had a strong impact on enzyme activities. Manimozhi and Kaviyarasan (2012) studied effect of carbon and nitrogen sources on laccase production of *Agaricus heterocystis* and observed that out of tested carbon sources only fructose showed highest fungus mycelial biomass and laccase enzyme production. Nitrogen sources ammonium tartarate and yeast extract showed highest mycelial biomass yield and laccase production. Among the different metal ions tested, only copper sulphate showed highest laccase activity and maximum biomass production.

Elsayed *et al.,* (2012) reported that carbon source soluble starch was found to be the best inducer for laccase production whereas galactose, fructose, maltose and CMC significantly reduced laccase production by *Pl. ostreatus* ARC280. For nitrogen sources the highest level of enzyme production was obtained with ammonium sulphate followed by L-arginine and urea showed lowest enzyme activity.

Nadeem *et al.,* (2014) screened *Pleurotus ostreatus* P1 by plate test on Kirk medium for laccase production by using guaiacol and syringaldazine as indicators and six different liquid culture media and showed hyper laccase production (5.5±0.33unit/ml) objective was achieved in medium 09-CBZ6 in combination with promising protein production (3mg/ml) under optimized fermentation conditions (pH- 5.5, temperature-30°C and Time-7th day). Laccase production by a long way intensified by the adding of glucose (1.5%, C/N:15) and inducer (CuSO4) gave promising results (13.72±0.30U/ml) and other inducers followed laccase production as Na_2SO_4> $ZnSO_4$> FeSO4> $CaCl_2$. Sodium azide was found a significant inhibitor (100%) for laccase activity than SDS (87%) and EDTA (83%).

Vivekanadan *et al.,* (2014) carried out experiments to study effect of various carbon, nitrogen and metal ions on the laccase production by *Aspergillus nidulans* KF974331 and reported that out of carbon sources tested sucrose showed maximum laccase production whereas among different nitrogen sources tested peptone showed positive factor for maximum laccase production. While in case of seven different metal ions tested for laccase production copper sulphate formed a major factor for maximum yield.

El-Batal *et al.,* (2015) concluded that laccase production by *Pleurotus ostreatus* has been shown to depend remarkably on the composition of the culture medium, carbon, nitrogen content and inducer compounds. They indicate that the factorial design can be a practical useful tool for optimizing the reaction parameters for enhancing the activity of laccase.

Afreen *et al.,* (2016) carried out an experiment on laccase production from non-nitrogen fixing cyanobacteria *Arthrospira maxima* (SAE-25780) and concluded that *Arthrospira maxima* (SAE-25780) showed a constitutive production of laccase which increased up to 80% in the presence of inducer compound guaiacol. The optimal condition for laccase production was at 30^0C, 10 mM sucrose as a carbon source, 10 mM sodium nitrate as a nitrogen source and 2 mM copper as metal activator.

6.3. Effect pH and temperature on laccase production:

The effect of pH and temperature is limited in case of laccase production. The optimal value of pH varies according to the substrate because substrate causes different reaction for

laccase. Most of the studies show that pH between 4.5 to 6.0 is suitable for laccase production. The optimal temperature of laccase differs from one strain to another. It has been found that 25^0 to 30^0 is the optimal temperature for laccase production.

Banerjee and Vohra (1991) optimized proper culture conditions for *Curvularia* sp. and concluded that the initial pH 4.0 and temperature 30^0C were found to be significant for maximum laccase production from *Curvularia sp.* Arora and Gill (2005) carried out optimization of proper culture conditions for laccase production from white rot fungus *Phlebia floridensis* and reported that, laccase production and manganese peroxidase production was found to be maximum on pH 4.5. Janusz *et al.,* (2006) concluded that the soil dwelling fungus *Rhizoctonia praticola* represents a new source of extracellular laccase and for the highest production of laccase enzyme, fungus requires a high value of initial medium pH (7.5-8).

Niladevi and Prema (2008) studied optimization of various cultural and nutritional parameters for the production of laccase by *Streptomyces psammoticus* and showed that the maximum laccase yield was attained at pH 7.5 and temperature 32^0C. Rajeswari and Parvatham (2011) standardized optimum cultural conditions for laccase production from *Aspergillus* sp. and reported that pH 4.6 and temperature 40°C were found to be optimum and computed half-life of laccase activity in minutes were 120 (40°C), 60 (50°C) and 35 (60°C) respectively. Christie and Shanmugam (2012) carried out optimization of *Alternaria arborescence* and *Fusarium oxysporium* for laccase enzyme production and showed maximum production of enzyme in *Alternaria arborescence* (800U/l) at 30^0C and 4.5 pH followed by *Fusarium oxysporium* with (600 U/l) at 45^0C and 5 pH after 15 days of incubation.

Manimozhi and Kaviyarasan (2012) isolated *Agaricus heterocystis* and carried out an experiment on optimum temperature and pH for fungal mycelial biomass and laccase activity and reported that the highest yield of biomass was at 30^0C and pH 6.5 whereas laccase activity was found to be maximum at 25^0C and pH 5.5. Fonseca *et al.,* (2013) studied influence of culture conditions on laccase production, growth and isoenzymes patterns in native white rot fungi and concluded that the selected four strains showed attractive and alternative source for laccase production influenced by combined effects of temperature and pH. Potential strain shows highest laccase activity at 29^0C and pH 4.5.

Rathinasamy and Thayumanavan (2014) observed that the growth of the fungus *Pleurotus ostreatus* IMI 395545 and the production of laccase were highly influenced by effective controlling. The factors like pH, temperature and percentage of dissolved oxygen tension (DOT) provision available in the bioreactor. Vantamuri *et al.,* (2015) isolated different fungal isolates and screened them for laccase production with guaiacol as an indicator compound. Out of 9 isolates tested only 2 isolates have the ability to produce laccase. Potent fungus was morphologically identified as *Coprinus comatus*. The laccase activity was highest with sucrose as carbon source and yeast extract as nitrogen source. The highest production of laccase was found to be at pH 5 and the optimum temperature for production was recorded at 30^0C.

Sidhu *et al.,* (2017) reported the optimum pH, incubation temperature and incubation period for the laccase production in submerged culture were found to be pH 6, temperature 30^0C, 7 day incubation period respectively.

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NextGen farming – Hybridization of Agriculture and Technologies for Resource Optimization and High Gain

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Keywords:
NextGen farming, Hybridization of Agriculture and Technologies

Abstract:
Use of technologies in agriculture is not new concept. From ancient time people are using various tools for farming like wooden-animal driven ploughing, cultivations to use of modern mechanical tractors, harvesters. One must admit that all these tools have simplified the agriculture work process in large extent by saving manpower, time and money.

With the advancements in technology, NextGen farming is now possible with Agriculture 4.0 that allows use of robots in crop cultivation, monitoring of crop health, digitized harvesters. This paper discusses technological solutions provided by industry 4.0 revolution that can be used in NextGen farming.

Introduction

Farming is the prominent business in India. It also contributes a significant figure to the Gross Domestic Product (GDP). The Indian agricultural sector is predicted to increase to US$ 24 billion by 2025. It also contributes a significant figure to the National Gross Domestic Product (GDP). Total food grain production in the country is estimated over 149.92 million tonnes for FY 2022-23.

Indian agriculture and allied activities have witnessed a green revolution, a white revolution, a yellow revolution and a blue revolution. During green revolution, various agricultural universities and institutions had worked on research and technology activities that were transferred to the farm and enhanced agricultural productivity in many regions of the world between 1950 and the late 1960s. The white revolution refers to increase in milk production making India as one of the leading a milk producing nations. In 1986-87, India set the goal of increasing edible oil production to reduce the dependency on foreign countries for edible oil giving rise the yellow revolution. During 7th five year plan, Indian government has promoted increase in food products from aquaculture mainly fisheries to improve farmers income. It has been considered as blue revolution.

Now, the most of manufacturing industries are moving towards industry 4.0 standards aiming the use of new technologies in the production and distribution of products.

The Industry 4.0

Industry 4.0 is revolutionizing the way companies manufacture, improve and distribute their products. Producers are incorporating new technologies, including Internet of Things (IoT), cloud computing, data science and AI with machine learning into their production facilities and throughout their operations. This digital technologies lead to increased automation, predictive maintenance, self-optimization of process improvements and, above all, a new level of efficiencies and responsiveness to customers not previously possible.

Industry 1.0

In the 18th century, with the power of water and steam, machineries were developed to perform various industrial task. This is widely known as the first industrial revolution or Industry 1.0. It allowed increase in mass production with machines power instead of purely human and animal power. The products were complete with machines rather than human hands.

Industry 2.0

The Industry 2.0 i.e. the second industrial revolution was fascinated with the use of oil, gas and electric power. Machines were then operated with electricity with more speed and telecommunication devices brought mass production and some degree of automation to manufacturing processes.

Industry 3.0

The third industrial revolution or industry 3.0 started in the 20th century. The new technologies like computers, advanced telecommunications factories were digitized with some extent. Automation of some processes, collection and sharing data made work management easier.

Industry 4.0

We are now in the fourth industrial revolution, also referred to as Industry 4.0. Characterized by increasing automation and the employment of smart machines and smart factories, informed data helps to produce goods more efficiently and productively across the value chain. Flexibility is improved so that manufacturers can better meet customer demands using mass customization—ultimately seeking to achieve efficiency with, in many cases, a lot size of one. By collecting more data from the factory floor and combining that with other enterprise operational data, a smart factory can achieve information transparency and better decisions.

Design Principles of Industry 4.0:

The important and basic principle of Industry 4.0 is that by connecting systems, work pieces and machines, businesses are creating intelligent network extend in the entire value chain that can control each other independently. There are four design principles of Industry 4.0. These principles can support companies to identify and help in implementing the Industry 4.0 scenarios.[1]

- Interconnection – The ability of systems, sensors, people and machines connect and communicate with each other through Internet of Things (IoT).
- Information transparency - The information transparency given by Industry 4.0 technology which provides huge amount of useful information to operators which can be needed to make the appropriate decisions.
- Technical assistance – Technical assistance system is support to humans by visualizing and aggregating information for solving short noticed problems and making important decisions. It also helps by conducting list of tasks which are unsafe, unpleasant or too exhausting for human co-workers [9].
- Decentralized decisions – The cyber physical systems support to perform tasks and make decisions as independently as possible. The tasks can be delegated to the higher level only in case of conflicting goals, exceptions or interferences.

Technological Force in Industry 4.0

Cloud computing

Storage and Exchange of data is prime demand in any industry operations. Cloud computing has provide the effective solution for Industry 4.0 strategy. Information of Raw material, assembly lines status, output of various units demands connectivity and integration in

supply chain, marketing and sales, and service management. Cloud computing makes this integration possible. Eliminating the need of EDP unit in every factory, cloud computing can reduce start-up costs of business.

Internet of Things (IoT)

Collecting small details of machines and process with identification is now possible with rise of Internet of Things (IoT) making it a key component of smart industries. With IoT, machines or environment in the factories are equipped with sensors with an identifiable address so that its minute details can be recorded or exchanged at the central place or other web-enabled devices.

Using high-tech IoT devices in smart factories leads to higher productivity and improved quality. Replacing manual inspection business models with AI-powered visual insights reduces manufacturing errors and saves money and time. By applying machine learning algorithms, manufacturers can detect errors immediately, rather than at later stages when repair work is more expensive

AI and machine learning

Artificial Intelligence and machine learning provide machine ability to analyse the inputs and generate optimal solutions for the given problem. Huge volume of data is generated through different sensors including drone cameras and algorithms of machine learning given the power to process these real time input to take much needed actions based on it.

Industries generate large volume of information at every step of business units. AI and machine learning can help in providing inspection, predictability and decision making automation of operations and business processes. For instance: Industrial machines are prone to breaking down during the production process. Using data collected from these assets can help businesses perform predictive maintenance based on machine learning algorithms, resulting in more uptime and higher efficiency.

Data Science

Data science is the study of data to extract meaningful information for better planning and implementations. It is a multidisciplinary approach that combines principles and practices from the fields of mathematics, statistics, artificial intelligence, and computer engineering to analyze large amounts of data.

Edge computing

The demands of real-time production operations mean that some data analysis must be done at the "edge"—that is, where the data is created. This minimizes latency time from when data is produced to when a response is required. For instance, the detection of a safety or quality issue may require near-real-time action with the equipment. The time needed to send data to the enterprise cloud and then back to the factory floor may be too lengthy and depends on the reliability of the network. Using edge computing also means that data stays near its source, reducing security risks.

Agriculture 4.0

Scientist have proposed the concept of site specific farming or precision farming running on principal of site specific farming and gathering real time information or data about soil, weather, crop etc. and analyse it for proper decision making. Agriculture 4.0 marks the extensive use of technologies in managing the agricultural work efficiently. The technologies of industry 4.0 like Internet of Things is very good at real time data collection, Artificial Intelligence, machine learning and data science make it possible to predict the trends and generate solutions or help in decision making. These techniques can be incorporated at various stages of crop cultivation, crop

management and product sale. Agriculture 4.0 suggest the optimum input of irrigation, fertilizers, and pesticides targeting very specific part of land.

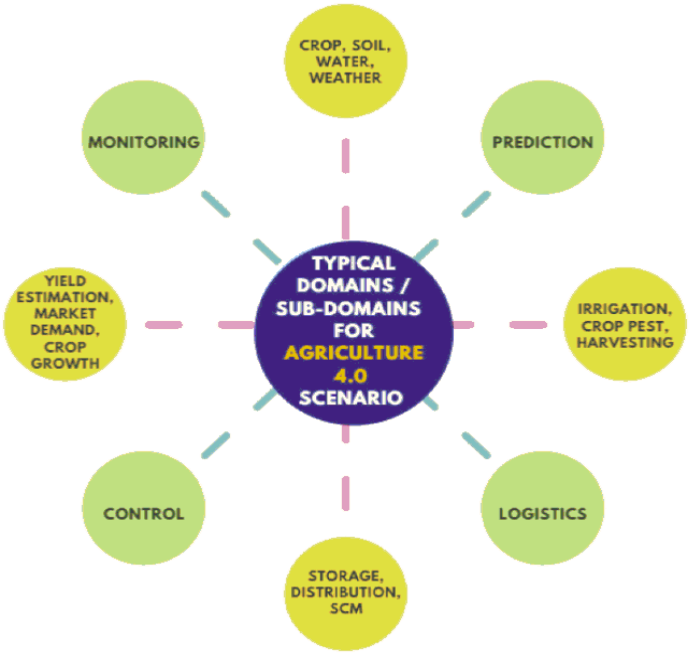
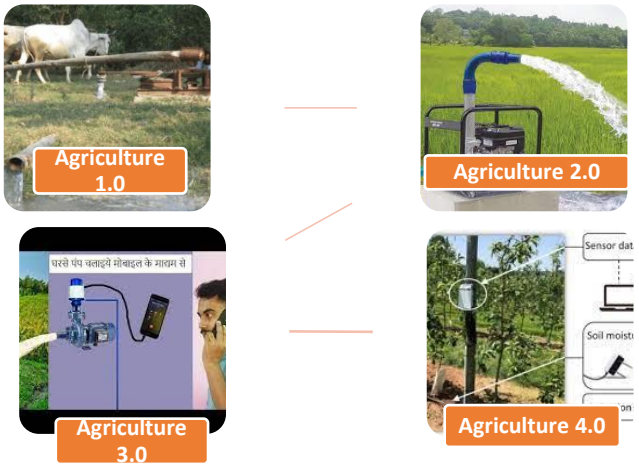


Figure 2 : Domains of Agriculture[1]

As part of Agriculture 4.0, the Internet of Things, Artificial Intelligence, and Nanotechnology, among other things, are gaining traction. It has changed the industrial process and has had a significant impact on agriculture and value chains. The farming industry is adopting genome editing, smart breeding tech, and combining digital AI-based technologies with microbial soil mapping to increase output quality and develop pest-resistant seeds [1,2]. The fundamental IoT security principles include monitoring data flow by encryption methods to safeguard critical data. This is done by employing AI-based security technologies to spot indicators of suspicious behaviour in real-time and storing data in the block chain to ensure its integrity. To fully profit from IoT, farmers must first become acquainted with the notion of data security and develop and adhere to internal security regulations [3,4].



Smart Farming

Smart farming encompasses all agricultural processes that aim to increase efficiency rather than capacity. The terms "smart farming" and "precision farming" are often used interchangeably. However, these two terms must be clearly delimited.

"Precision farming" focuses mainly on arable farming (e.g. site-specific management and documentation) while "smart farming" is more holistic, aiming to relieve the farmer in all parts of his business with intelligent electronics, and to bring about additional cost savings.

The **expected benefits** of digitization for smart farming can be summarized as follows:

There is large scarcity of physical labour required in farming. With use of technologies this dependency can be reduced up to some extents. Digitized tractors, sowers, harvesters, AI controlled irrigation systems may be an good alternatives in this aspect

IoT based systems found very useful in gathering real time data about soil moistures, temperature, and nutrient requirement. With such systems, Timely and precise actions are possible in supplying such important items to the crop.

Overall, it can be expected that process improvements can be achieved through continuous monitoring and an increase in knowledge, for example through AI applications.

Agriculture is carried out with large amounts of data, so it must be possible to store and make available whenever and wherever required. Cloud computing is far superior in sharing the data on your figure tip with mobile like devices.

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Indian Scenario of Mosquito borne Diseases

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Keywords:
Mosquito-borne diseases, India, Public health interventions, Vector control programs, Healthcare systems, Public awareness campaigns and Insecticide resistance.

Abstract:

Mosquito-borne diseases are a major public health issue in India, with a significant impact on morbidity and mortality rates. India is endemic to several mosquito-borne diseases, including malaria, dengue, chikungunya, and Japanese encephalitis. The burden of these diseases is especially high in rural areas where access to healthcare is limited.

Malaria is the most significant mosquito-borne disease in India, with over 85% of the country's population at risk of infection. However, the incidence of malaria has been declining in recent years due to various public health interventions.

Dengue and chikungunya are other significant mosquito-borne diseases in India. Both diseases have become more prevalent in urban areas, with periodic outbreaks occurring throughout the country. Japanese encephalitis, which is primarily a rural disease, is also a cause of concern in several states of India.

The Indian government has implemented several measures to combat mosquito-borne diseases, including vector control programs, public awareness campaigns, and strengthening of healthcare systems. However, challenges such as inadequate funding, inadequate infrastructure, and resistance to insecticides have hindered the effective implementation of these measures.

In conclusion, mosquito-borne diseases remain a significant public health challenge in India, and a concerted effort is required from the government, healthcare professionals, and the public to combat these diseases effectively

Introduction:

Mosquito-borne diseases have been a major public health concern in India for decades, with a significant impact on the country's morbidity and mortality rates. India is endemic to several mosquito-borne diseases, including malaria, dengue, chikungunya, and Japanese encephalitis. These diseases are prevalent in both rural and urban areas, and their burden has been particularly high in areas with poor sanitation, inadequate access to clean water, and inadequate healthcare systems.

Malaria, the most significant mosquito-borne disease in India, has been responsible for a large number of deaths and hospitalizations in the country. However, the incidence of malaria has declined in recent years due to various public health interventions. Dengue and chikungunya are also emerging as significant public health concerns, particularly in urban areas, with periodic outbreaks affecting large numbers of people. Japanese encephalitis, which is primarily a rural disease, is also a cause of concern in several states of India.

The Indian government has implemented several measures to combat mosquito-borne diseases, including vector control programs, public awareness campaigns, and strengthening of healthcare systems. However, these measures face several challenges, including inadequate funding, inadequate infrastructure, and resistance to insecticides. This article will discuss the current scenario of mosquito-borne diseases in India, their impact on public health, and the measures being taken to combat them.

Discussion:

The burden of mosquito-borne diseases in India has been significant, particularly in rural areas where access to healthcare is limited. Malaria has been the most significant mosquito-borne disease in India, with over 85% of the population at risk of infection. However, the incidence of malaria has declined in recent years due to various public health interventions, including the use of insecticide-treated bed nets, improved diagnosis, and treatment, and targeted indoor residual spraying.

Dengue and chikungunya have emerged as significant public health concerns in recent years, particularly in urban areas. The incidence of these diseases has been on the rise, and periodic outbreaks affect large numbers of people. The primary mode of transmission of these diseases is the Aedes mosquito, which breeds in stagnant water. Therefore, efforts to control these diseases include measures to eliminate breeding sites, such as regular cleaning of water storage containers, and promoting the use of insecticide-treated bed nets and mosquito repellents.

Japanese encephalitis is another significant mosquito-borne disease in India, particularly in rural areas. The disease is transmitted by the Culex mosquito, which breeds in paddy fields and other water bodies. Vaccination is an effective measure to prevent the disease, and several states in India have implemented mass vaccination programs targeting high-risk populations.

The Indian government has implemented several measures to combat mosquito-borne diseases, including vector control programs, public awareness campaigns, and strengthening of healthcare systems. However, these measures face several challenges, including inadequate funding, inadequate infrastructure, and resistance to insecticides. Additionally, climate change may lead to changes in the distribution of mosquito-borne diseases in India, making it necessary to continually reassess and adapt strategies to combat these diseases.

In conclusion, the burden of mosquito-borne diseases in India has been significant, and a concerted effort is required from the government, healthcare professionals, and the public to combat these diseases effectively. This effort should focus on targeted vector control measures, promotion of personal protective measures, strengthening of healthcare systems, and continuous monitoring and evaluation of interventions to assess their effectiveness.

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A Review on the Larvicidal Efficacy of Secondary Metabolites from Medicinal Plant Extracts against the Mosquito Genera *Aedes*.

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Keywords:
Phytoextract, Larvicidal Activity, Integrated Mosquito Management (IMM), *Aedes*

Abstract:

Mosquitoes are responsible for the majority of deadly diseases, including chikungunya fever, dengue fever, yellow fever, and malaria. Vector-borne illnesses are spreading globally, which has a negative effect on society and the economy. Aedes is a type of mosquito that aids in the spread of dengue fever, chikungunya, zika fever, mayaro, and yellow fever worldwide. Larvicides are becoming less effective against mosquitoes, and their resistance to synthetic insecticides is rising. This review aims to shed light on the use of biolarvicides to control the vectors *Aedes (Ae.) aegypti* and *Ae. albopictus*. 120 bioactive substances from 71 plant species were tested for larvicidal abilities, and diverse methods and solvents were employed by different workers to extract them. Biolarvicides are environmentally friendly, biodegradable, safe, and target-specific and should receive more attention for controlling vector species.

Introduction

Aedes (Ae.) aegypti and *Ae. albopictus* are the primary vectors of the emerging arboviruses dengue fever, chikunya, zika fever, mayaro, and yellow fever. Mosquitoes prefer to breed in stagnant water, such as flower vases, uncovered barrels, buckets, and abandoned tyres, and are especially driven to lay eggs in water containers with the right concentrations of particular fatty acids linked to bacteria that degrade food. A number of methods have been developed to control mosquitoes, including mechanical methods like eradicating breeding grounds, draining reservoirs, and installing screens on doors and windows, biological methods like using fish and invertebrates that feed on the larval stages of mosquitoes, and the use of plant extracts that have antilarviciadal properties for *Aedes.* (Puccioni-Sohler et al., 2017) (VEP et al., 2010)

Dengue: A Global and Indian Perspective

Dengue is a major public health issue in India, with 50 million cases reported between 2010 and 2019. Periodic dengue outbreaks have occurred in the nation, with the worst one in 2017 when over 180,000 cases were reported and more than 300 people died. Environmental elements such as temperature, rainfall, and humidity, as well as rapid urbanisation and population growth, all play a significant role in the transmission of dengue. India has implemented a number of interventions to address the dengue problem, including improved clinical management, public education campaigns, and vector control measures. (Guzman et al., 2016) (Leo, 2012) (Kakkar, 2012) .

Plants can be a promising source of eco-friendly larvicides as they biosynthesize a diverse array of biodegradable secondary metabolites with insecticidal activity. The efficacy of phytochemicals against mosquito larvae can vary significantly depending on plant species, plant parts used, solvent used, and vector species.(Shaalan et al., 2005) (M. F. Nethengwe, 2012).

Sr. No.	Plant species	Plant part	Solvent/fraction/ compound	mg/L(LC)	Source
1	*Abuta grandifolia*	Fruit	Dichloro methane	2.6	Ciccia *et al.* (2000)
2	*Acorus calamus*	Rhizome	Steam distillate	3.6 – 7.7	Ranaweera (1996)
3	*Acorus calomus*	--	Commercial oils	52 – 74	Sharma *et al.* (1994)
4	*Agave sisalana*	Sisal fibre	Na	322	Pizarro *et al.* (1999)
5	*Allium sativa*	Bulb	Crushed in water	160 (42)	Thomas and Callaghan
6	*Alnus glutinosa*	Old litter	Tannic acid	>20,000	Rey *et al.* (1999a)
7	*Alnus glutinosa*	Old litter	Tannic acid	13,000	Rey *et al.* (2001b)
8	*Alnus glutinosa*	Old litter	Polyphenols	200 – 400	Rey *et al.* (2001a,b)
9	*Alnus glutinosa*	Old litter	Different aged	25	David *et al.* (2000)
10	*Angelico glauca*	--	Commercial oils	52 – 74	Sharma *et al.* (1994)
11	*Annona squamosa*	Leaf	Water	2400	Monzon *et al.* (1994)
12	*Anthemis nobilis*	Flower	na	34	Soliman and El-Sherif
13	*Argemone mexicana*	Leaf	Benzene	30 – 55	Karmegam *et al.* (1997)
14	*Azadirachta indica*	Leaf	Water	4800	Monzon *et al.* (1994)
15	*Azadirachito indica*	Seed	Crushed seeds	100 (59)	Sinniah *et al.* (1994)
16	*Bryonopsis laciniosa*	Whole	Goniothalamin	21.5	Kabir *et al.* (2003)
17	*Cassia obtusifolia*	Seed	Methanol	40 (51)	Jang *et al.* (2002)
18	*Cassia tora*	Seed	Methanol	20 (59)	Jang *et al.* (2002)
19	*Callitris glaucophylla*	Wood	Steam distilled	0.69	Shaalan *et al.* (2003)
20	*Calophyllum inophyllum*	Leaf	Ethyl-acetate	16	Pushpalatha and
21	*Calophyllum inophyllum*	Seed	Ethyl-acetate	7.1	Pushpalatha and
22	*Calotropis procera*	Latex	Aqueous	28	Markouk *et al.* (2000)
23	*Cannabis sativa*	Leaf	Ethanol	1000	Jalees *et al.* (1993)
24	*Caulerpa scalpelliformis*	Whole	Acetone	53.7	Thangam and Kathiresan
25	*Cedrus deodara*	Different	Steam distillation	63.2	Kumar and Dutta (1987)

26	*Citrus limon*	Peel	Crushed in water	160 (80)	Thomas and Callaghan -1999
27	*Cleome viscosa*	Whole	Petroleum ether	10.7	Kalyanasundaram and Babu (1982)
28	*Codiaeum variegatum*	Leaf	Water	37,600	Monzon *et al.* (1994)
29	*Cotula cinerea*	Whole	Ethyl ether	310	Markouk *et al.* (2000)
30	*Curcuma domestica*	Rhizome	Petroleum ether	4.5	Ranaweera (1996)
31	*Cymbopogon flexuosus*	Different parts	Steam distillation	91.4	Kumar and Dutta (1987)
32	*Cymbopogon martini*	Different parts	Steam distillation	100	Kumar and Dutta (1987)
33	*Cymbopogon nardus*	Grass	Petroleum ether	6.3	Ranaweera (1996)
34	*Cymbopogon nardus*	Different parts	Steam distillation	105.4	Kumar and Dutta (1987)
35	*Daucus carota*		Commercial oils	36	Sharma *et al.* (1994)
36	*Dictyota dichotoma*	Whole	Acetone	61.7	Thangam and Kathiresan (1991a)
37	*Dirca palustris*	Seed	Hexane	10	Ramsewak *et al.* (2001)
38	*Euphorbia antiquorum* L.	Latex	Chloroform	82.17	Chandrasekaran R., et al., (2017)
39	*Euphorbia hirta* L.	Stem bark	Petroleum ether	272.36	Rahuman AA,et al., (2008)
40	*Euphorbia rothiana* Spreng.	Leaf	Distilled water	8.28	Banumathi B, et al.,(2017)
41	*Euphorbia tirucalli* L.	Stem bark	Petroleum ether	4.25	Rahuman AA,et al., (2008)
42	*Excoecaria agallocha* L.	Leaf	Methanol	41.74	Anil KV, et al., (2016)
43	*Hypericum japonicum* Thunb.	Whole plant	Methanol	7.37	Puthur S, et al., (2018)
44	*Hyptis suaveolens* Poit.	Leaf	Petroleum ether	64.49	Hari I, Mathew N. (2018)
45	*Jasminum fructicans*	Leaf	na	6	Soliman and El-Sherif
46	*Jatropha curcus*	Leaf	Benzene	175 – 188	Karmegam *et al.* (1997)
47	*Khaya senegalensis*	Seed	Hexane	5.1	Shaalan *et al.* (2003)
48	*Languas galanga*	Rhizome	Petroleum ether	8.3	Ranaweera (1996)
49	*Lansium domesticum*	Leaf	Water	5000	Monzon *et al.* (1994)
50	*Lavandula afficinalis*	Different parts	Steam distillation	83.6	Kumar and Dutta (1987)
51	*Melia azadirachta*	Different parts	Steam distillation	88.5	Kumar and Dutta (1987)

52	*Melia azedarach*	Seed	Acetone	30 – 40	Al-Sharook *et al.* (1991)
53	*Melia volkensii*	Seed	Acetone	20 – 30	Al-Sharook *et al.* (1991)
54	*Melia volkensii*	Fruit	Hexane: Ethyl	50	Mwangi and Rembold -1988
55	*Melia volkensii*	Fruit kernel	Methanol: water	5.4	Mwangi and Mukiama - 1988
56	*Mentha arvensis*	Different parts	Steam distillation	83.8	Kumar and Dutta (1987)
57	*Minthostachys setosa*	Whole	Dichloro methane	9.2	Ciccia *et al.* (2000)
58	*Ocimum basilicum*	Whole	Petroleum ether	47.3	Kalyanasundaram and Babu (1982)
59	*Origanum majoranal*	Leaf	na	54	Soliman and El-Sherif -1995
60	*Phyllanthus amarus* Schumach.	Leaf	Petroleum ether	90.92	Rahuman AA,et al., (2008)
61	*Phyllanthus emblica* L.	Fruit	Hexane	298.93	Kumar S., et al., (2012)
62	*Rhinocanthus nasutus*	Leaf	Petroleum ether fraction	22	Pushpalatha and Muthukrishnan (1999)
63	*Solanum trilobatum* L.	Leaf	Acetone	125.87	Premalatha S, et al., (2013)
64	*Solanum variabile* Mart.	Leaf	Ethanol	188	Porto KRDA, et al., (2018)
65	*Tagetes minuta*	Whole	Methylene chloride	173.8	Markouk *et al.* (2000)
66	*Thymus capitatus*	Whole	Volatile oils fraction	16	Perich *et al.* (1994)
67	*Valarian wallichii*		Commercial oils	49	Mansour *et al.* (2000)
68	*Vetiveria zizanioides*	Rhizome	na	52 – 74	Sharma *et al.* (1994)
69	*Vicia tetrasperma*	Seed	Methanol	67	Soliman and El-Sherif -1995
70	*Vitex negurdo*	Whole	Petroleum ether	100 (67)	Jang *et al.* (2002)
71	*Withania somnifera*	Leaf	Benzene	66.8	Kalyanasundaram 1982

Conclusion:

Botanical extracts have been shown to have a range of toxic effects, including deleterious effects on mosquito eggs, larvicidal properties, and growth regulating effects. When combined, these effects can produce impressive results, increasing the application possibilities for vector control. This review suggests that botanical phytochemicals should not be discounted as future alternative to synthetic insecticides, as joint-action may prolong the usefulness of synthetic insecticides.

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BRIEF OF OUTCOME

1. Understanding the interplay between ecosystems and human activities, which can help to develop sustainable practices that minimize the impact of human activities on the environment.
2. Identification of key areas of conservation concern, which can guide conservation efforts and help to protect endangered species and ecosystems.
3. Development of new technologies and techniques for monitoring and managing biodiversity, which can help to improve conservation outcomes.
4. Advancement in the fields of medicine and biotechnology, including the development of new treatments and drugs based on natural compounds.
5. Strengthening of interdisciplinary collaborations between scientists, policy-makers, and communities, which can lead to more effective and inclusive conservation strategies.
6. Creating opportunities for scientific training and education, which can help to build a skilled workforce capable of addressing current and future challenges in biodiversity conservation. Overall, the present and future perspective of life science research for sustainable development and biodiversity conservation can play a critical role in promoting long-term environmental sustainability, human well-being, and scientific advancement.

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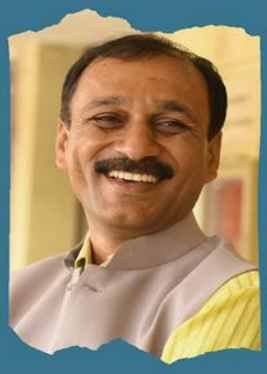

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