Bharati Vidyapeeth's

Dr. Patangrao Kadam Mahavidyalaya, Sangli

Fourth National Conference on

Recent Trends In Pure And Applied Sciences (RTPAS-2022)

21st and 22nd January 2022

RTPAS 2022

ISSN: 2454 6348

Doi: 10.26479/2022.0802.05

Special Issue

Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences

Index

Oxidation of benz and *m*-toluic acid hydrazides by thallium(iii) in acidic medium

Amit S. Varale*[1], Yashodhara S. Varale[2], Sachin V. Bangale[3], Milind B. Ubale[4], G. S. Gokavi[5]

*[1]Department of Chemistry, Athalye-Sapre-Pitre College, Devrukh, Dist-Ratnagiri

[2]Dr.Ambedkar College of Commerce and Economics, Wadala, Mumbai

[3]G.M. Vedak Arts, Commeerce and Science College, Tala Dist-Raigad

[4]Vasantrao Naik Mahvidhyalaya, Aurangabad

[5]Department of Chemistry, Shivaji University, Kolhapur

E-mail: amitvarale@gmail.com

Abstract

The reaction between thallium (III) and benz and *m*-toluic acid hydrazideswas carried out in a mixture of perchloric and hydrochloric acid medium. The reaction proceeds through formation of complex with reactant, which decomposes in subsequent steps to give product. Effect of acrylonitrile shows, that there is no formation of free radicals. The increase in $[H^+]$ and $[Cl^-]$ decreases the rate of the reaction. The increase in ionic strength does not affect the rate of reaction. The effect of temperature was studied at four different temperatures ranging from 15 to 30^oC. The activation parameters were also determined and a mechanism is predicted.

Keywords: kinetics, thallium(III), oxidation, Benzoic acid hydrazide i.e. Benz hydrazide (BAH), m-Toluic acid hydrazide (m-TAH)

Introduction

Thermodynamics is interested only in the initial and final states of a system, the mechanism whereby the system is converted from one state to another and the time is of no importance. Time is not

one of the thermodynamic variables. The most important subject in thermodynamics is the state of equilibrium and consequently, thermodynamics is the more powerful tool for investigating the conditions at equilibrium. Kinetics is concerned fundamentally with the details of the process whereby a system gets from one state to another and with the time required for the transition. Equilibrium can also be treated in principle on the basis of kinetics as that situation in which the rates of the forward and reverse reactions are equal. The converse is not true; a reaction rate cannot be understood on the basis of thermodynamics alone.

Therefore, a branch of chemistry, which deals with the study of reaction rates, i.e. chemical kinetics may be considered a more fundamental than thermodynamics.

Thallium(III) salts, which are wellknown oxidants in organic chemistry [1] have not yet been employed for the oxidation of hydrazides of carbon-nitrogen bonds: 1) Cleavage of oximes and semicarbazones to obtain aldehydes or ketones [2] and 2) the preparation of alkynoic esters or allenic esters from 5-pyrazolanes and their condensed analogs [3]. The oxidation of cyclopropane by thallium(III) acetate in acetic acid leads to cleavage of C-H bonds [4].The reaction is first order in both reactants. Interest in the use of thallium(III) in the oxidation of organic compounds has increased only recently and research in this regard is not been extensive. The potential of this oxidant is realized more and more as is evident from the considerable amount of work that is lately being done. Thus, the selectivity of thallium (III) is higher than its neighbours in the periodic table, mercury(II) and lead (IV) and also thallium (III) is a better oxidant than the other two. The kinetics of oxidation of simple olefins was studied in detail by Henry [5].

Literature survey reveals that, although several oxidants are used for oxidation of hydrazides and their mechanisms have been established, there is no report on the oxidation of hydrazides by thallium(III). Hydrazide derivatives are the type of organic compounds containing a nitrogen-nitrogen covalent bond with one of the substituents being an acyl group. They have gained prominence because of their antibacterial, anti-inflammatory, anticancer, antiplatelet, antimalarial, analgesic and antioxidant activity [6,7]. The objective of the present study is not only to develop method for the oxidation of hydrazides to their corresponding carboxylic acids but also to determine order of reaction and to propose the plausible mechanism of the reaction. The hydrazides are structurally related and are having different substituents.

Material and Method

Benzoic and m-Toluic acid hydrazides used are of 1M. Thallium(III) solution was prepared by dissolving Tl_2O_3 (ACROS) in 1.0 mol dm^{-3} HCl and the concentration was ascertained by iodometric titration. The Benzoic and m-Toluic acid hydrazides were prepared from reportedprocedure[8] and

characterised by determining their melting points. Stock solution of Benzoic and m-Toluic acid hydrazides were prepared in 50% v/v, 1,4-dioxane. Ionic strength was kept constant.

The reactions were carried out in 50 % v/v 1-4 dioxane (s.d.fine.chem) under pseudo-first order conditions keeping concentration of hydrazide in large excess over that of the oxidant. The solutions containing the reactants and all other constituents were thermally equilibrated separately, mixed and the reaction mixture was analysed for unreacted thallium(III) iodometrically by titrating against standard thiosulphate. The pseudo-first order rate constants were determined from the slopes of linear log[Tl(III)] versus time plots. The results were reproducible up to ± 5%. Kinetic runs were followed to about three half-lives of the reactions. Under the experimental conditions oxidation of 1,4-dioxane did not occur.

End product analysis. For identification of products the reaction was carried out by using aqueous solution of hydrazide, thallium(III), HCl and $HClO_4$. The flask containing reaction mixture was kept in thermostated water bath maintained at 50°C for 24 h to complete the reaction, the residue obtained after filtration was analysed for acid as follows:

(i) The presence of carboxylic acid group was detected by testing with bicarbonate.

(ii) The formation of acid was confirmed by IR and its melting point.

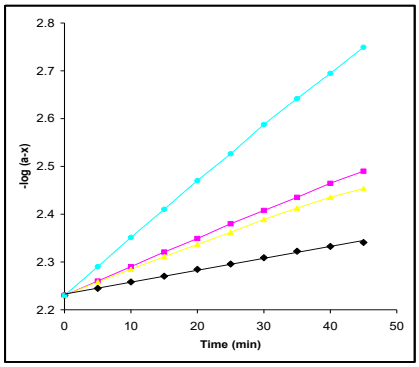
$$RCONHNH_2 + 2\,Tl(III) + H_2O \rightarrow R\text{-}COOH + N_2 + 4H^+ + 2\,Tl(I) \qquad \ldots (1)$$

Results and Discussion

The reaction occurs rapidly in perchloric acid medium but in the presence of hydrochloric acid the rate is measurable. Therefore, the reaction was carried out in a mixture of both acids. The effect of reactants on the reaction was studied at constant [HCl] and [$HClO_4$] of 0.1 mol dm^{-3} each and ionic strength of 0.6 mol dm^{-3}. Concentration of oxidant was varied from 6.4×10^{-4} to 6.4×10^{-3} mol dm^{-3} keeping the [hydrazide] constant at 1×10^{-1} mol dm^{-3}. Since, the pseudo-first order rate constants were fairly constant ($3.6 \pm 0.1\times10^{-4}$ s^{-1} for BAH and *m*-TAH), the order with respect to [oxidant] is unity. The effect of [hydrazide] was studied between the concentration range from 1×10^{-2} to 1×10^{-1} mol dm^{-3} keeping the [oxidant] constant at 3.0×10^{-3} mol dm^{-3}.The pseudo-first order rate constants increases with increase in concentration and the order with respect to hydrazide is found to be fractional.

To study the effect of [H^+] and [Cl^-],[oxidant], [hydrazide] and ionic strength were kept as 3.0×10^{-3}, 1×10^{-1} and 0.6 mol dm^{-3}, respectively. To vary [H^+] and [Cl^-], $HClO_4$ and NaCl were used. Increase in [H^+] from 0.13 to 0.60 mol dm^{-3} decreases $10^{-4}k$ (s^{-1}) from 4.20 to 0.15 for BAH and from 28.71 to 0.21 for *m*-TAH at 25°C. Increase in [Cl^-] from 0.13 to 0.60 mol dm^{-3} decreases $10^{-4}k$ (s^{-1}) from 2.80 to 0.095 for BAH and from 3.03 to 0.12 for *m*-TAH at 25°C. The relative permittivity was varied by changing the 1,4-dioxane content from 5 to 40% v/v. The rate was found to decrease with decrease in relative permittivity.

Added acrylonitrile in the concentration range 0.5 to 2.5 vol.% by keeping concentrations of oxidant, reductant, perchloric acid, hydrochloric acid and ionic strength fixed did not produce any precipitate due to polymerisation of the added acrylonitrile on the pseudo-first order rate constants indicating absence of free radical.



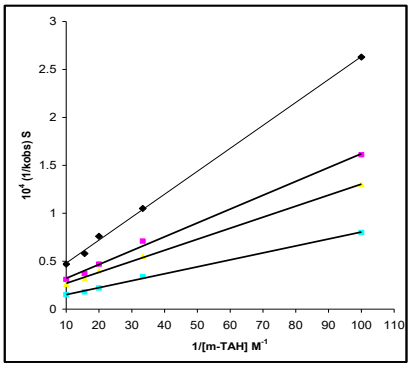


Fig. 1. Effect of temperature **Fig. 2**. The Michaelis-Menten plot

Since there is no formation of free radicals in the reaction, the reaction proceeds with two-electron transfer step. The order in thallium(III) was found to be unity and the order in hydrazide was found to be fractional. Such fractional order in substrate concentration is due to the prior complex formation equilibrium between the reactants.

S c h e m e 1

$$\mathrm{Tl(III)} + \text{hydrazide} \rightleftharpoons \text{complex} \qquad K_c$$

$$\text{complex} \rightarrow \mathrm{Tl(I)} + \text{intermediate} \qquad k_1$$

$$\mathrm{Tl(III)} + \text{intermediate} \rightarrow \mathrm{Tl(I)} + \text{products} \qquad \text{fast}$$

The Michaelis - Menten plots of $1/k_{obs}$ versus 1/[hydrazide] (Fig. 2) were linear with an intercept in support of the complex formation. Therefore, in agreement with the results obtained the mechanism of the reaction can be represented as in Scheme 1. Equation (2) gives the rate according to Scheme 1. Since, total [Tl(III)] exists in the form of free [Tl(III)] and the complex (equation (3)) therefore, the [Tl(III)] free is given by equation (5). The overall rate law is now expressed by equation (6) and the pseudo-first order rate constant k_{obs}, - by equation (7).

$$\text{Rate} = k_1\,[\text{complex}] = k_1K_c\,[\text{hydrazide}]_{free}\,[\mathrm{Tl(III)}]_{free} \qquad (2)$$

$$[\mathrm{Tl(III)}]_{total} = [\mathrm{Tl(III)}]_{free} + [\text{complex}] \qquad (3)$$

$$[\mathrm{Tl(III)}]_{total} = [\mathrm{Tl(III)}]_{free} + K_c\,[\text{hydrazide}]\,[\mathrm{Tl(III)}]_{free} \qquad (4)$$

$$[\mathrm{Tl(III)}]_{free} = [\mathrm{Tl(III)}]_{total}/(1 + K_c\,[\text{hydrazide}]) \qquad (5)$$

$$\text{Rate} = k_1K_c\,[\text{hydrazide}]\,[\mathrm{Tl(III)}]_{free} \qquad (6)$$

$$k_{obs} = k_1K_c\,[\text{hydrazide}]/(1 + K_c\,[\text{hydrazide}]) \qquad (7)$$

The rate law (equation (7) is verified by plotting $1/k_{obs}$ against 1/[hydrazide] at four different temperatures and from the slopes and intercepts of these plots the values of k_1 and K_c were calculated and are given in Table 1.

The effect of hydrogen and chloride ion concentrations on the reaction is due to the protonation of hydrazides[9]and different chloro–complexes of thallium(III) present in the solution. in acid medium according to equation (9). Hydrazides are known to be protonated, therefore, total [hydrazide] can be expressed by equation (10 and thereby the fact that there was no effect of free [hydrazide] by equation (11). Since the rates of reaction decreases as the $[H^+]$ increases, free hydrazide is the active species, this is in support of ionic strength on the reactions indicating one of the reactant is neutral.

$$RCONHNH_2 + H^+ \rightleftharpoons RCONHNH_3^+ \qquad K_H \qquad (8)$$

$$[Hydrazide]_{total} = [hydrazide]_{free} + [hydrazide]_{protonated} \qquad (9)$$

$$[Hydrazide]_{total} = [hydrazide]_{free} + K_H[hydrazide]_{free} \qquad (10)$$

$$[Hydrazide]_{free} = [hydrazide]_{total} /(1+ K_H [H^+]) \qquad (11)$$

Thallium(III) forms strong complexes with chloride ions of the formula $TlCl_n^{3-n}$ where n is the number of chlorides complexes with thallium(III) as represented in equilibria (12) to (15). The values of respective stability constants are $K_1 = 1.38 \times 10^8$, $K_2 = 3.98 \times 10^{13}$, $K_3 = 6.02 \times 10^{15}$ and $K_4 = 1.0 \times 10^{18}$ $mol^{-1}dm^3$.

$$Tl^{3+} + Cl^- \rightleftharpoons TlCl^{2+} \qquad K_1 \qquad (12)$$

$$TlCl^{2+} + Cl^- \rightleftharpoons TlCl_2^+ \qquad K_2 \qquad (13)$$

$$TlCl_2^+ + Cl^- \rightleftharpoons TlCl_3^+ \qquad K_3 \qquad (14)$$

$$TlCl_3 + Cl^- \rightleftharpoons TlCl_4^+ \qquad K_4 \qquad (15)$$

All the thallium(III) will exists as $TlCl_2^+$ and its concentration can be expressed by equation (16). The $[TlCl_2]^+_{free}$ can now be given by equation (18) where $\beta_1 = K_3/K_2 = 151$ and $\beta_2 = K_4/K_3 = 166$, further, using equations (18) and (19) the concentrations of $[TlCl_2]^+_{free,}$ $TlCl_3$ and $TlCl_4^-$ were calculated at different chloride ion concentrations and compared with the change in rate constant as the chloride ion concentration varied.

$$[Tl(III)]_{total} = [TlCl_2^+]_{total} = [TlCl_2^+]_{free} + [TlCl_3] + [TlCl_4] \qquad (16)$$

$$[TlCl_2^+]_{total} = [TlCl_2^+]_{free} (1+\beta_1[Cl^-] + \beta_2[Cl^-]^2) \qquad (17)$$

$$[TlCl_2^+]_{free} = [TlCl_2^+]_{total}/(1 + \beta_1 [Cl^-] + \beta_2[Cl^-]^2) \qquad (18)$$

The concentration of both of $[TlCl_2^+]_{free}$ and $TlCl_3$ parallel the values of rate constants as $[Cl^-]$ changes, but the order $[Cl^-]$ is -1.5, which makes $[TlCl_2^+]_{free}$ as the only active species.

Scheme 2

$$TIC1_2^+ + \text{hydrazide} \rightleftharpoons \text{complex} \qquad K_c$$

$$\text{complex} \rightarrow RCONHNH + T1C1_2\text{-} + H^+ \qquad k_1$$

$$RCONHNH + H_2O + TlCl_2^+ \rightarrow RCOOH + N_2 + 2H^+ + TlCl_2^- \qquad \text{fast}$$

where R is alkyl group.

The mechanism considering $TlCl_2^+$ of oxidant and free hydrazide of the substrate as the active species can now be represented by Scheme 2 with respective rate law and the expression for the pseudo-first order rate constants by equations (19) and (20). The rate law (equation(20) was verified by plotting $1/k_{obs}$ against 1/[hydrazide] and $1/k_{obs}$ against $[H^+]$ which were found to be linear. From the slopes and intercepts of these plots the values of K_c and K_H were determined. The values of K_c are given in Table 1 and those of K_H were found to be 13 and 16 mol^{-1} dm^3 for heterocyclic acid hydrazides, respectively.

$$\text{Rate} = \frac{k_1 K_c\,[\text{hydrazide}]_{total}\,[TlCl_2^+]_{total}}{(1+K_c\,[\text{hydrazide}])\,(1+K_H[H^+])\,(1+\beta_1[Cl^-]+\beta_2\,[Cl^-]^2)} \qquad (19)$$

$$k_{obs} = \frac{k_1 K_C\,[\text{hydrazide}]_{total}}{(1+K_c\,[\text{hydrazide}])\,(1+K_H[H^+])\,(1+\beta_1\,[Cl^-]+\beta_2[Cl^-]^2)} \qquad (20)$$

The electrophilic character of $TlCl_2^+$ among the thallium(III) chlorocomplexes is highest, thus making it the reactive species.

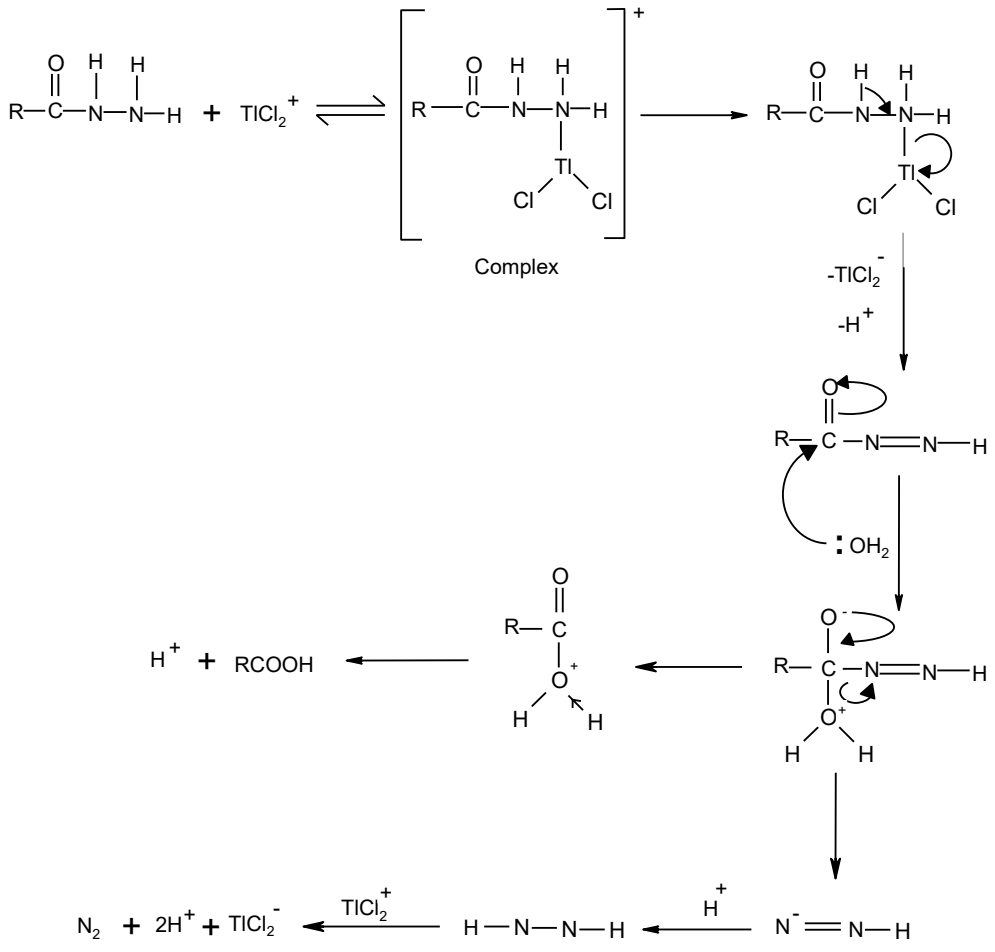
Table 1. Values of K_c and k_1

$[HCl] = 0.1$ mol dm^{-3}; $[HClO_4] = 0.1$ mol dm^{-3};$[Tl(III)] = 3.0 \times 10^{-3}$ mol dm^{-3}; $I = 0.6$ mol dm^{-3}

Hydrazide	K_c (mol dm^{-3})				$k_1 \times 10^4$ (s^{-1})			
	15°C	20°C	25°C	30°C	15°C	20°C	25°C	30°C
*B*AH	9.85	12.12	12.25	12.50	1.03	1.25	1.48	2.15
m-TAH	12.50	13.33	11.25	12.00	3.33	5.55	7.40	11.12

Mechanism

Scheme 3



R=Alkyl group for acid hydrazides

The detailed mechanism involves electrophilic substitution on the nitrogen of the hydrazide with the formation of N-Tl bond, which decomposes in the subsequent step with direct two-electron

transfer from hydrazide to thallium to give an intermediate followed by fast steps (Scheme 3). Such N-T1 bond formation has been postulated during thallium(III) oxidation of nitrogen-containing compounds[10].

The activation parameters, with respect to slow step, k_1, ΔH^*(kJ mol^{-1}), ΔG^* (kJ mol^{-1}) and ΔS^* (JK^{-1}mol^{-1}) were found to be 59.74, 87.94, - 94.34 for BAH and 32.76, 107.04 and -249.26 respectively for *m*-TAH[11-14]. Considerable decrease in the entropy of activation is due to formation of more ordered transition state as shown in Scheme 3. The mechanism involves neutral hydrazide as the active substrate thus the reaction is unaffected by the change in the ionic strength[15-16]. The increase in 1,4-dioxane content in the reaction medium decreases; the rate such an effect of the solvent is due to the stabilization of the complex formed between reactants in a medium of low relative permittivity[17-21].

Conclusion

The order of reactivity of benz and toluic acid hydrazides under investigation is:BAH <*m*-TAH. In case of toluic acid hydrazides the electron donating inductive effect of alkyl group is weaker and has negligible effect on reactivity. Our study on oxidation of benz and m-Toluic acid hydrazide is helpful to study thermodynamic pararameters and equilibrium constamnt of the reaction it also tells us the effect of substituent on the rate of reaction. In the future we are also going to study Hammet parameters.

Acknowledgment-

The authors are thankful to Prof.G.S.Gokavi, Shivaji University,Kolhapur and my Guide Prof.(Smt) N,P,Hilage for their help and support to carry out this work.

References

1. Mckillop, A Taylor, E.C. Organic syntheses by "Oxidation with Metal Compounds", Mijs, W.J., de Jonge, C.R.H. J., Eds., Plenum: New York., 1986, 695-701.

2. Narang, R Narasimhan, B and Sharma, S "A Review on Biological Activities and Chemical Synthesis of Hydrazide Derivatives," *Curr. Med. Chem.*, 2012,19, 569–612.

3.Suarez, J Ranguelova, K A Jarzecki, A K. Manzerova, J. Krymov, Zhao, X. Metlitsky, L.Gerfen, G J. and Mangliozzo, R S "An Oxyferrous Heme/Protein-based Radical Intermediate Is Catalytically Competent in the Catalase Reaction of Mycobacterium tuberculosis Catalase-Peroxidase (KatG)*," *J. Biol. Chem.*, 2009,11(284),7017–7029 doi: 10.1074/jbc.M808106200.

4. Stefane, B Kotevar, M and Polanc, S "Ceric(IV) Ammonium Nitrate In the Selective Conversion of Hydrazides to Esters," *Tetrhahedran Letter.*, 1999,40,4429–4432.

5. Kadam S D and Gokavi, G S. "Anderson type Hexamolybdochromate (III) catalyzed oxidation of acetic acid hydrazide by $KBrO_3$ in acidic medium," *Res. J. Chem. Sci.*,2016, 6(4), 17–23.

6. Narang, R Narasimhan, B and Sharma, S. "A Review on Biological Activities and Chemical Synthesis of Hydrazide Derivatives," *Curr. Med. Chem.*, 2012, 19,569–612.

7.Saini, M. Kumar, P Kumar, K. and Narasimhan, B "Synthesis , in vitro antimicrobial, anticancer evaluation and QSAR studies of N- (substituted) -4-(butan-2-lindeamino)benzohydrazides," *Arab. J. Chem.*, 2013, 7, 448–460, doi: 10.1016/j.arabjc.2013.05.010.

8. Fawzy, A. Ahmed, S A Althagafi, I L Morad, M H and Khairou, K S. "Kinetics and Mechanistic Study of Permanganate Oxidation of Fluorenone Hydrazone in Alkaline Medium," *Adv. Phys. Chem.*, 2016, 1–9, doi: 10.1155/2016/4526578.

9. X. Shen and R. W. Giese, "Hydrazide as a ligand moiety in immobilized metal ion affinity chromatography. Separation of BO-IMI and BODIPY-hydrazide," *J. Chromatogr. A*, vol. 777, no. 2, pp. 261–265, 1997, doi: 10.1016/S0021-9673(97)00370-1.

10. Safavi, A. Karimi, M. A. Hormozi Nezhad, Kamali, R and Saghir, N. "Sensitive indirect spectrophotometric determination of isoniazid," *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*,2004,60(4), 765–769. doi: 10.1016/S1386-1425(03)00288-9.

11. Osunlja, A A. Idris, S. O. and Iyun, J. F. "Kinetics and mechanism of the methylene blue-permanganate ion reaction in acidic medium," *Archives of Applied Science Research*, 2012, 4(2), 772-780.

12. Varale, A. S. Hilage, N. P."Oxidation of Nicotinic Acid Hydrazide by Thallium (III) in Acidic Medium: A Kinetic and Mechanistic Study" *Int J Chem Sci,* 2009,**7(**3), 2173-2178.

13Varale, A. S. Hilage, N. P."Comparative Kinetic Study of Oxidation of Toluic Acid Hydrazides by Thallium(III) in Acidic Medium"*Oxid Commun,*2009, 32(4) 867-874.

14. Varale, A. S. Hilage, N. P.'Kinetic and Mechanistic Study of Oxidation of Isonicotinic Acid Hydrazide by Thallium (III) in Acidic Medium'*Int J Chem Tech Res,*2009, 1(2) 270-273.

15. Varale, A. S. Hilage, N. P."Kinetic and Mechanistic Study of Oxidation of Salicylic acid Hydrazide by Thallium (III) in Acidic Medium"*Int J Chem Tech Res*,2011,3(17), 357-359.

16. Varale, A. S. Hilage, N. P."Comparative Kinetic and Mechanistic Study of Oxidation of Benzoic, *o*-Toluic Benzoic, *p*-Toluic Benzoic acid Hydrazides with Thallium (III) in Acidic Medium"*Oriental J Chem,*2011,27(1), 113-118.

17. Varale, A. S. Hilage, N. P."Comparative Kinetic and Mechanistic Study of Oxidation of Heterocyclic Acid Hydrazides by Thallium(III) in Acidic Medium"*Oxid Commun.* 35(2),371-374.

18.Varale, A. S. Hilage, N. P."Oxidation of o-Toluic Acid Hydrazide by Thallium (III) in Acidic Medium. A Kinetic and Mechanistic Study"*Oriental J Chem,* 29(2,) 667-669.

19. Varale, A. S. Hilage, N. P."Study of Thermodynamic parameters for oxidation of p-hydroxy benzoic acid hydrazide by Thallium(III) in Acidic medium- A kinetic and mechanistic approach" *Oxidation Communications.,*2015, 38(3), 1204–1212.Peer Reviewed and Indexed withThe Impact factor for 2013 is 0.507.

20.Varale, A. S. Hilage, N. P."Oxidation of p-amino benzhydrazide to the Corresponding Acid by Thallium (III) in 1, 4-Dioxane Medium - A Kinetic and Mechanistic Approach" published in Journal of Emerging Technologies and Innovative Research (JETIR) ISSN 2349-5162 Impact Factor 5.87, 2020, 7(3), 1607-1616**.**

21. Varale, A. S. Hilage, N. P."Oxidation of benzoic and n- Butyric acid hydrazides by Thallium (III) in Acidic Medium- A Kinetic and Mechanistic approach" published in International Journal of Grid and Distributed Computing, 2020, 13(2), 119 –125.

Liquid assisted grinding as environmentally benign protocol for synthesis of 6-amino-3-methyl-4-phenyl-1,4-dihydropyrano[2,3-*c*]pyrazole-5 carbonitrile derivatives as cystinyl amino peptidase inhibitors and antihypertensive agents

P. R. Kharade[1,2*], U. B. Chougale[1,2] K. N. Patil[3] and S. R. Dhongade[1]

[1]Research Laboratory in Heterocyclic Chemistry, Devchand College, Arjunnagar, Tal. Kagal, Dist. Kolhapur, 591269 (MH), India.

[2]Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti, Tal. Bhudargad, Dist. Kolhapur, 416209 (MH), India.

[3]Dr. Ghali College, Gadhinglaj, Tal. Gadhinglaj, Dist. Kolhapur, 416502 (MH), India.

ABSTRACT: A series of 6-amino-3-methyl-4-phenyl-1,4-dihydropyrano[2,3-c]pyrazole-5-carbonitrile derivatives were synthesized by liquid assisted grinding of pyrazolone, malono nitrile and different substituted aldehydes. The liquid system used for the synthesis is ethanol and water in 80:20 proportion respectively. The synthesized derivatives were characterized by spectral methods viz. IR, NMR and their structures were confirmed on the basic of spectral

data obtained. The structures of all the derivatives were further screened for their biological activities by using computer web-based program PASS. All the synthesized compounds were found as Cystinyl amino peptidase inhibitors and antihypertensive agents. The compound **1a (Pa value = 0.77)** was found to show highest activity as Cystinyl amino peptidase inhibitors and the compounds **2a** and **3a (Pa value = 0.54)** were found to show highest activity as antihypertensive agents.

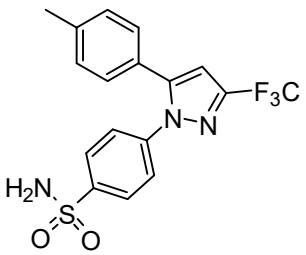
KEYWORDS: Pyrazolone, grinding, Cystinyl amino peptidase and antihypertensive agents.

***Corresponding Author: Mr. P. R. Kharade M. Sc., NET-JRF, SET, GATE**
Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti, Tal. Bhudargad, Dist. Kolhapur, 416209 (MH), India.

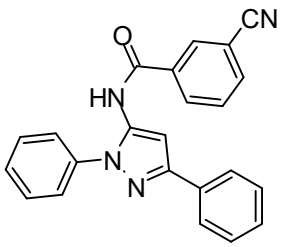
1.INTRODUCTION

Heterocyclic compounds possess wide applications as medical, agrochemicals, pharmaceuticals and functional materials [1-2]. Among them, Pyrazole fused heterocyclic scaffolds comprise main class of N-heterocyclics as important naturally occurring substances [3]. Many publications revealed that they have been used as antiallergenic, anticancer, hypotensive, antibacterial, antioxidant, antileishmanial, antifungal [4-8] etc. Further, pyranopyrazole derivatives are fused heterocyclic compounds which are biologically important as they show bactericidal and vasodilators activities [9-10].

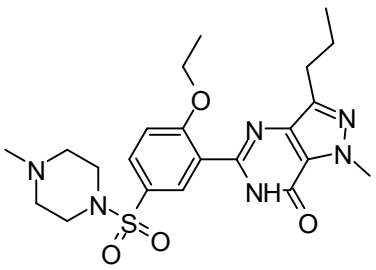
Due to such promising biological applications, many researches are attracted towards synthesis of pyranopyrazole derivatives. In recent years, variety of catalysts like TEA-Br [11], [Dsim] $AlCl_4$ [12], nano-TiO_2 [13], nano-CuI [14], nano-Fe_3O_4 [15], DABCO [16], pyrrolidine [17], CAN [18] and Chitosan hydrogel [19] etc were used for the synthesis of pyranopyrazoles. The synthesized and marketed drugs containing pyrazole and pyran core structures shown below.



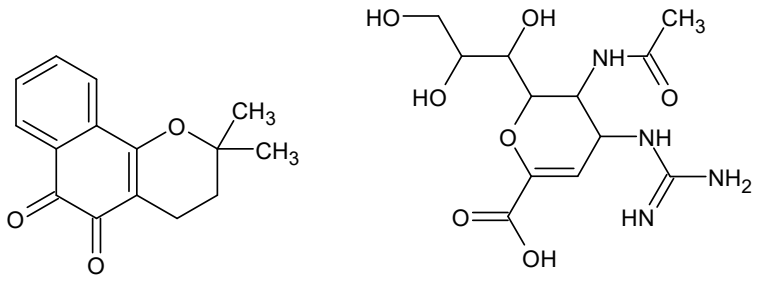
Celocoxib
anti-inflammatory



CDPPB
anti-psychotic



Sildenafil
Viagra- Sexual tonic



beta-lapachone zanamivir

Fig 1: Marketed drugs containing core pyrazole and pyran

Mostof these methods have certain disadvantages such as use of expensive catalytic system, high temperature reaction, longer reaction times and low product yields. To overcome these difficulties, we used grinding as greener tool for synthesis of pyranopyrazole derivatives using ethanol-water solvent system.

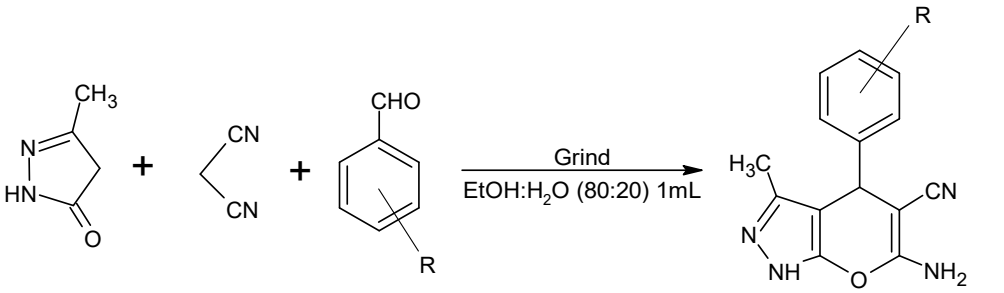
2. MATERIALS AND METHODS

IR spectra were recorded in KBr on FT/IR-4600 type A spectrophotometer.^{1}H NMR spectra were recorded on Bruker 400MHz spectrometer using TMS as an internal standard. Chemical shifts are reported in δ units and the coupling constants (J) are reported in Hertz. TLC was performed on an alumina backed silica plates with visualization by UV-light. Melting points were determined in open capillary tubes and were uncorrected.

EXPERIMENTAL

General procedure for the Synthesis of 6-amino-3-methyl-4-phenyl-1,4-dihydropyrano[2,3-*c*]pyrazole-5-carbonitrileDerivatives:

A clean, dry mortar and pestle were taken and charged with 3-methyl-1-phenyl-2-pyrazolin-5-one (1mmol), malononitrile (1.2 mmol) and substituted benzaldehydes (1.2 mmol). The mixture was ground for appropriate time in presence of 1 mL of EtOH:H_2O (80:20) solvent. After completion of reaction (as indicated by TLC), the product formed was quenched with water, filtered, dried and recrystallized from hot ethanol. The products were confirmed by comparing melting point data from literature and ^{1}H-NMR analysis.



R= -H, -mNO_2, -pNO_2, -pF, -pOH, -pOCH_3, -pCH_3

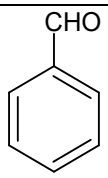
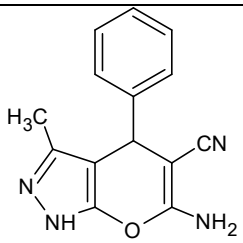
1a-1g

pyrano-pyrazole derivatives

Scheme 1

3. RESULTS AND DISCUSSION

The scope of the method was investigated with a series of substituted aromatic aldehydes. The results are summarized in Table 2. As seen from Table 2, the aromatic aldehydes carrying both electron-withdrawing (Entries 2-4) and electron-donating functional groups (Entries 5-7) underwent successful condensation with malononitrile at room temperature to afford the corresponding products in good yields. It seems that the electronic effects and the nature of the substituents on the aryl aldehyde ring have slight effect on both reaction yield and necessary time for the completion of the reaction. The electron-donating groups somewhat increased reactivity and afforded higher yields compared to electron-withdrawing groups. In addition, this reaction was affected by steric effect. In this case, the effects of functional groups in the aromatic aldehyde ring were opposite. Remarkably, the reactions were clean and all the products were obtained after only a filtration and simple washing with water and ethanol. Thus, a simple work-up gives the title products without of need of chromatographic purification. The time required, physical constant values and percentage yields are given in table below.

Entry	Aldehyde	Product	Time(min)	Yield (%)	M.P. (^{0}C)
1a			30	92	246-247

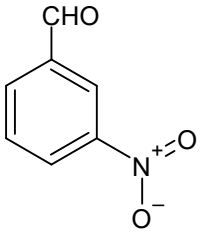
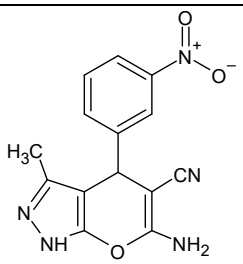
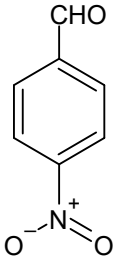
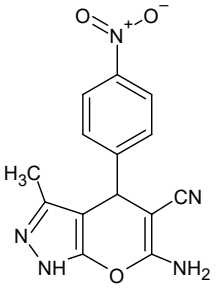
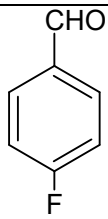
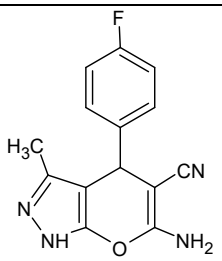
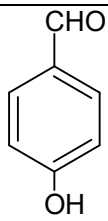
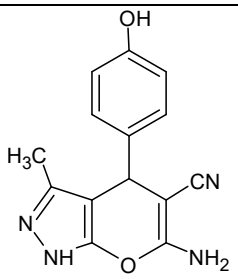
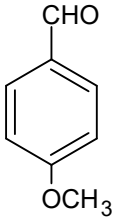
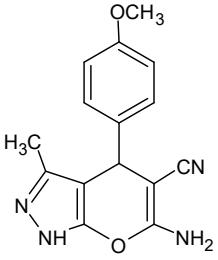
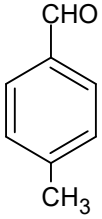
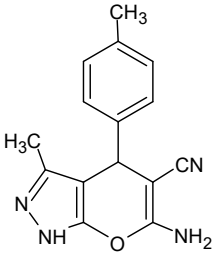
2b			25	87	194-195
3c			22	85	250-251
4d			20	89	244-245
5e			32	91	224-225
6f			35	89	210-211
7g			30	87	208-209

Table 1: Structures of synthesized compounds

Spectral data for compound 6f.

IR (KBr) cm^{-1}: 3478, 3417 (NH_2), 3087 (Aromatic), 2188 (–CN), 3241 (–NH–), 1481 (–NH–).

^{1}H NMR (400 MHz, DMSO-d6) : (δ ppm) 1.77 (s, 3H), 4.45 (s, 1H), 6.66–6.68 (d, 2H, *J* = 8.40 Hz, Ar–H), 6.74 (s, 2H, NH2), 6.92–6.95 (d, 2H, *J* = 8.40 Hz, Ar–H), 9.23(s, 1H, OH), 12.01 (s, 1H, NH).

Leucyl/cystinyl aminopeptidase (LNPEP) is a zinc-dependent aminopeptidase that cleaves vasopressin, oxytocin, lys-bradykinin, met-enkephalin, dynorphin A and other peptide hormones. [20]. The structures of synthesized compounds were subjected to computer web-based PASS program to find biological activities. Surprisingly, we found that the synthesized moieties can be used ascystinyl aminopeptidase inhibitors and antihypertensive agents. The predicted activities with probability of being active values (Pa values) summarized in the table given below.

Compound	Activity (Pa values)	
	Cystinyl amino peptidase	antihypertensive
1a	**0.77**	0.51
1b	0.72	**0.54**
1c	0.74	**0.54**
1d	0.72	0.45
1e	0.75	0.44
1f	0.72	0.49
1g	0.76	0.48

Table 2: PASS activities

The compound **1a (Pa value = 0.77)** was found to show highest activity as Cystinyl amino peptidase inhibitors and the compounds **2a** and **3a (Pa value = 0.54)**were found to show highest activity as antihypertensive agents.

4. CONCLUSION

In conclusion, 6-amino-3-methyl-4-phenyl-1,4-dihydropyrano[2,3-*c*]pyrazole-5-carbonitrile derivativeswere successfully obtained by usingliquid assisted grinding method. The structures were characterized by using physical data and spectroscopic methods.The structures of synthesized compounds were subjected to computer-web based PASS program to screen various biological activities.The screening data obtained through PASSprogram suggestedthat

the synthesized compounds can be used ascystinyl aminopeptidase inhibitors and antihypertensive agents. Certain disadvantages were overcame through this method such as low yields, longer reaction time, tedious workup process and use of toxic solvents.

5. ACKNOWLEDGEMENT
Financial support was provided to PRK under Research Initiation Scheme by Shivaji University, Kolhapur. The authors PRK and UBC are thankful to Principal, Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti for providing laboratory facilities.

6. CONFLICT OF INTEREST
Authors have no conflict of interest.

7. REFERENCES
1. Jampilek J. Heterocycles in medicinal chemistry. *Molecules*, 2019, 24(21): 3839.

2. Arora P, Arora V, Lamba H. S. and Wadhwa D. Importance of heterocyclic chemistry: a review. *Int. J. Pharm. Sci. Res.*2012, 3(9): 2947-2954.

3. Santos N. E., Carreira A. R. F., Silva V. L. M. and Braga S. S**.** Natural and Biomimetic Antitumor Pyrazoles, A Perspective. *Molecules*. 2020**,** 25(6): 1364.

4. Fustero S, Sánchez-Roselló M, Barrio P and Simón-Fuentes A. From 2000 to Mid-2010: A fruitful decade for the synthesis of pyrazoles. Chem. Rev., 2011, 111: 6984–7034.

5. Ansari A, Ali A and Asif M. Biologically active Pyrazole Derivatives. New J. Chem., 2017, 41: 16– 41.

6. Zaki M. E. A., Soliman H. A., Hiekal O. A. and Rashad A. E. Z. Pyrazolopyranopyrimidines as a class of anti-inflammatory agents. *Naturforsch C*, 2006, 61:1–5.

7. Abdelrazek M., Metz P., Metwally N. H. and El-Mahrouky S. F. Synthesis and molluscicidal activity of new cinnoline and pyrano [2,3-c]pyrazole derivatives. *Archiv. Der. Pharmazie*, 2006, 339(8):456–460.

8. El-Assiery S. A., Sayed G. H. and Foudan A. Synthesis of some new annulated pyrazolo-pyrido (or pyrano) pyrimidine, pyrazolopyridine and pyranopyrazole derivatives. *Acta Pharmaceutica*, 2004, 54(2):143–150.

9. Nasr M. N. and Gineinah M. M. Pyrido[2, 3-d] pyrimidines and pyrimido[5',4':5,6] pyrido [2, 3-d] pyrimidines as new antiviral agents: synthesis and biological activity. *Arch. Pharm. Med Chem*., 2002,335(6):289-295.

10. Ahluwalia V.K., Dahiya A. and Garg V. Reaction of 5-amino-4-formyl-3-methyl (or phenyl)- 1-phenyl-1H- pyrazoles with active methylene compounds: Synthesis of fused heterocyclic rings. *Indian J Chem*. *B*., 1997, 36:88-90.

11. Kumar, H., Saini, D., Jain, S., Jain, N. Pyazole scaffold: a remarkable tool in the development of anticancer agents. *Eur. J. Med. Chem*., 2013, 70:248–258.

12. Ahmad, R.M.Z., Mohammad, A.Z., Ehsan, N.M.T., Vahid, K., Abdolkarim, Z. Synthesis of 6-amino-4-(4-methoxyphenyl)-5-cyano-3-methyl-1-phenyl-1,4-dihydropyrano[2,3-*c*]pyrazoles using disulfonic acid imidazolium chloroaluminate as a dual and heterogeneous catalyst. *New J. Chem.,*2013, 37:4089-4094.

13. Shaterian H.R, Azizi H. Mild, four-component synthesis of 6-amino-4-aryl-3methyl-1,4-dihydropyrano[2,3-c]pyrazole-5carbonitriles catalyzed by titanium dioxide nano-sized particles.

Res. Chem. Intermed. 2014, 40(2):661–667.

14. Safaei-Ghomi J, Ziarati A, Tamimi M. A novel method for the one-pot five component synthesis of highly functionalized pyranopyrazoles catalyzed by CuI nanoparticles. *Acta Chim. Slov.,*2013, 60(2): 403- 410.

15. El-Aleem M. A. and El-Remaily A. A. Synthesis of pyranopyrazoles using magnetic Fe_3O_4 nanoparticles as efficient and reusable catalyst. *Tetrahedron.*2014, 70(18): 2971 – 2975.

16. Keyume A., Esmayil Z., Wang L. and Jun F. Convenient DABCO catalyzed one-pot synthesis of multi-substituted pyrano[2,3-c]pyrazole dicarboxylates. *Tetrahedron*, 2014, 70(26): 3976-3980.

17. Liju W. and Ablajan K. Pyrrolidine-Catalysed Four Component OnePot Synthesis of dihydropyrano[2,3-C]Pyrazole Derivatives. *Curr. Org. Synth*., 2014, 11(2): 310- 316.

18. Ablajan K., Liju W., Kelimu Y. and Jun F. Cerium ammonium nitrate (CAN)-catalyzed four-component one-pot synthesis of multi-substituted pyrano[2,3-c] pyrazoles under ultrasound irradiation. *Mol. Diversity*, 2013, 17(4): 693-700.

19. Patil K. and Helavi V. Synthesis of Pyranopyrazoles by using Chitosan Hydrogel as a green and recyclable catalyst. *Asian J. Research Chem.* 2018, 11(2):477-484.

20. Rogi T., Tsujimoto M., Nakazato H., Mizutani S. and Tomoda Y.Human placental leucine aminopeptidase/oxytocinase. A new member of type II membrane-spanning zinc metallopeptidase family". *J Biol Chem*., 1996, 271(1): 56–61.

Studies on use of yttrium sulphide as the storage electrode in photoelectrochemical (pec) storage cell

U.K. Mohite

Department of Physics, M.B.S.K. Kanya Mahavidyalaya, Kadegaon,Dist.- Sangli-415304(M.S.)

ABSTRACT: The study of yttrium sulphide as storage electrode was carried out by designing a special three electrode storage cell system. It consists of three electrodes, namely, storage electrode, photoelectrode and counter electrode. Electrodeposited yttrium sulphide film and CdSe film on to a stainlesssteel substrate has been used as a storage electrode and photoelectrode respectively. The graphite rod was used as a counterelectrode. These three electrodes were immersed in two rectangular transparent plastic boxes containing suitable electrolytes. Boxes were bridged together by agar-agar gel. The cell was illuminated by a high intensity lamp. The electrical characteristics in the mode of charging and discharging were studied.

KEYWORDS: Storage cell, storage electrode, photoelectrode, counterelectrode and agar-agar gel.

1. INTRODUCTION:

Now a day, for a sustainable society, energy is unquestionably one of the grand challenges [1, 2]. In this modern world, the social prosperity and economic development depend on the sustainable energy conversion and storage [2]. Since from 19^{th} century,due to vast consumption of non- renewable fossil fuels resulted in a severe anxiety for energy deficiency and the corresponding carbon emissions,creates new environmental issues. There is urgent need of clean, affordable and reliable energy that can substitute fossil fuels and limits the carbon emission issue. Therefore, the interest of researchers focused towards the development of technology to make availability of clean and renewable energy, especially the intermittent energy, energy conversion and storage [3, 4]. Now a day, there is vast demand for electrochemical energy conversion and storage devices, especially portable devices, consumer electronics, and electric vehicles [5–7]. Derek P Gregory has been reported use of rare earth hydrides for storing hydrogen in both stationary and mobile applications [8]. Therefore, it should require rapid development of new materials with high performance in energy conversion and storage devices. In our opinion, scientists underestimated this field and started working with material having low cast and easy availability. The best materials, for examples, so far reported with relatively with high efficiency and stability for long time are CdSe, WSe_2, CuIn Se_2[9-12].

Photo electrochemical cell can be converted into rechargeable electrochemical storage cell, when storage electrode, capable of undergoing a reversible chemical change is used in it [13]. A reversible chemical reaction occurs at the storage electrode of the type,

$$AX + ne^- \leftrightarrow A + X^{n-}$$

Where A is storage electrode, X is solute present in the electrolyte. Construction of such a cell requires stable low resistance separator which minimizes direct chemical reaction of the electro active redox species, and the selected redox couples suitable to semiconductor photoelectrode. The PEC cells employing third electrode as a storage electrode have been reported in the literature [14-19].

2. MATERIALS AND METHODS:

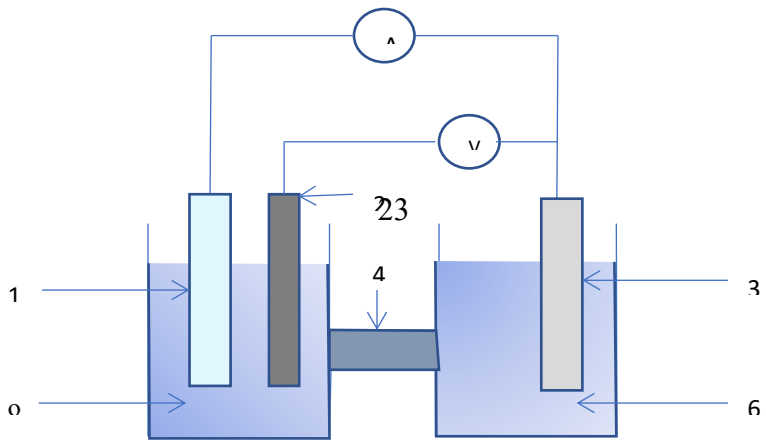
Preparation of electrode films and electrolytes:

Yttrium-sulphide films have been electrodeposited from the non-aqueous formaldehyde bath [0.05M Y $(NO_3)_3$ - 0.05M CH_3CSNH_2 - 0.05M CH_3COONa] onto a stainless steel substrate at room temperature. The CdSe films are electrodeposited from the aqueous bath [0.1M $CdCl_2$ - 0.05M SeO_2] onto stainless steel substrates. The PEC properties of the film were tested with the electrolyte 0.1 M (Na_2S - S – NaOH) as an electrolyte and graphite as a counterelectrode. In order to increase the photo effect, the films were annealed at 200^0C.

The electrolytes are prepared by using analytical grade chemicals in doubly distilled water. The stable electrolyte for the photoanode (CdSe) is polysulphide [20]. It was prepared by taking A.R. Grade Sodium hydroxide and sulphur from. B.D.H., India, and A. R. Grade sodium sulphide, from the Fluka. Appropriate amount of NaOH and Na_2S were dissolved in double distilled water at room temperature. In this solution, sulphur powder was added and mixture warmed up to 55^0C with constant stirring. The mixture was maintained at this temperature till all sulphur powder dissolves. The solution was cooled to room temperature, filtered and preserved in the glass stopper air tight bottle. The colour of the the solution was yellowish pink. The yttrium sulphide films are stable in the ferri-ferrocyanide electrolyte. This electrolyte was prepared by taking appropriate amount of potassium ferricyanide and potassium ferrocyanide of analytical grade, dissolved in double distilled water and preserved in the glass stopper air tight bottle.

Design of the Three Electrodes Storage Cell:

The design of three electrode battery was reported by many researchers in various journals [21-25]. Here, cell consists of three electrodes, namely, CdSe as a photoelectrode, graphite as counterelectrode and yttrium sulphide as storage electrode. Two rectangular transparent plastic boxes were fixed with M-seal by conducting bridge of 3 cm in length formed with Agar-Agar gel. The size of each rectangular box is $4.0 \times 1.5 \times 7.5 cm^3$. One compartment of cell consists of CdSe as a photoanode ($5 cm^2$area) and graphite rod ($6.2 cm^2$area) as the counterelectrode. The volumes of the electrolytes were 35 cc in each compartment of the cell. The electrolyte 0.1 M (Na_2S-NaOH) was used in first compartment. The other compartment consists of 0.1 M [$K_3Fe(CN)_6$] – $K_4Fe(CN)_6$] electrolyte with yttrium sulphide storage electrode which is kept in dark.



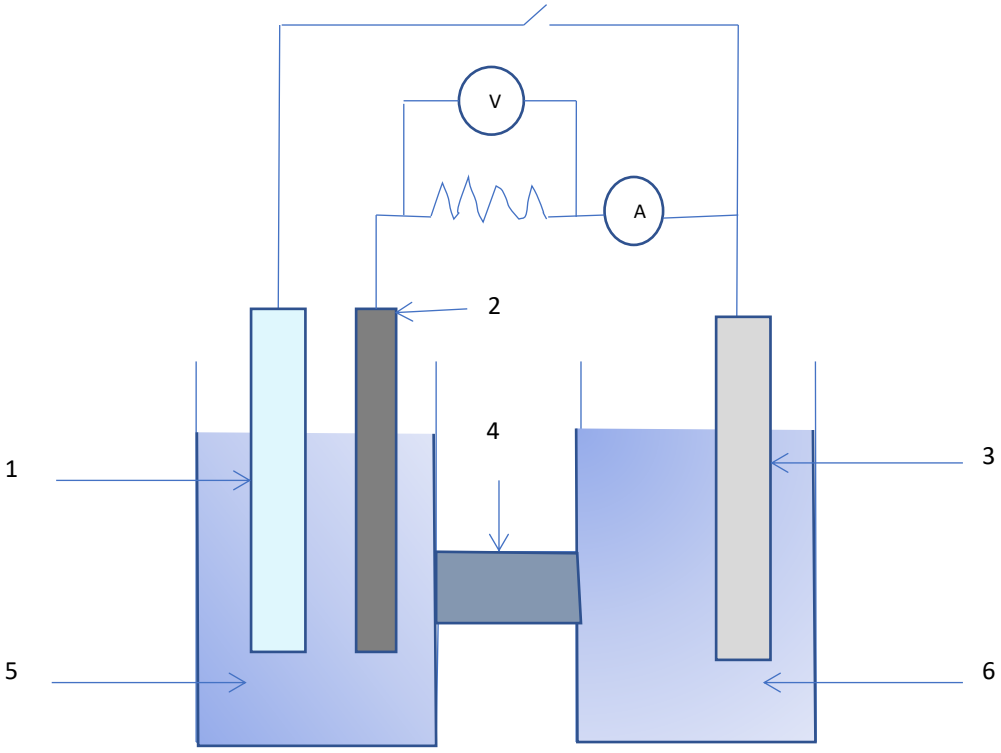


Fig.2: Schematicdiagram of the redox storage cell during discharging

The cell was illuminated by using 500 Watt tungsten filament lamp. The light intensity was $200mW/cm^3$. The electrical characteristics in the mode of charging were studied with the circuit diagram shown in fig.1 and fig.2 respectively. In fig. 1)- CdSe photoanode, 2)-counterelectrode, 3) Y-S storage electrode, 4) Agar-Agar gel, 5) 0.1 M (Na_2S - S – NaOH) and 6) $K_3Fe(CN)_6$] – $K_4Fe(CN)_6$. The current and voltages were recorded using the digital current and volt meters respectively.

3. RESULT AND DISCUSSION:

The Configuration of the Cell and Charge Transfer Mechanism:
The configuration of the cell was as follows:

n - CdSe/ 0.1M (Na_2S - S – NaOH) / C / 0.1M - [$K_3Fe(CN)_6$] – $K_4Fe(CN)_6$] /Y – S.

During charging the photoreaction occurring at the two electrodes can be described as follows:

$$n - CdSe + h\upsilon \rightarrow e^- + h^+ \qquad (h\upsilon > E_g)$$

Due to localized electric field at junction,

$$e^- + h^+ \rightarrow e^-{}_{bulk} + h^+{}_{surf}$$

(i.e. near the interface of the semiconductor electrolyte)

$$2h^+ + S^{2-} \rightarrow S \quad \text{(at the CdSe)}$$

i.e. oxidation of electrolyte would occur, which is present near interface.

$$e^-{}_{bulk} \rightarrow e^- \qquad \text{Storage electrode}$$

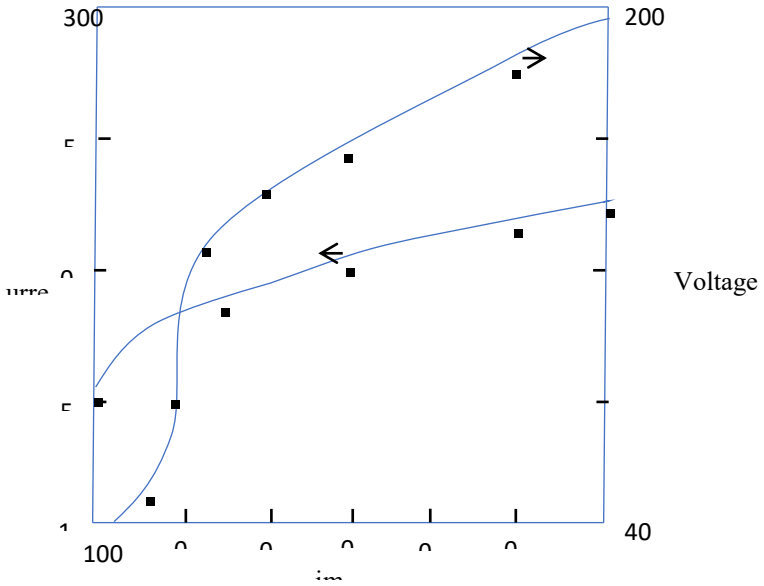
(Transfer to through back of semiconductor to the storage electrode)

$$2\,e^- + Y\text{-}S \rightarrow Y + S^{2-}$$

when Y-S was the storage electrode.

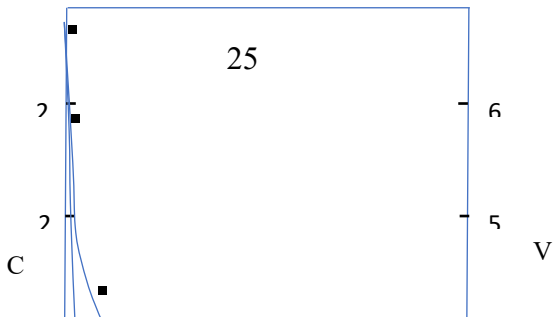
Charging and Discharging Studies of the CdSe/ 0.1M (Na_2S - S – NaOH) / C / 0.1M - [$K_3Fe(CN)_6$] – [$K_4Fe(CN)_6$] /Y – S.

During the period of charging, photocurrent raised from 0.15 to $0.225 mA/cm^2$, while cell voltage rose from 30 to 200 mV as shown in fig.3 within the period of 120 minutes.



cycle of the cellFig 3:

Discharging of the cell using 6 KΩ load across the storage electrode and counterelectrode results in an initial current of 25 $\mu A/cm^2$and after the period of two hours current drops to 6 $\mu A/cm^2$.The cell voltage also decreased from 600 to 300mV as shown in fig.4. It can be seen that the potential drops rather severely under load. This is attributed to resistance losses in the systems such as those in the photoactive layer and electrolyte, polarization losses at the counterelectrode and semiconductor metal contact.



4. CONCLUSION:

From above studies of charging and discharging characteristics, it is concluded that yttrium sulphide film may be used as a storage electrode.

ACKNOWLEDGEMENT:
The author thankful to Principal Dr. V. Y. Kadam, M.B.S.K. Kanya Mahavidyalaya, Kadegaon, for encouragement and provision of necessary facilities. The author is thankful to Dr. C. D. Lokhande, former Professor and Head of the Physics Department of Shivaji University, Kolhapur. Also, author is thankful to and all departmental faculties for kindly sharing their knowledge with me, for their suggestions and fruitful discussions.

REFERENCES:

[1] Arico AS, Bruce P, Scrosati B, TarasconJM ,Van Schalkwijk W,Nanostructured materials for advanced energy conversion and storage devices,Nat. Mater. 2005;4: 366–367.
[2] Chu S, Majumdar A, Opportunities and challenges for a sustainable energy future, Nature ,2012;488:294–303 .
[3] Liu C, Li F, Ma LP, Cheng HM, Advanced Materials for Energy Storage, Adv. Mater. 2010;22: 28–62 .
[4] Manthiram A, Fu Y, Chung SH, Zu C, Su YS,Rechargeable Lithium–Sulfur Batteries Chem. Rev. 2014; 114: 11751–11787.
[5] Lv W, Li Z , DengY ,Yang QH, Kang F,Graphene-based materials for electrochemical energy storage devices: Opportunities and challenges,Energy Storage Mater. 2016;2: 107–138.
[6] Yu-Guo Guo, Jin-Song Hu, and Li-Jun Wan, Nanostructured Materials for Electrochemical Energy Conversion and Storage Devices, Adv. Mater. 2008; 20: 2878–2887.

[7] Luhan Ye,Kechun Wen,Zuoxiang Zhang,Fei Yang,Yachun Liang,Weiqiang Lv and et. al., Highly Efficient Materials Assembly Via Electrophoretic Deposition for Electrochemical Energy Conversion and Storage Devices, Advanced Energy Materials.2016: Review.
[8] Gregory Derek P, Prospects for the use of hydrogen as an energy carrier; Proceedings of the 13th Rare earth conference held at Wheling, West Virginia. 1977;1:16-19.
[9] J. Manassen, G. Hodes and D, Cahen, Semiconductor liquid junction solar cells,Electrochemical soc.1977: 77:34-38.
[10] **XiaohuiZhang**, **BinYao**, **YongfengLi**, **ZhanhuiDing**, **HaifengZhao**, **LigongZhang** and et. al.Influence of WSe_2 buffer layer at back electrode on performance of $Cu_2ZnSn(S,Se)_4$ solar cells, Solar Energy.2020;199:128-135.
[11] Gutirrez MT and Ortega J, Photoelectrochemical Study of Electrodeposited Polycrystalline CdSe in Ferro-Ferricyanide System, J. Electrochem. Soc.1989;136:2316-2320.
[12]Ueno Y,Kaigawa H,Ohashi T,Sugiura T and Minoura H,Chemical bath precipitation of CdSe particles for use in a photoelectrochemical cell, Solar Energy Mat.1987:15:421-430.
[13] Sharon **Maheshwar, Veluchamy** P, **Natarajan** C, Kumar **Dhananjay,** Solar rechargeable battery—principle and materials, Electrochimica Acta.1991:36:1107-1126.
[14] Joost Manassen, Gary Hodes and David Cahen,Photoelectrochemical Energy Conversion and Storage: The PolycrystallineCdSe Cell with Different Storage Modes, Journal of Electrochemical Soc.1977:124:532-534.

[15] Liuyue Cao, Maria Skyllas-Kazacos, and Da-Wei Wang, Solar Redox Flow Batteries: Mechanism, Design, and Measurement, Advance Sustainable Systems; Review.2018:1-33.
[16] Gadave KM, Bhosale CH and Lokhande CD, Electrochemical storage cell formed with HgS films,Bull. of electrochem.,1993;9:262-263.
[17] Lokhande CD, Uplane MD and Pawar SH,Studies on photoelectrochemical storage celis formed with CdS:Cu electrode,Solid State Communication.1982;43:623-626.
[18] Dhumure SS and Lokhande CD, Studies on photoelectrochemical storage cells formed with chemically deposited CdSe and Ag2S electrodes, Sol. Energy Matter. and Sol.Cells.1993;29:183-194.
[19] Pawar SH, Patil MP and Shinde VN, Photoelectrochemical redox storage cell using electrodeposited CdSe as semiconductor-septum Bull.of Electrochem.,1993;9:252-254.

[20] Dhumure SS and Lokhande CD, Studies on photoelectrochemical storage cells formed with chemically deposited CdSe and Ag2S electrodes,Solar Energy Materials and Solar Cells.1993; 29:183-194.

[21] Costard J, Ender M, Weiss and Ivers-Tiffee E,Three-Electrode Setups for Lithium-Ion Batteries. Journal of the Electrochemical Society.2017;164(2):80-87.
[22] Song JY, Lee HH, Wang YY and Wan CC,Twoand Three-Electrode Impedance Spectroscopy of LithiumIon Batteries, Journal of Power Sources.2002:111: 255-267.

[23) Robert D. Minter, Daniel J. Robles, Conner Fear, Yevgen Barsukov and Partha P. Mukherjee,Three-electrode Coin Cell Preparation and Electrodeposition Analytics for Lithium-ion Batteries,Journal of visualized Experiments. 2018;135:57735.

[24] Goodenough JB and Park KS, The Li-Ion Rechargeable Battery: A Perspective,

Journal of American Chemical Society. 2013; 135 (4): 1167-1176.

[25] Johannes Landesfeind, Daniel Pritzl, and Hubert A. Gasteiger, An Analysis Protocol for Three-Electrode Li-Ion Battery Impedance Spectra,2017;164:1773-1783.

Study to understand the major stress related responses during the period of Covid -19 Pandemic lockdown by Student Stress Survey

Dr. Sheetal Bhopinder Singh Juneja
Assistant professor

Department of zoology

Dhote Bandhu Science College, Gondia (M. S)

Email: sheetalbhaskar107@gmail.com

Abstract

During Covid -19 Pandemic College students experienced Stress related responses due to fear of contagion and to limitations of personal and relational life. The intensity and frequency of behavioral, cognitive, and emotional responses of students during this period has also been affected. The COVID-19 pandemic has affected the mental health and social, emotional, psychological, and educational well-being of everyone. Taking Cognizance of this , Women Cell of Dhote Bandhu Science College Gondia, Maharashtra, India prepared a questionairee using Student Stress Survey template . The purpose of this questionnaire was to capture feedback about major stressers they experienced during the academic year with Covid -19 and how they handled that. This paper documents the findings from online interview surveys conducted through google form in a large institutional system in Dhote Bandhu Science College, Gondia, M. S. India. The study provided a brief, valid and reliable measure to assess perceived stress to understand the impact of lockdown amongst College students which will be helpful in developing tailored interventions fostering their wellbeing.

Keywords: Covid -19, student Stress, College Students, Physical and Mental Well Being.

Introduction:

In March 2020 WHO declared Covid-19 to be a global Pandemic , resulting in lockdown and life restrictions(1). During Covid -19 Pandamic College students experienced Stress related responses due to fear of contagion and to limitations of personal and relational life (2,3). The intensity and frequency of behavioral, cognitive, and emotional responses of students during this period has also been affected. The COVID-19 pandemic has affected the mental health and social, emotional, psychological, and educational well-being of everyone(4,5). Taking Cognizance of this Women Cell of Dhote Bandhu Science College Gondia, Maharashtra, India prepared a questionairee using Student Stress Survey template . The purpose of this questionnaire was to capture feedback about major stressers they experienced during the academic year with Covid -19 and how they handled that.

Total 256 students (211 girls and 44 boys) participated in the survey.

Methodology &STUDY area:

Materials and Methods

Participants and Sampling

Online survey data were collected from 23 to29th October 2021 with students of the Institution- Dhote Bandhu Science College Gondia (M. S) .This period fully corresponded to the condition when Educational Institution was reopened after the pandemic lockdown due to COVID-19 in Maharashtra and students have experienced various stress factors with massive social restrictions. The participants were contacted through Mentor- Mentee Whatsapp group. Students were contacted and given all the information about the study, and they were asked their participation on a voluntary basis.

All the participants were fully informed about the aims of the study and about the confidentiality of the data, and they were also assured that the data would be used only for the purpose of the research and refusal to participate would not affect their current and future course of study in any way.

Overall, 256 students voluntarily enrolled in the study and completed online google forms.

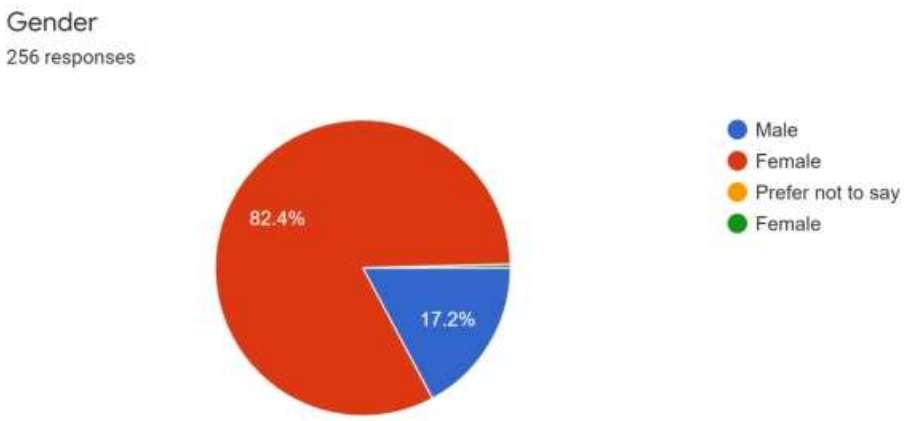
Measures

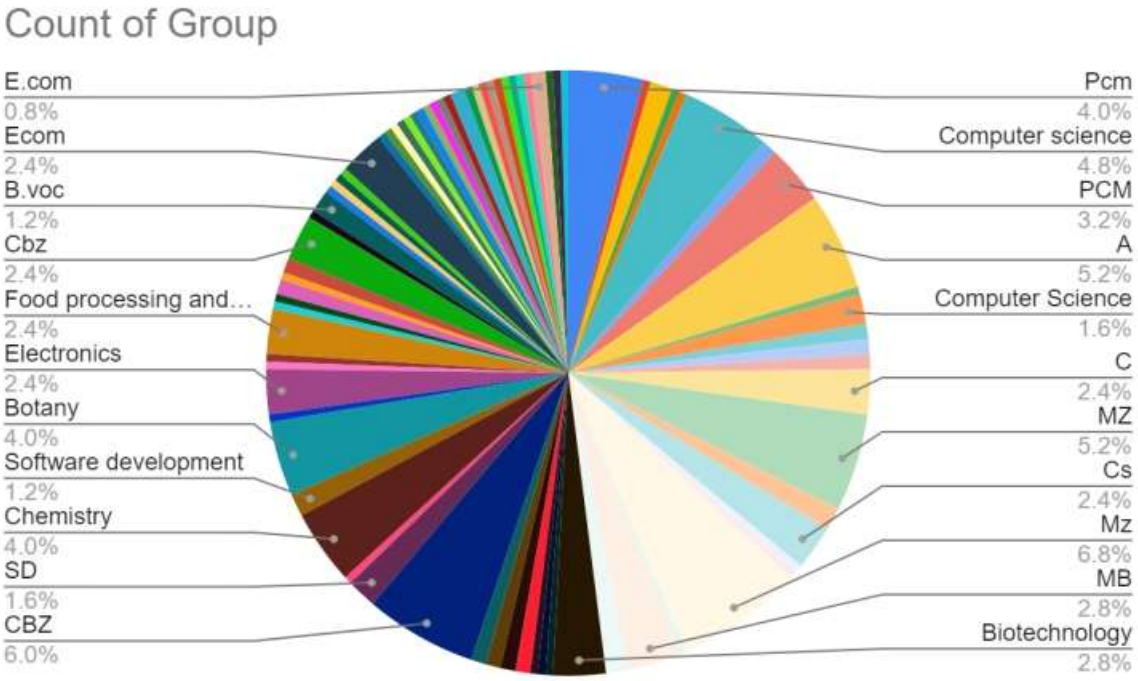
The questionnaire included a section dealing with background information (i.e., Gender, Age, Degree Program, Year of study) and the proposed 7-item COVID-19 Student Stress Questionnaire,

RESULTS

The total sample consisted of 211 girls and 44 boys, with a combined mean age of 19.92

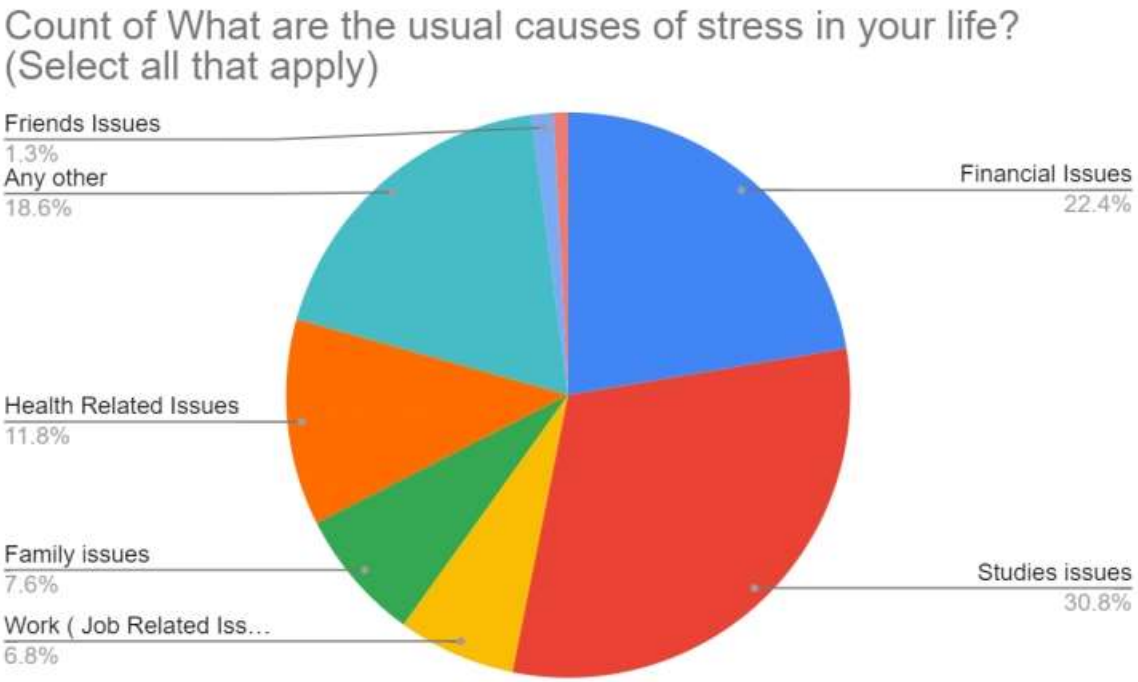
(*SD* = 1.50) years.



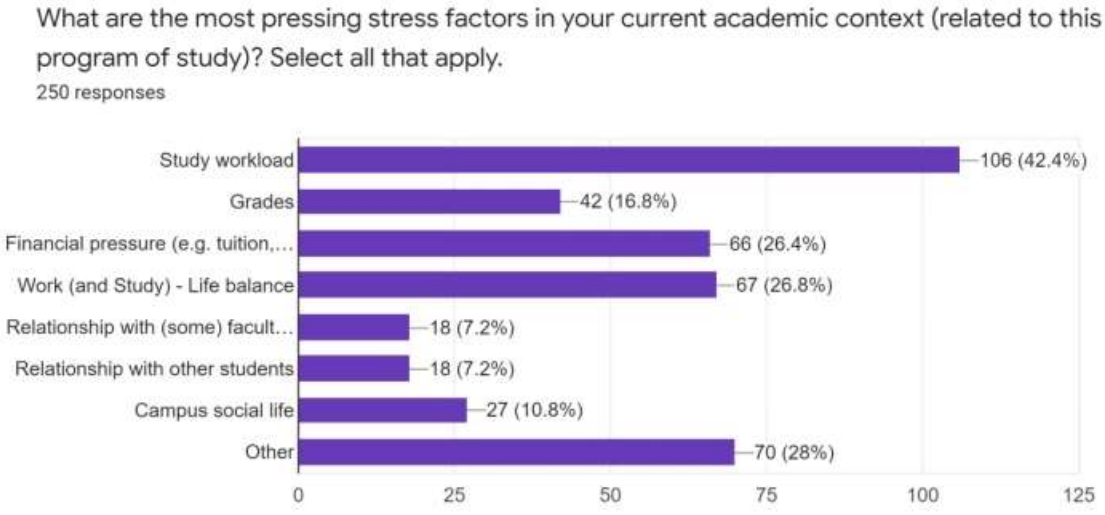


The sample was composed of students enrolled in E com (*n* = 10, 3.2%),

B. Voc (*n* = 44, 1.2%), Food Processing and technology(n= , 2.4%) , Electronics (n= , 2.4%), Botany (n= , 4%), Software development (n= , 2.8 %), Chemistry(n= , 6.4%), CBZ (n= , 6.0%), PCM and Psychology (*n* = 460, 89.5%) degree programs;

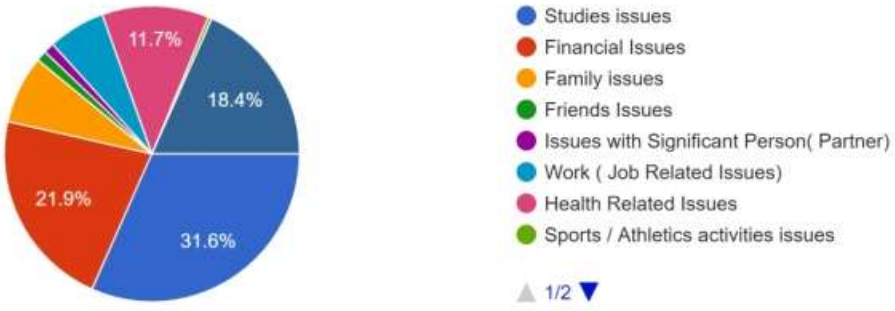


the majority of them were final year students (1st year *n* = 400, 77.8%; 2nd year *n* = 46, 8.9%; 3rd year *n* = 68, 13.3%).



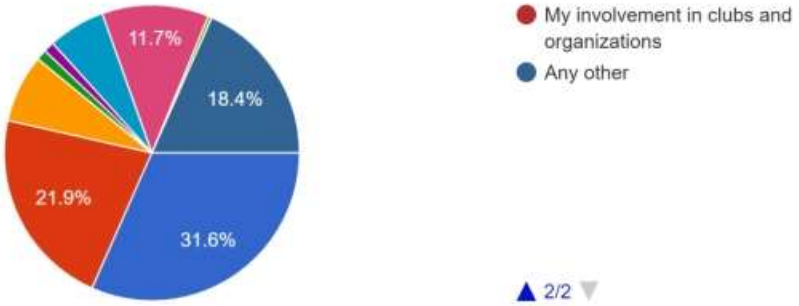
What are the usual causes of stress in your life? (Select all that apply)

256 responses

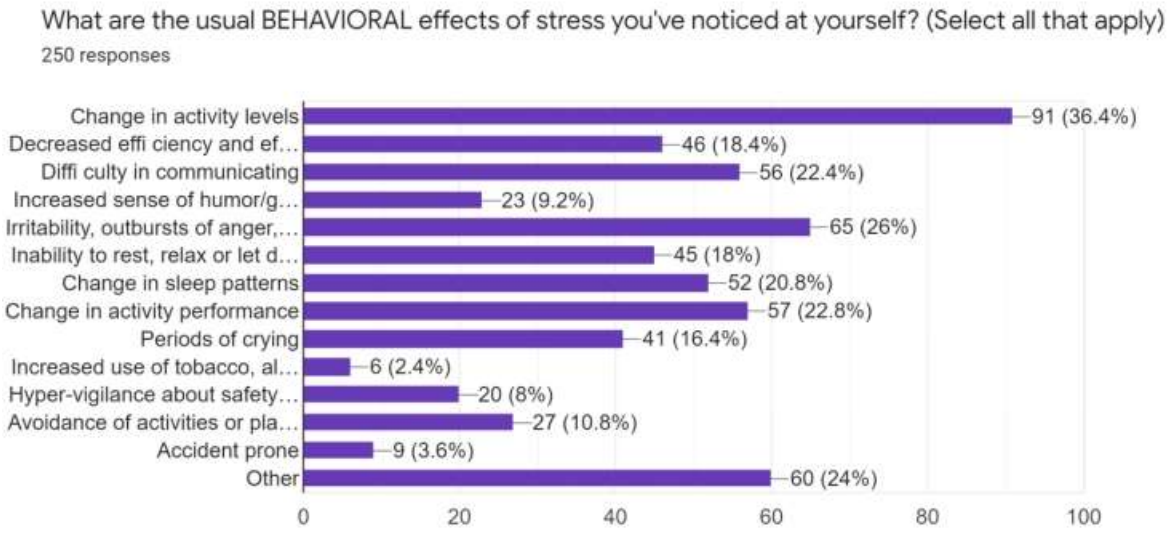


What are the usual causes of stress in your life? (Select all that apply)

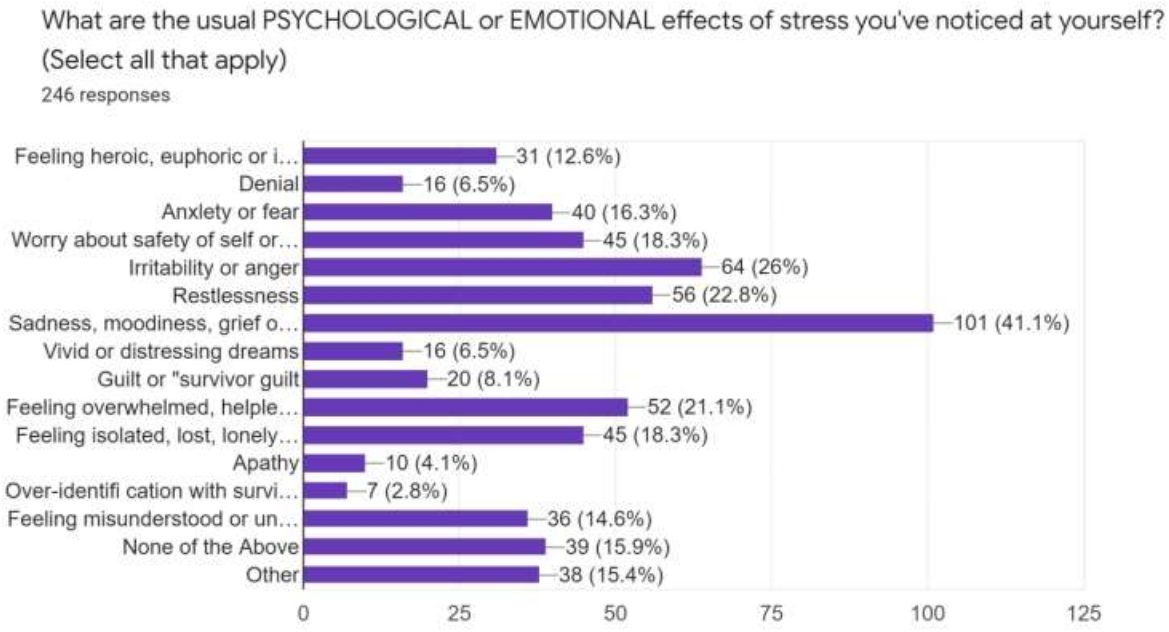
256 responses



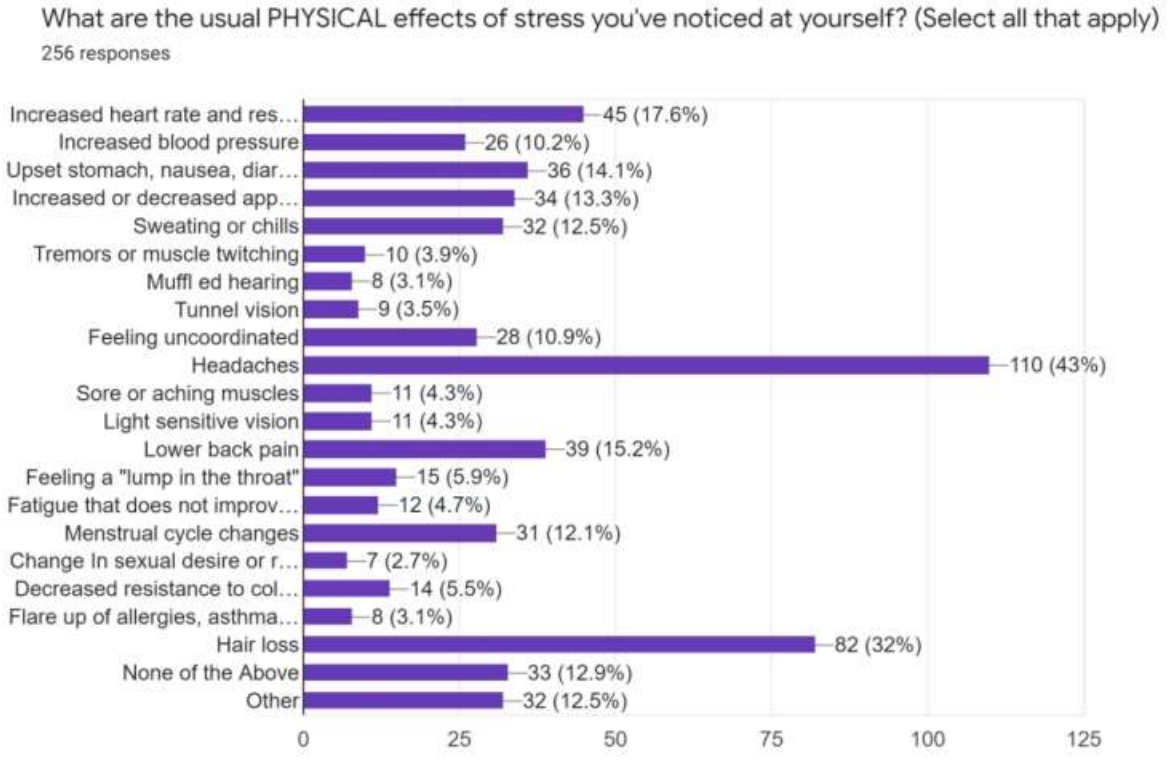
Due to on and off closure and opening of the educational institutions 30.8 % students reported worries related to studies while 22.4% told financial issues , 6.8% students reported job related issues to be stress related cause.



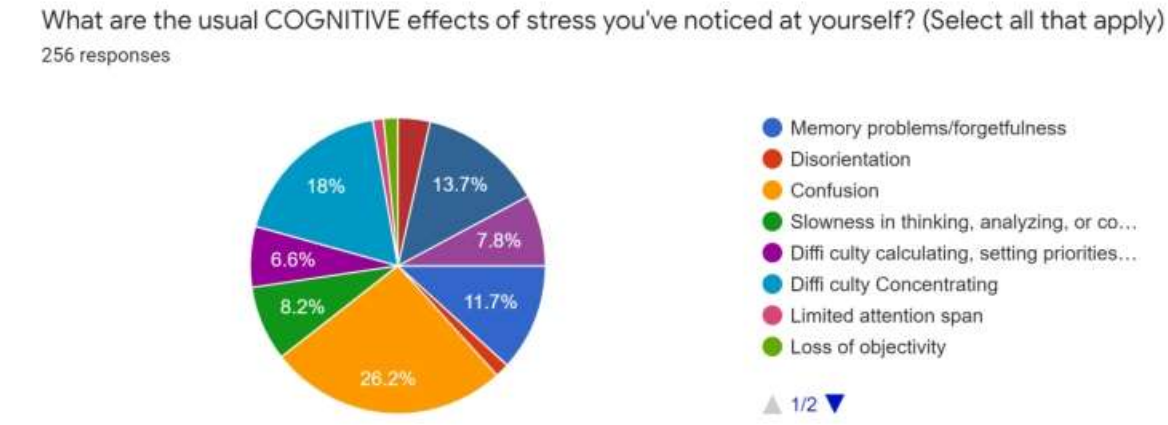
36.4% students reported Change in activity levels , 22.8% reported Decreased efficiency & Change in activity Performance-, 20.8% students reported Change in Sleep Patterns while 22.8% students reported change in behaviour.



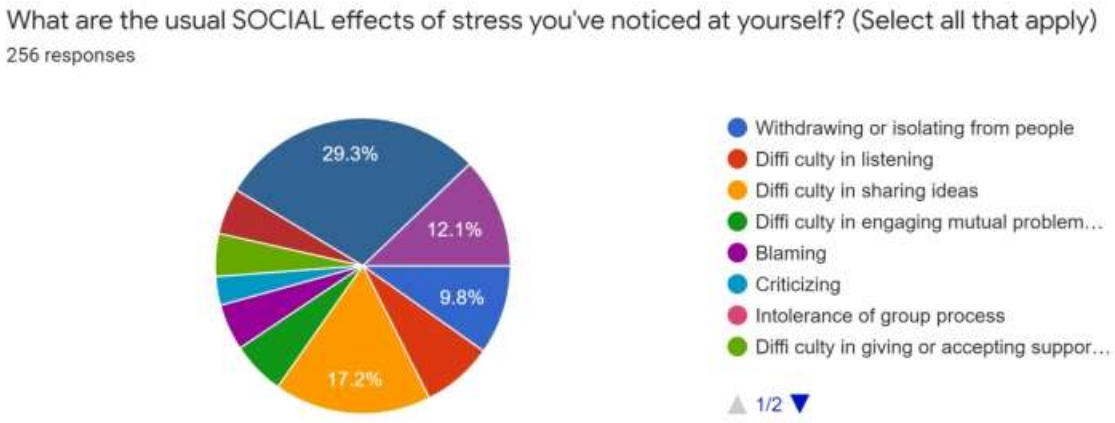
41.1% students reported Sadness, Moodiness,grief, 22.8% reported Restlessness, 18.3% reported feeling isolated, lost and lonely, 18.3% worry about self safety while 14.6% felt misunderstood by the others.



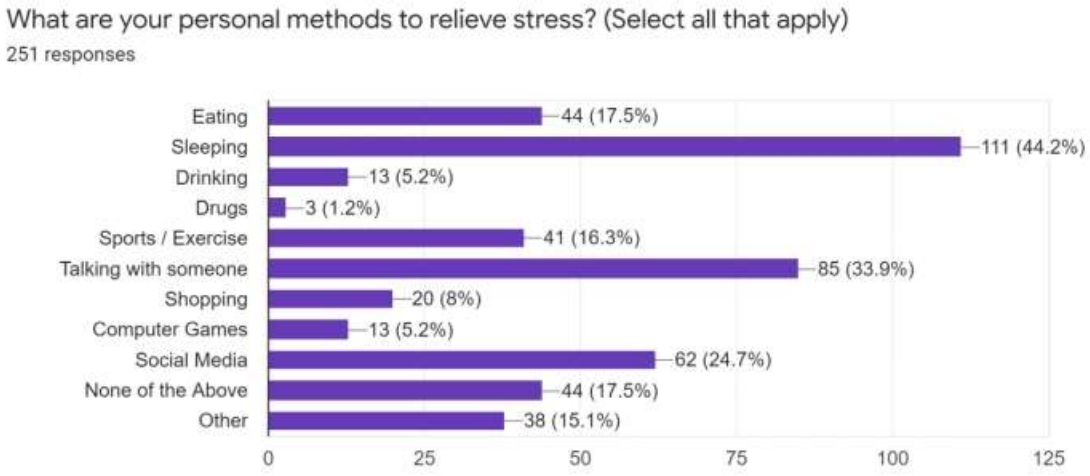
Amongst Physical Effects 17.6% students observed Increased Heart rate during the period of Covid-19. 12.15 girl students reported changes in menstrual cycle, 43% reported headaches .



26.2% students reported Confusion during these days.



29.3% students experienced Social Isolation. Most of the students experienced it when any of the family member got affected from the corona virus then they found themselves isolated.



Sleeping, Addiction to Social Media , Sports, Exercise, Chit chatting on phone and Online shopping were reported to be the best ways of relieving from stress.

Students Views on what the Institution should do to overcome these stress factors

'Due to corona virus pandemic there are so many student who are financially not strong because there parents also facing financial pressure. So that Some concession should be provide to students'.

'Good guidance for education and job, Institution giving the excellent education through vertual meetings , it could be very useful if they continue this education in a regular traditional way'.

'Help us to guide the career opportunities and support us'.

'Financial support and mentally support'.

‘ Arrange a programme for parents to tell them or make them understood that how important it is to be frank with their children. Tell them to make conversation with their kids’’

‘Providing scholarship is the better way to get rid of this problem (scholarship due to corona pandemic must increase)’.

‘Government bus will properly routine their schedule for traveling to help us the students’

‘The mind feels energized by the inspirational words of the teachers. There comes a passion to do something different in life’ .

Conclusion;

Higher education students experiences truly stressful situations a lot many times. Students feedback is essential for any academic institution . Such surveys can help us to understand different factors responsible for their stress. By doing so we can support our students to cope up with that more efficiently. It can also be helpful in developing tailored interventions fostering their wellbeing.

Acknowledgements: I want to acknowledge my Sincere thanks to Prof. Sanjay Timande , Head of the MicroBiology Department for all the guidance . I owe my thanks to our Principal Dr. Anjan Naidu for permitting and Collegeuees of our Institution Dhote Bandhu Science College Gondia for Circulating the Survey Form amongst the Mentees and Encouraging them for their Participation.

Conflict of Interest: There is no Conflict of Interest.

References:

1.Brooks, S. K., Webster, R. K., Smith, L. E., Woodland, L., Wessely, S., Greenberg,N., et al. (2020). The psychological impact of quarantine and how to reduce it: rapid review of the evidence. Lancet,2020. **395,** 912–920. doi: 10.1016/S0140-6736(20) 30460-8

2. Cao, W., Fang, Z., Hou, G., Han, M., Xu, X., Dong, J., et al. (2020). The psychological impact of the COVID-19 epidemic on college

students in China. Psychiatry Res.2020. 11, 287:293. doi: 10.1016/j.psychres.20 20.112934

3. Chen, B., Sun, J., and Feng, Y. (2020). How have COVID-19 isolation policies affffected young people’s mental health? - Evidence from

Chinese college students. Front. Psychol.,2020. 11:1529. doi: 10.3389/fpsyg.2020.01529

*4. Zurlo MC, Cattaneo MF, Volta D and Vallone F1,2020.*Frontiers in Psychology, 2020.11, Article 576758.

www.frontiersin.org. 18 Student Stress Survey Questions for Questionnaire + Template

5. Maria Clelia Zurlo1*, Maria Francesca Cattaneo Della Volta1,2 and Federica Vallone1,2020. COVID-19 Student Stress Questionnaire: Dstress/

Phytochemical Potential of Medicinal Plants –Influential Immunity Boostersfor the Pandemic of Covid-19

A. P. Patil[1*], P.D. Natekar[2] S.A. Ajerekar[3]

1. HOD, Department of Botany, R.B. MadkholkarMahavidyalaya,Chandgad, District-Kolhapur,
Maharashtra, India.

2. P.G. Department of Botany, DattajiraoKadam Arts, Science and Commerce College, Ichalkaranji, District-Kolhapur, Maharashtra, India.

3. Department of Botany, R.B. MadkholkarMahavidyalaya,Chandgad, District-Kolhapur, Maharashtra, India

---

ABSTRACT: In the present study medicinal plant from Chandgad region are reported with their ayurvedic medicinal importance and active principles. A total of 25 medicinal plants belonging to 22 different families were recorded. Out of which 2 were monocotyledons and 23 were dicotyledons. Present investigation was undertaken to study plant resources and updated practical activities like extracts preparations or poultice applied to the body to alleviate inflammation as well as to cure human health disorders. In our study, source of medicine were leaves in 6 plants-*Azadirachta indica* A. Juss. ,*Ocimum sanctum* Linn. *Nyctanthes arbortristis*Linn.,.*Centella aciatica* L. Urb., *Adhatoda vasica* Medic.,*Tridex procumbens* L.fruits in 11 plants-*Emblica officinalis* Gaerth.*Piper nigrum* L., *Citrusaurantium* L., *Garcinia indica* Du petit Thou.Choisy ,*Carica papaya* Linn. *Punica granatum* Linn.,*Helicteres isora* L.*Syzygium cumini* L. Skeels,*Aegle marmelos* Corr., *Terminalia chebula* Retz., *Solanum virginianum* L., stem in 3 plants- *Holarrhena pubescns* Wall. ex G.Don. , *Tinospora cordifolia*.,*Glycyrhiza glabra*(Willd.) Hook.f. & Thomson,L. , rhizome in 2 plants-*Curcuma longa* L.,*Zingiber officinale* Rosc., Floral bud in 1 plant-*Syzygium aromaticum*(L.)

Merr.&L.M.Perry.,bulb in 1 plant- *Allium sativum* L. and bark in 1 plant- *Cinchona offcinalis* (L.) Ruiz. It is necessary to conserve the precious biodiversity of plants as it gives powerful immunity boosters designed for the pandemic of covid-19 situation. In our studies total soluble proteins content was recorded in all medicinal plants. The total soluble proteins in the dried fruits of *Terminalia chebula*Retz. was found to be **66.9**g 100^{-1}g fr.wt.

KEYWORDS: Phytochemical, Medicinal plants, Uses, Families,Habit.

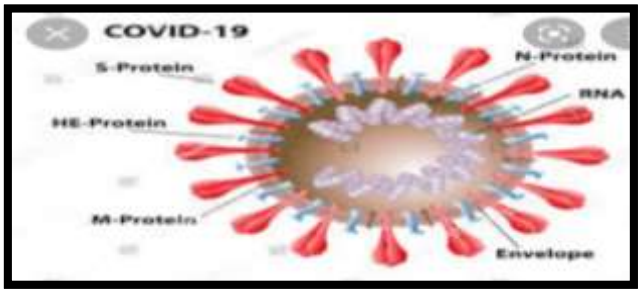
Corresponding Author:Anjali P. Patil1*Ph.D., email- anjalimane1972@gmai.com

1. HOD, Department of Botany, R.B. Madkholkar Mahavidyalaya, Chandgad, District-Kolhapur, Maharashtra, India.

1.INTRODUCTION

Medicinal plants provide a major resource for herbal industry. Nearly 80% of people rely on traditional herbal medicine to meet their primary health care needs due to their effectiveness[16]. Approximately 6000 medicinal plants are officially registered as herbal drugs in Ayurveda.

Structure of Covid-19 virus



Corona viruses are a group of related RNA viruses that cause diseases in mammals and birds. Corona viruses constitute the subfamily Orthocoronavirinae, in the family Coronaviridae, order Nidovirales and realm Riboviria. They are enveloped viruses with a positive-sense single-stranded RNA genome and a nucleocapsid of helical symmetry. The genome size of corona viruses ranges from approximately 26 to 32 kilobases, one of the largest among RNA viruses. They have characteristic club-shaped spikes that project from their surface, which in electron micrographs create an image reminiscent of the solar corona, from which their name derives.

Their size is highly variable with average diameters of 80 to 120 nm. The total molecular mass is on average 40,000 kDa. They are enclosed in an envelope embedded with a number of protein molecules. The lipid bilayer envelope, membrane proteins, and nucleocapsid protect the virus when it is outside the host cell. Four human corona viruses produce symptoms that are generally mild e.g. Cold, Cough,Fever.

Human corona virus OC43 (HCoV-OC43), β-CoV

Human corona virus HKU1 (HCoV-HKU1), β-CoV

Human corona virus 229E (HCoV-229E), α-CoV

Human corona virus NL63 (HCoV-NL63), α-CoV

Three human corona viruses produce severe symptoms e.g.Malerial Fever, Bronchitis, Respiratory tract infections, Fits, Nerve Disorders.

Severe acute respiratory syndrome corona virus (SARS-CoV), β-CoV (2003)

Middle East respiratory syndrome-related corona virus (MERS-CoV), β-CoV (2012)

Severe acute respiratory syndrome corona virus 2 (SARS-CoV-2), β-CoV (2019)

These cause the diseases commonly called SARS, MERS, and COVID-19 respectively.Medicinal plants have curative properties due to presence of chemical substances like alkaloid, glycoside, oil, gum, tannin, resin, minerals, steroids, starch, acid, mucilage, phenol, coumarin, anthraquinone, flavonoid, anthocyanin, saponin, vitamin and glucosilinates. Plant parts like root , bulb, rhizome ,bark, stem, leaves , flowers, fruits , seeds are utilized in the drugs preparation. Some important plants are useful for prevention of diseases.

2. MATERIALS AND METHODS

Medicinal plant survey was conducted during the last two years.The study area design for the medicinal plants was Chandgad taluka.For collecting varied information, we have visited to study area in rainy, winter and summer seasons. In that, we observed varieties of plant species in vegetative stage and reproductive stage. Several plant specimens were collected, reported along with their ethno-medicinal systems. This paper includes documents of dialogue along with twenty five rural people living near forest area of Chandgad, Here, Kolindre, Mirvel, Satwane, Kurtanwadi, Pohachiwadi, Adkur and Waghotre. Ethnobotanical information about medicinal plants is given by the local people. Geological co-ordinate of study area is $74^{0}18^{'}38^{''}$ E, $15^{0}95^{'}47^{''}$N. Recorded information [Table -1] was confirmed using literature cited in the ayurvedic medicinal books, regional flora, recent research papers from and relevant literature

[2], [4], [8], [10], [11], [13]. For extraction of total soluble proteins 1 g. fresh plant material was homogenized in 0.14M cold saline [NaCl] solution. The extract was filtered and centrifuged at 5000 rpm for 15 minutes. The supernatant was used as a source of proteins. For estimation of proteins method described by Gornall *et.al.* [1949] was used. When proteins are treated with an alkaline solution of copper sulphate the peptide linkages are broken down giving a characteristic violet colour to the solution. This reaction is termed as Biuret reaction and first demonstrated on Biuret which is the product of pyrogenic decomposition of urea. One ml of plant extract and one ml distilled water was mixed with eight ml ofBiuret reagent and it was incubated at 37^{0} for 30 minutes utilizing water bath. The absorbance of violet colour was measured at 540 nm. Simultaneously a set of reaction mixtures containing different concentrations from Blank, 0.4, 0.8,1.2,1.6 and 2 ml of standard Casein -20 mg/ ml was prepared to obtain a standard curve of proteins. This curve was used to determine the amount of proteins [17].

3. RESULTS AND DISCUSSION

In the present research work botanical name,vernacular name,family, habit, uses , chemical content , flowering and fruiting period of the plants were studied by using flora, internate facility and field visits. We have collected ethnobotanical information of medicinal plants [Table-1] given by the rural people which is needful to cure human diseases.

Table 1- Documentation of the ethnobotanical information of medicinal plants given by the rural people and Protein content in the medicinal plants parts.

Sr. No.	Name of the Vaidya	Botanical name, Vernacular name,family,habit and chemical contents of the plants	Proteins (g 100^{-1}g fr.wt.)	Use of the plants	Flowering and Fruiting period
1	Shri.J.B. Satwanekar	*Allium sativum*L. , Lasun (Family-Liliaceae) Herb Allicin,diallyldisulphid,vinyldithins ,ajoene,allyl propyl disulphide,Sulphur	3.9	Oil or cheese fried bulbs cure fever	Feb.-May

2	Shri. M. G.Patil	*Emblica officinalis*Gaerth., Avala(Family-Euphorbiaceae) Tree, Vitamin C,embilicanin-A and B-ellagic acid,quercetin,phyllemblic acid, alanine,aspartic and glutamic acid,lysine,proline,chromium,copper. iron ,niacin.	11.6	Fruit churna or boiled fruits with salt cure acidity	Apr.-May
3	Shri. J.B. Satwanekar	*Azadirachta indica* A. Juss,Kadulimb.(Family- Meliaceae) Tree,Tyurosine,proline,azadiractine, quercetine,nimbidine,nimbin,nimbandiol,nimbolide,quercitin,sistosterol, hexacosanol.Margolone	3.0	Leaf juice with honey cure skin infection	May-Sept.
4	Shri.V.S.Desai	*Piper nigrum* L., Miri (Family-Piperaceae) Climber , Piperine, piperidine	4.1	Seed powder with cheese cure malarial fever.	May-June
5	Shri.M..S. Kalkudrikar	*Curcuma longa* L., Halad(Family-Zingiberaceae) Herb ,Curcumine	3.0	Powder with milk or alcohol cure fever,cold,cough	July-Aug.
6	Shri.I.M. Patel	*Citrus aurantium* L., Limbu (Family-Rutaceae) Tree, Citric acid, hydroxyl citric acid, vitamin-c	6.2	Fruits cure stomach disorders	Apr.-Jun.
7	Shri. Shri.J.B. Satwanekar	*Garcinia indica* Du petit Thou. Choisy Kokam (Family-Cluisiaceae) Tree, Isogarcinol,cambogin,cambogenol,	6.4.	Fruits cure acidity	Nov.-Feb.

		Garcinol, garcinic acid, hydroxyl citric acid, malic acid, tartaric acid,			
8	Sou.V.M. Desai	*Carica papaya* L. Papai (Family- Caricaceae) Tree,Vitamin C,A,E, mineral Magnesium, Iron,Sodium, flavonid,carbohydrate potassium, pantothenic acid,folate acid, fibers, enzymes- papaintha and papain, terpenoid,alkaloid, tannin, glycoside,saponins,steroids.	11.2	Cure stomach disorders,used as abortifacient	Throughout the year
9	Shri. R.M.Patil	*Ocimum tenuiflorum*L., Tulsi (Family- Lamiaceae) Shrub, Eugenol,eugenol methyl ether,carvacrol,methyl, chavicol,cineole,linalool	6.5	Leaf juice cure Skin disease,cough ,cold, bronchitis	Throughout the year
10	Shri. R.M. Patil	*Punica granatum* L., Dalimb (Family-Lythraceae) Tree, Sugars, ascorbic acid, pectin, N,Zn,Fe,Co,I, Na, Mn, fibres protein,lipid,glycerol,linoleic acid, stearic acid, peletierine,punicic acid	3.7	Fruits cure stomach disorders	Apr.-May
11	Sou.S.V. Shinde	*Nyctanthesarbor-tristis* L., Parijatak (Family- Oleaceae) Tree, Crocin-3, Astragalin, Nicotiflorin, Nictanthoside	8.5	Leaf juice mixed in water cure fever	Aug.-Dec.
12	Shri. P.M.Disoza	*Holarrhena . pubescns* Wall. ex G.Don. , Kuda (Family – Apocynaceae) Tree,steroid,alkaloid,tannin,phenol, saponin,resin, coumarin,ergasterol	4.5	Stem water extract cure fever	Apr.-July

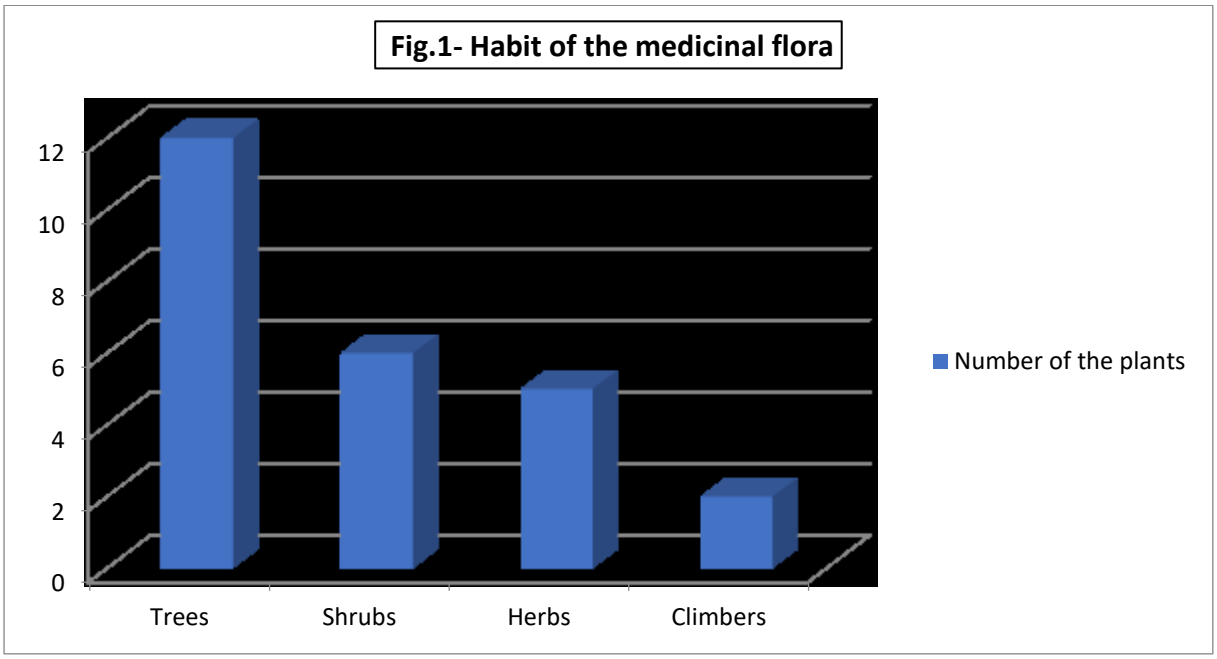
13	Shri. S. V. Shinde	*Centella asiatica* (L.)Urb., Brahmi (Family- Apiaceae) Herb, Bramhine	10.2	Leaf oil - Hair tonic	Aug.-Sept.
14	Shri. A.N. Patil	*Tinospora cordifolia* (Willd.) Hook.f. & Thomson, Gulvel(Family-Menispermaceae) Climber Tinosporin,columbin,chasmanthin,palmarin,berberin,tinosporin,giloinisin, B,sitosterol,nonacosan,pyrrolidine,furanolacton.	7.2	Stem powder mixed with water cure fever, dyspepsia, leprosy	May-June
15	Shri. A.N. Patil	*Helicteres isora* L. Muradshenge, (Family- Malvaceae) Shrub,Diosgenin	18.2	Fruit powder mixed with water cure stomach disorders	Apr.-Dec.
16	Shri.R.M.Patil	*Syzygium cumini* (L.)Skeels, Jambhul (Family- Myrtaceae) Tree, Myricetin, 3-L-arabinoside, dihydromyricetin, betulinic acid, friedellin.	36.0	Cure acidity, maintain blood Sugar level. dissolve kidney stone.	Mar.-Apr.
17	Shri. P.M.Disoza	*Justicia adhatoda* L., Adulsa (Family-Acanthaceae) Shrub,Vasicinolone,vasicol,peganine,vacisine,maiontone,sistosterol,glucoside,kaempferol	2.4	Leaf extract cure asthma,cold cough, dysentery, malaria, increases blood flow and cure skin disorders	Dec.-Feb.

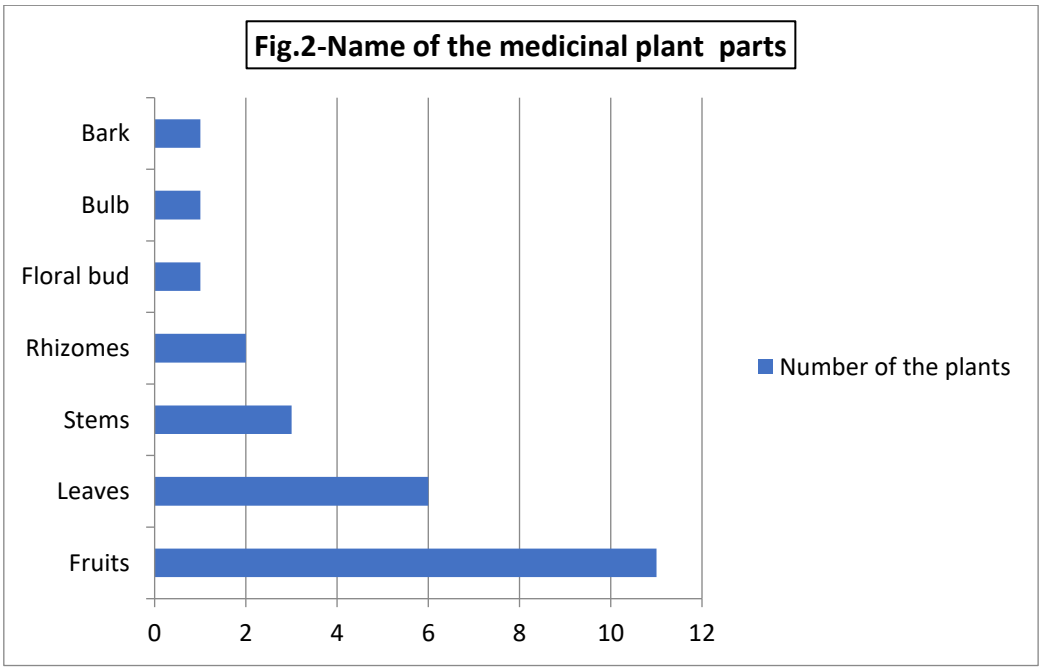
18	Shri. S.T.Ardalkar	*Aegle marmelos* (L.) Corr., Bel (Family- Rutaceae) Tree, Aegeline, marmin, phellandrene, rutin, linolenic acid. The drug of bel is called Belae fructus,	6.4	Cure diarrhoea, Fruit pulp mix with turmeric and paste is applied externally in case of pimples.	Jan.-May
19	Shri. S.T.Ardalkar	*Tridax procumbens* L., Dagadphool (Family-Asteraceaae) Herb, Steroids, carotenoids, fatty acids, sterol, tannin.	4.5	Leaf poultice cure wounds	July-Oct.
20	Shri. A.S.Patil	*Terminalia chebula* Retz., Hirda (Family- Combretaceae) Treecitric acid, hydroxyl citric acid,vitamin-c,	**66.9**	Fruit powder and honey cure stomach disorder acidity, thirst reliever	May-June
21	Sou. S.K. Meghane	*Syzygium aromaticum*(L.) Merr.&L.M.Perry, Lavang (Family-Myrtaceae) Treeprotein,carbohydrate,tannin,oleanolic acid,eugenol acetate,cayeophyllene,eugenol,	**54.1**	Cure toothache	June-Oct.
22	Sou.P.P. Meghane	*Glycyrrhiza glabra* L., Jashtamadha (Family-Fabaceae) Shrub, Estragole, anethole, eugenol, indole y-nonalactone, cumic alcohol.	8.9	Sore throat	June-Aug.

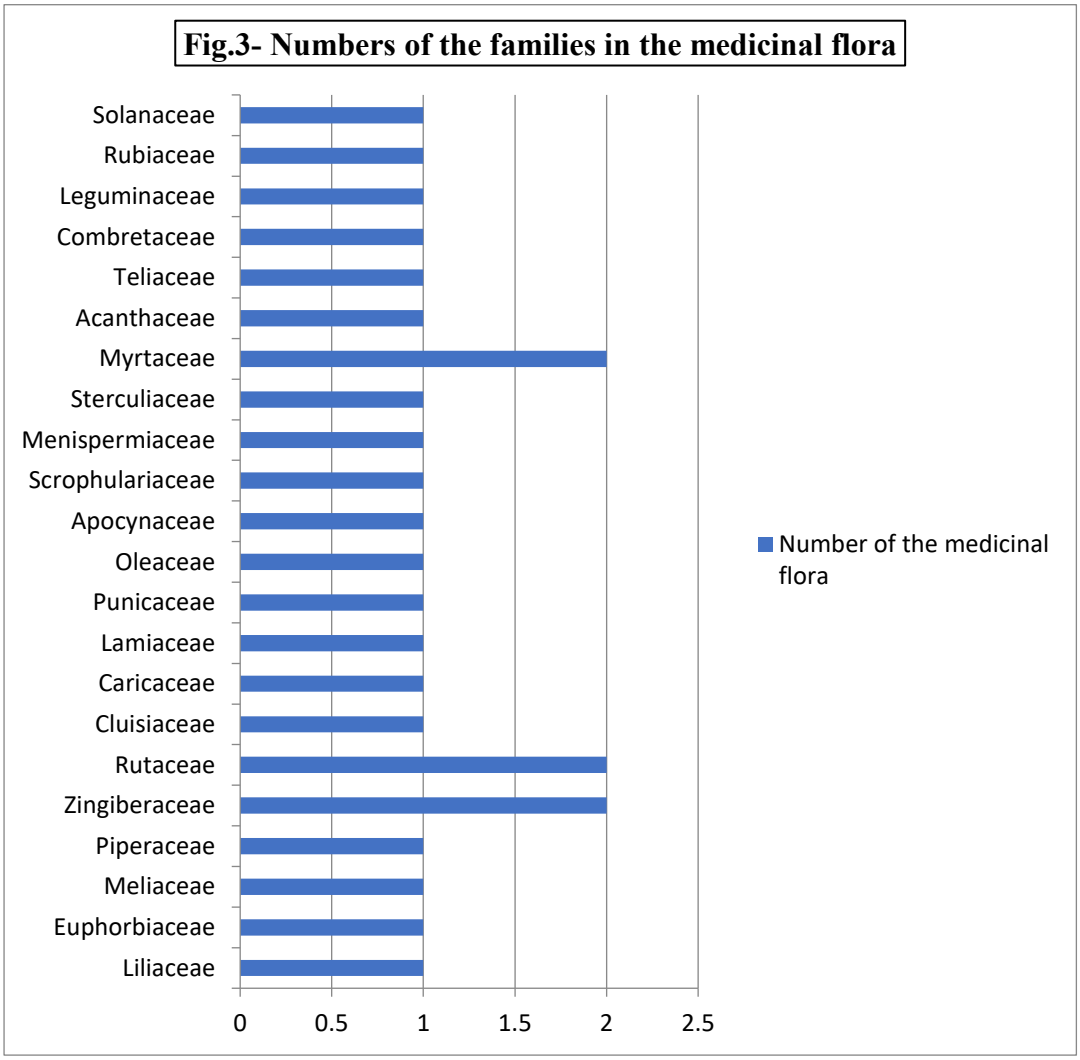
23	Sou.P.P. Meghane	*Zingiber officinale* Roscoe Aale (Family- Zingiberaceae) Shrub, Zingiberine,gingerol,shogaols,zingerone,paradol,curcumene,bergamotene, camphene,bisabolene,bourbormene,borneol,acetate,calamene,cedrol,citral, citronellol.	3.7	Sore throat	May-July
24	Sou.P.P. Meghane	*Cinchona offcinalis* L., Dalchin (Family-Rubiaceae) Tree, quinine and quinidine.	6.1	Bark powder with cheese cure Malarial fever	Sept.-Dec.
25	Sou. S.K. Meghane	*Solanum virginianum* L., Belvange(Family- Solanaceae) Herb,	**46.0**	Fruit powder cure toothache,cold ,cough	Nov.-May

The present investigation was carried out to explore traditional utilization of medicinal plants. Active principles of plants have always played sustainable role in human welfare by satisfying needs ranging from food to medicines. In our studies tree habit [Fig.-1] and use of fruits [Fig.-2] were dominant in the medicinal flora. We have reported 25 medicinal plants species [Photoplate 1 and 2] from 22 different families [Fig.-3]. In some species the seed coats of the seeds are hard due to which seed germination is inhibited for such types of seeds mechanical and chemical scarification methods can be used [9]. Chemicals used for the treatment of seeds are Gibberellic acid, Indole-3 butyric acid, Maleic hydrazide. Concentrated sulphuric acid. It is essential to conserve and proliferate these plants to avoid the pandemic of covid -19 [1], [3], [5], [6], [7], [12], [14], [15]. The protein content was estimated from the plant parts of the medicinal flora. . The total soluble proteins in the dried fruits of Terminalia *chebula* Retz.Was found to be 66.9g 100^{-1}g fr.wt. Better contents of proteins have been recorded in floal buds of the *Syzygium aromaticum* (L.) Merr. &L.M.Perry. which was **54.1** g 100^{-1}g fr.wtand it was **46.0** g 100^{-1}g fr.wt.in the *Solanum virginianum* L. fruits. The data on estimation of proteins has been presented in Table-1. The values of proteins from our studies

were associated with the proteins content research findings in the medicinal plants studied by . N. G. Mager [5].



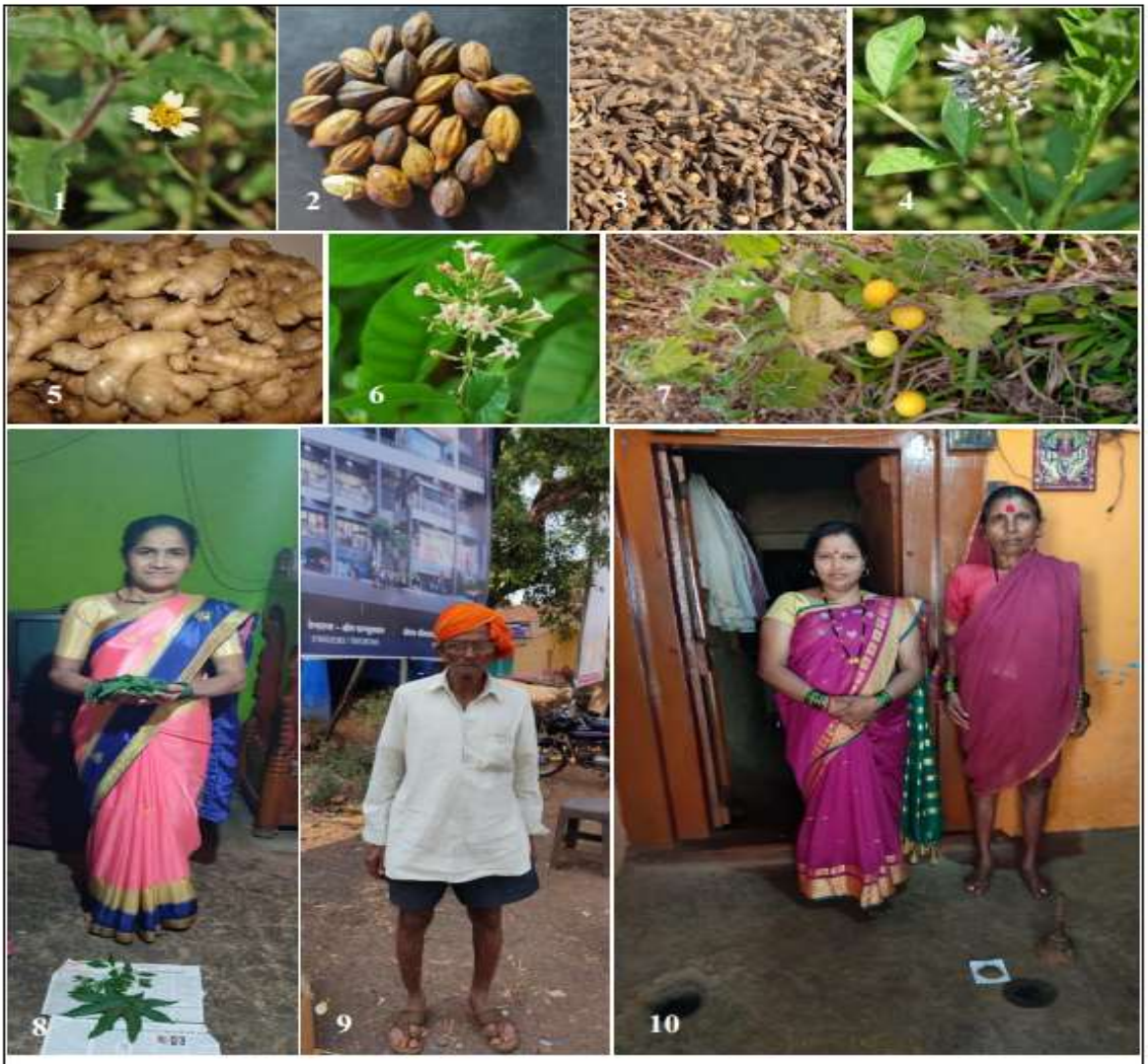




PHOTOPLATE-1-

1 *Allium sativum* L., 2 *Emblica officinalis* Gaerth., 3 *Piper nigrum* L., 4 *Azadirachta indica* A. Juss., 5 *Curcuma longa* L., 6 Citrus aurantium L., 7 *Garcinia indica* Du petit Thou. Choisy., 8 *Centella asiatica* (L.) Urb., 9 *Ocimum tenuiflorum* L., 10 *Punica granatum* Linn., 11 *Nyctanthes arbor-tristis* L., 12 *Holarrhena pubescens* Wall. ex G. Don., 13 *Tinospora cordifolia* (Willd.) Hook.f. & Thomson., 14 *Helicteres isora* L., 15 *Carica papaya* Linn., 16 *Syzygium cumini* (L.) Skeels., 17 *Justicia adhatoda* L., 18 *Aegle marmelos* (L.) Corr.

PHOTOPLATE-2-



1 *Tridax procumbens* L., 2 *Terminalia chebula* Retz., 3 *Syzygium aromaticum* (L.) Merr. & L.M.Perry., 4 *Glycyrrhiza glabra* L., 5 *Zingiber officinale* Roscoe., 6 *Cinchona offcinalis* L., 7 *Solanum heterodoxum* var. *heterodoxum* Dunal., 8 Shrimati S. V. Shinde, 9 Shri. S. T. Ardalkar, 10 Sou P. P. Meghane and Sou. S. K. Meghane.

4. CONCLUSION

Medicinal plants diversity have probable applications in conservation ,cultivation and drug discovery due to occurrence of chemical contents. Various products like churns, ointments poultice , extracts are derived from medicinal flora. In conclusion it can be revealed that the medicinal plant parts were leaves in 6 plants , fruits in 11 plants , stem in 3 plants , rhizome in

2 plants ,bulb in 1 plant, floral bud in 1 plant and bark in 1 plant possess significant amount of proteins to build up theresistant human health against variable diseases.

ACKNOWLEDGEMENT

Authors are thankful to Dr. P. R. Patil, Principal of R.B. MadkholkarMahavidyalaya, Chandgad, District-Kolhapur and Dr. V. S. Dhekale Principal of DattajiraoKadam Arts, Science & Commerce College Ichalkaranji, Dist.-Kolhapur for library and laboratory facilities. PDN is personally thankful to the SARTHI Institute, Pune for financial assistance.We also thankful to the Mrs. Shital Walwadkar for valuable guidance. Dr. APP offer special thanks to Shri. S.N. Patil, Shri. N.P. Chandekar and Shri. S.B. Hasure from R.B. MadkholkarMahavidyalaya, Chandgad, District-Kolhapur for the collection of the plant specimens.

REFERENCES

[1]Boger S, Robert J,Craker LE,LangeD.Medicinal and Aromatic plants.http://ikisan.com.2006.

[2]Home — the plant list [Internet]. Theplantlist.org. [cited 2022 Feb 2]. Available from: http://www.theplantlist.org

[3] Jadhav PM. Kapoor N. Thomas B. Hingorani L. Antiviral potential of selected Indian medicinal plants against herpes simplex virus 1 and 2, North American Journal of medical science.2012; 4, (12): 641-647.

[4] Kapse H.Ayurvedic Medicinal plants. Published by Son publication ,Pune.2003.

[5] Mager NG. Tel v Mede .Published by Secretary,MaharashtraRajyaSahityaanisanskrutiMandal, NavinPrashasanBhavan , Mumbai.1979;1.s

[6] Rao N, Thammanna K.Medicinal Plants of Ritual Hills. Department of Garden, TirupatiDevasathanams, Tirupati, India.1987.

[7] Sabale AB. Mane AA. Influence of maleic hydrazide on growth behavior of*Allium cepa* L.J. Ecobiol. 2003; 15 (6):463-467.

[8] Sharma BD, Karthikeyan S, Singh NP. Flora of Maharashtra State, Monocotyledons, Botanical Survey of India, Flora of India.1996; 2.

[9] Shendge SM, Malpure NV, Pawar NV,Bhise BS, Yadav SR. Quantitative assessment of plant resourses from Parner and Sangamnertahsils of Ahmednagar district (Maharashtra)-National seminar on Plant resources of Western Ghat. “SuvarnaSahyadri”Karnataka biodiversity board, Vanvikas 18 th cross,Malleshwara,Bangalor.2006.
[10] Singh NP, Karthikeyan S. Flora of Maharashtra State. Botanical Survey of India.2000; 1.
[11] Tropicos [Internet]. Tropicos.org. [cited 2022 Feb 2]. Available from: http://www.tropicos.org

[12] Trivedi PC.Medicinal Plants Traditional knowledge, Published by I.K.International Publishing House Pvt.Published by-B.S.I.,P-8,Brabourne Road,Culcutta. 2007.

[13] Yadav SR. Sardesai MM. Flora of Kolhapur District. 2002.

[14] www.google.com- Wikipedia, Online.
[15] http//www.researchgate net.
[16] WHO, IUCN and WWF. Guidelines on the conservation of medicinal plants.Published by IUCN, Gland, Switzerland.1993.
[17] Gornall A.G. , Bardawill C.J. and David M.M., J.Biol.Chem.177.751 Cited by Chaykin S. In: Biochemistry Laboratory Techniques Wiley Eastern Private Ltd. New Delhi., 1949

Design, synthesis, characterization and biological evaluation of some new aryl-pyrazole based chalcones as anticancer agents.

U. B. Chougale[1,2*],P. R. Kharade[2] , H. V. Chavan[3], S. M. Deshmukh[4], K. N. Patil[5], S. R. Dhongade[1*]

1.Research Laboratory in Heterocyclic Chemistry, Devchand College, Arjun-nagar, Tal. Kagal, Dist. Kolhapur, 591269 (MH), India.

2. Department of Chemistry, Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti, Tal. Bhudargad, Dist. Kolhapur, 416209 (MH), India.

3. Department of Chemistry, A S. P. College, Devrukh, Dist. Ratnagiri (Autonomous), 415804 (MH), India.

4. Department of Chemistry, V.N.B.N. Mahavidyalaya, Shirala, Dist. Sangali, 415408 (MH), India.

5. Department of Chemistry, Dr. Ghali College, Gadhinglaj, Tal. Gadhinglaj, Dist. Kolhapur, 416502 (MH), India.

ABSTRACT: In the present work series of aryl-pyrazole based chalcones were synthesized through stepwise manner and screened for their anticancer activity. Initially a precursor compound 5-chloro-3-methyl-1-phenyl-1-H-pyrazole-4-carbaldehyde (**4**)was synthesized from theformylation of 3-methyl-1-phenyl-2-pyrazolin-5-one (**3)**which was obtained from the condensation of starting compounds ethyl aceto acetate (1) and phenyl hydrazine (2). Finally the precursor compound (4) on claisen-schmidt condensation with various active hydrogen compounds yields final chalcone derivatives. The structures of synthesized derivatives were confirmed on the basis of their spectral data and then confirmed structures were screened for

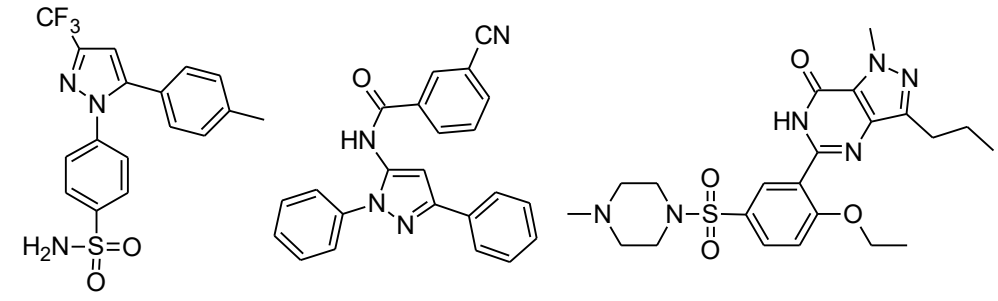
their anticancer activity. Among the tested compounds, compound**6l** Displays potential anticancer activity against MCF-7 breast cancer cell line.

KEYWORDS: Vilsmeier-Haack Reaction, Aryl-pyrazoleChalcones, anticancer, active methylene compound, Breast cancer cell line.

Corresponding Author: Dr. U.B. Chougale* Ph.D.
Department of Chemistry, Karmaveer Hire Arts, Science, Commerce and Education College, Gargoti, Shivaji University, Kolhapur, Maharashtra, India.

1.INTRODUCTION

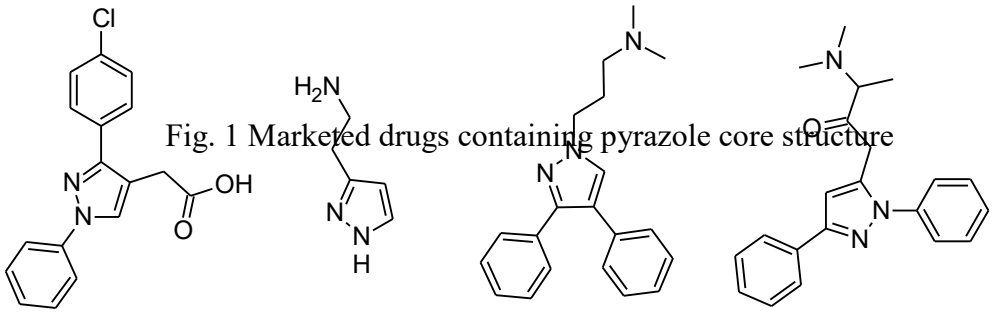
Pyrazole, a five membered two nitrogen containing heterocyclic compound exhibits diversified biological properties like anticancer [1], antimicrobial [2], anti-inflammatory [3], anti-diabetic [4], antifungal [5], anti-depressant [6] and anticonvulsant [7] etc. In last few year several pyrazole derivatives have been synthesized and marketed as Celocoxib (anti-inflammatory), CDPPB (antipsychotic), Sildenafil or Vigra (sexual tonic), Lonazolac (anti-inflammatory), Betazole (H_2-receptor agonist, Fizolamine (anti-depressant), Difenamizole (analgesic) etc.



Celocoxib
anti-inflammatory

CDPPB
anti-psychotic

Sildenafil
Viagra- Sexual tonic



Lonazolac
anti-inflammatory

Betazole
H_2- Receptor agonist

Fezolamine
anti-depressant

Difenamizole
analgesic

Fig. 1 Marketed drugs containing pyrazole core structure

For the synthesis of various pharmaceuticals and agrochemicals, pyrazole and it's well known analogues have been used as basic building blocks to modify and enhance the biological activities of synthesized derivatives. As a result of which pyrazole derivatives with broad spectrum of activities like anticancer [8-12], antimicrobial [13-15], anti-depressant [16, 17], antidiabetic [18], insecticidal [19], **α**-Amylase inhibitors [20], antifungal [21, 22]and analgesic [23-25] activities have been synthesized.

In present study we focused on the synthesis of some novel pyrazole derivatives with anticancer activity. Now a day's cancer is a major health issue in human being as it causes average 13% of all the death. Hence designing and synthesizing new anticancer therapeutic agents is one of the biggest challenge as well as fundamental goal for researchers in the field of medicinal chemistry.

2. MATERIALS AND METHODS

IR spectra were recorded in KBr on FT/IR-4600 type A spectrophotometer. ^{1}H NMR and ^{13}C spectra were recorded in $CDCl_3$ on Bruker 400MHz spectrometer using TMS as an internal standard. Chemical shifts are reported in δ units and the coupling constants (J) are reported in Hertz. Mass spectra were obtained with a Shimadzu LCMS-2010EV. TLC was performed on an alumina backed silica plate with visualization by UV-light. Melting points were determined in open capillary tubes and were uncorrected.

Synthesis of 3-methyl-1-phenyl-2-pyrazolin-5-one (3)

Under Solvent free condition a mixture of phenyl hydrazine (4.3g, 3.94 mL, and 0.04 mol) and ethyl acetoacetate (5.2g, 5.2 mL, 0.04 mol) was taken in a 100 mLround bottom flask and heated at 120ºC with constant stirring for 4h on an oil bath. After completion of the reaction checked on TLC, the reaction mixture was cooled and diethyl ether (20 mL) was added to it. The obtained solid was filtered, washed with diethyl ether and recrystallized from ethanol to obtain the pure product 3-methyl-1-phenyl-2-pyrazolin-5-one **(3)** in excellent yield.

Synthesis of 5-chloro-3-methyl-1-phenyl-1*H* pyrazole-4-carboxaldehyde (4) by Vilsmeier-Haack formylation reaction

Vilsmeier-Haack formylationof a mixture of 3-methyl-1-phenyl-2-pyrazolin-5-one **3** (2.205g, 0.018 mol) and dimethyl-formamide (DMF) (10 mL, 0.13 mol) was carried out in a three-neck round-bottomed flask equipped with reflux condenser under an inert atmosphere. The reaction mixture was cooled at 0ºC and treated with $POCl_3$ (4.6g, 2.8mL, 0.03 mole), maintaining the temperature between 10-15ºC. After complete addition, the reaction mixture was heated on a water bath for about 3h, cooled, and poured into ice water with vigorous stirring to obtain the desired compound 4 in good yield. The product obtained was recrystallized from ethanol as yellow needles.

General procedure for the synthesis of aryl-pyrazole based chalcones (6a-l)

In a round bottomed flask, a mixture of compound 5-chloro-3-methyl-1-phenyl-*1H*-pyrazole-4-carboxaldehyde **(4)** (0.220 g, 1 mmol) and active hydrogen compound (**5**)(1 mmol) was dissolved in ethanol (15 mL) under stirring. To this solution sodium hydroxide (0.12 g, 3 mmol) was added dissolved in minimum quantity of water and stirring continued for 2-3h. The completion of reaction was checked by TLC. After completion of reaction, the solid product obtained was filtered-off and washed with little cold ethanol. The crude product was dried and recrystallized from ethanol to get desired product (**6**) in pure form.

Spectral data of representative compounds

(3*Z*)-3-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-1,3-dihydro-2*H*-indol-2-one (6a)

FT-IR υ_{max}cm^{-1}: 615, 687, 781, 899, 999, 1221, 1366, 1460, 1500, 1613, 1708, 1739, 3019, 3328.**^{1}H NMR (CDCl$_3$, 400 MHz):**δ2.32 (s, 3H, ***-CH$_3$Pyr***), 6.91(d, J= 8Hz, 1H, ***-ArHoxindole***, 6.98 (d, J= 8Hz, 1H, ***-ArHoxindole***), 7.20 (d, J= 7.6 Hz, 1H, ***-ArHoxindole***), 7.24 (d, J= 7.6 Hz, 1H, ***-ArHoxindole***), 7.64 (dd, J= 1.2 Hz, J= 8Hz, 2H, ***-ArH***), 7.46 (d, J= 7.2 Hz, 1H, ***-ArH***), 7.50-7.56 (m, 3H, ***2 x –ArH, 1 =CH***), 8.23 (bs, 1H, ***-NH-***). **^{13}C NMR (CDCl$_3$100 MHz):** 13.79, 109.98, 114.53, 121.90, 122.05, 124.12, 124.53, 124.83, 124.91, 127.12, 128.53, 129.05, 129.17, 129.29, 129.86, 137.83, 141.39, 149.40, 169.46.**MS (EI)** ***m/z:*** 335.95 $(M)^+$

5-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-1,3-diazinane-2,4,6-trione (6b)

FT-IR υ_{max}cm^{-1}: 681, 763, 882, 971, 1027, 1156, 1325, 1535, 1613, 1680, 2930, 3346. **^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.29 (s, 3H, ***Pyr-CH$_3$),***7.44 (d, J =7.1 Hz, 1H, ***-ArH***), 7.49 – 7.55 (m, 3H, ***2-ArH, 1 =CH***), 7.62 (dd, J = 7.2 Hz, 1.2 Hz, 2H, ***2-ArH***), 8.29 (bs, 2H, ***2-NH***-). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.33, 118.7, 121.8, 122.03, 124.7, 129.20, 129.46, 130.20, 132.6, 133.0, 139.4, 145.2, 150.4, 166.70, 167.76. **MS (EI) *m/z:*** 330.65 $(M)^+$

5-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-1,3-dimethyl-1,3-diazinane-2,4,6-trione (6c)

FT-IR υ_{max}cm^{-1}: 688, 693, 736, 893, 976, 1019, 1165, 1245, 1389, 1557, 161, 1684, 2981. **^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.28 (s, 3H, ***Pyr-CH$_3$***), 3.6 (s, 6H, 2 x ***N-CH$_3$***), 7.42 (d , J = 7.2 Hz, 1H -***ArH***), 7.51-7.56 (m, 3H, ***2 x -ArH***, 1 ***=CH***), 7.62 (dd, J = 7.2 Hz, 1.2 Hz, 2H, ***2 x -ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.45, 28.2, 29.10, 118.7, 121.89, 122.32, 124.7, 129.12, 129.92, 130.45, 132.6, 133.0, 139.4, 145.2, 151.2, 164.4, 168.22. **MS (EI) *m/z:*** 358.90 $(M)^+$

2-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl-methylidene]-5,5-dimethylcyclohexane-1,3-dione (6d)

FT-IR υ_{max}cm^{-1}: 617, 678, 718, 989, 1024, 1212, 1363, 1406, 1517, 1622, 1686, 2889, 965, 3024.**^{1}H NMR (CDCl$_3$ 400 MHz):**δ 1.5 (s, 6H, 2 x –***CH$_3$ Dimedone***), 2.31 (s, 3H, ***Pyr-CH$_3$***), 3.44 (s, 4H, 2 x –***CH$_2$Dimidone***), 7.42 (d, J = 7.2 Hz 1H –***ArH***), 7.46-7.52 (m, 3H, ***2x ArH***, 1 ***=CH***), 7.60 (dd, J = 7.1 Hz, 1.1 Hz, 2H, ***ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 14.25, 29.56, 30.18, 32.0, 52.66, 54.84, 124.7, 128.8, 129.34, 129.34, 129.87, 131.43, 132.66, 133.80, 135.08, 139.4, 145.2, 187.4, 195.65. **MS (EI) *m/z:*** 342.80 $(M)^+$

(4*Z*)-4-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-5-methyl-2-phenyl-2,4-dihydro-3*H*-pyrazol-3-one (6e)

FT-IR υ_{max}cm^{-1}: 1027, 1130, 1185, 1265, 1308, 1389, 1425, 1587, 1670, 1685, 2885, 3017. **^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.35 (s, 3H, ***Pyr-CH$_3$),*** 2.57 (s, 3H, ***-CH$_3$***), 7.44 – 7.49 (m, 6H, 5 x -***ArHPyr***, 1 ***=CH***), 7.53 – 7.58 (m, ***5H, -ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.45, 15.45, 119.08, 12.21, 121.8, 122.10, 124.7, 126.32, 129.2, 129.22, 129.26, 129.82, 130.12, 131.60, 132.6, 133.0, 139.2, 139.4, 145.2, 147.6, 167.93. **MS (EI) *m/z:*** 376.95 $(M)^+$

(2*E*)-3-(5-Chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-3,4-dihydronapthalen-1(2H)-one (6f)

FT-IR υ_{max}cm^{-1}: 736, 862, 976, 1128, 1324, 1463, 1598, 1628, 1685, 2880, 2917, 3001. **^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.39 (s, 3H, ***-Pyr-CH$_3$*)**, 3.22 (t, 2H, -CH$_2$-***CH$_2$***-), 3.92 (t, 2H, =***C-CH$_2$***-), 7.42 – 7.46 (m, 2H, ***-ArHtetralone***), 7.50 – 7.93 (m, 8H, ***7 x –ArH, 1 =CH***-). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.30, 31.04, 32.04, 121.8, 122.46, 124.7, 127.6, 128.3, 128.5, 129.2, 130.43, 131.32, 132.4, 132.6, 133.0, 134.2, 139.4, 140.7, 142.9, 145.2, 190.30. **MS (EI)** ***m/z:*** 348.48 $(M)^+$

(5*Z*)-5-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-1,3-thiazolidine-2,4-dione (6g)

FT-IR υ_{max}cm^{-1}: 872, 917, 1165, 1270, 1389, 1452, 1578, 1654, 1688, 2898, 2965, 3025, 3289, 3465. **^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.32 (s, 3H, ***Pyr-CH$_3$),***7.42 (s, 1H, ***=CH***), 7.46 (d J = 7.2 Hz, 1H, ***-ArH***), 7.51- 7.56 (m, 2H, ***-ArH***), 7.64 (dd, J = 8 Hz, 1.2 Hz, 2H, ***-ArH***), 8.16 (bs, 1H, ***-NH-***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.22, 121.97, 122.89, 124.7, 124.9, 129.1, 129.33, 130.67, 131.43, 132.6, 139.4, 145.2, 167.6, 169.89. **MS (EI)** ***m/z:*** 319.90 $(M)^+$

(5*Z*)-5-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-2-sulanylidene-1,3-thiazolidin-4-one (6h)

FT-IR υ_{max}cm^{-1}: 763,827,971, 1156, 1275, 1398, 1425, 1587, 1623, 1659, 1689, 2963, 3017, 3265, 3430.**^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.32 (s, 3H, ***Pyr-CH$_3$),***7.36 (s, 1H, ***=CH***), 7.46 (d J = 7.2 Hz, 1H, ***-ArH***), 7.51- 7.56 (m, 2H, ***-ArH***), 7.64 (dd, J = 8 Hz, 1.2 Hz, 2H, ***-ArH***), 7.98 (bs, 1H, ***-NH-***).**^{13}C NMR (CDCl$_3$ 100 MHz):** 14.23, 121.83, 122.32, 124.7, 124.98, 129.03, 129.37, 130.21, 130.88, 132.6, 139.4, 145.2, 169.8, 196.1.**MS (EI)** ***m/z:*** 336.10 $(M)^+$

(*2E*)-2-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-2,3-dihydro-1*H*-inden-1-one (6i)

FT-IR υ_{max}cm^{-1}: 1091, 1112, 1182, 1267, 1324, 1415, 1598, 1629, 1689, 2921, 2987, 3043.**^{1}H NMR (CDCl$_3$, 400 MHz):**δ2.44(s, 3H, ***-CH$_3$*)**, 3.94 (s, 2H, ***-CH$_2$-*)**, 7.42-7.47 (m, 2H, ***-ArHindanone***), 7.50-7.91 (m, 7H, ***6 x –ArH, 1 x =CH***), 7.92 (d, J= 7.6 Hz, 1H, ***-ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 14.00, 32.52, 114.83, 123.13, 124.47, 125.09, 126.13, 126.80,

127.60, 128.56, 129.12, 134.69, 136.35, 137.88, 138.05, 149.67, 149.89, 193.65. **MS (EI)** ***m/z:***335.20$(M)^+$

(2*E*)-2-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-6-methoxy-2,3-dihydro-1*H*-inden-1-one (6j)

FT-IR υ_{max}cm^{-1}:1024, 1103, 1166, 1218, 1280, 1357, 1380, 1492, 1590, 1621, 1689, 3000, 3064.**^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.41 (s, 3H, ***Pyr-CH$_3$***), 3.86 (s, 2H, ***-CH$_2$-***), 3.88 (s, 3H, ***-OCH$_3$***), 7.22 (dd, J= 2.8 Hz, 8.4 Hz, 1H, ***-ArH indanone***), 7.37 (d, J= 2.4 Hz, 1H, ***ArH indanone***), 7.42 (d, J= 8.8 Hz, 1H, ***-ArH indanone***), 7.46 (dd, J= 1.6 Hz, 7.2Hz, 1H, ***-ArH***,), 7.50-7.54 (m, 3H, ***2 x -ArH, =CH***), 7.57-7.59 (m, 2H, ***-ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):**14.03, 31.82, 55.67, 105.77, 114.82, 123.0, 124.01, 125.08, 126.84, 126.87, 128.55, 129.12, 137.11, 137.88, 139.23, 142.50, 149.86, 159.52, 193.60. **MS (EI)** ***m/z:*** 365.15 $(M)^+$

(2*E*)-2-[(5-chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)-methylidene]-5,6-dimethoxy-2,3-dihydro-1*H*-inden-1-one (6k)

FT-IR υ_{max}cm^{-1}: 1025, 1095, 1128, 1253, 1309, 1376, 1417, 1455, 1496, 1589, 1631, 1691, 2834, 3004, 3193. **^{1}H NMR (CDCl$_3$ 400 MHz):**δ 2.42 (s, 3H, ***Pyr-CH$_3$***), 3.84 (s, 2H, ***-CH$_2$-***), 3.95 (s, 3H, ***-OCH$_3$***), 3.99 (s, 3H, ***-OCH$_3$***), 6.95 (s, 1H, ***-ArH indanone***), 7.34 (s, 1H, ***-ArH indanone***), 7.44 (m, 2H, ***-ArH***), 7.49-7.52 (m, 2H, ***-ArH***), 7.56-7.58 (m, 2H, ***-ArH, =CH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.99, 32.16, 56.20, 56.30, 105.08, 107.14, 114.91, 121.51, 125.07, 126.55, 128.0, 128.91, 129.11, 131.05, 137.09, 137.93, 144.97, 149.57, 149.78, 155.47, 192.42. **MS (EI)** ***m/z:***394.90 $(M)^+$

(2E)-5-Bromo-2-[(5-chloro-3-methyl-1-phenyl-1H-pyrazol-4-yl)-methylidene]-2,3-dihydro-1H-inden-1-one (6l)

FT-IR υ_{max}cm^{-1}: 763, 826, 967, 1182, 1342, 1436, 1589, 1618, 1690, 2927, 3018.**^{1}H NMR (CDCl$_3$, 400 MHz):**δ 2.41 (s, 3H, ***Pyr-CH$_3$***), 3.86 (s, 2H, ***-CH$_2$-***), 7.20 (dd, J = 2.8 Hz, 8.4 Hz, 1H, ***-ArH indanone***), 7.34 (d, J= 2.4 Hz, 1H, ***ArH indanone***), 7.41 (d, J= 8.8 Hz, 1H, ***-ArH indanone***), 7.46 (dd, J= 1.6 Hz, 7.2Hz, 1H, ***-ArH***,), 7.50-7.54 (m, 3H, ***2 x -ArH, =CH***), 7.57-7.59 (m, 2H, ***-ArH***). **^{13}C NMR (CDCl$_3$ 100 MHz):** 13.2, 35.8, 121.8 x 2, 124.3, 124.7, 124.8,

126.6, 129.0, 129.2 x 4, 132.6, 133.0, 139.4, 139.5, 145.0, 145.2, 191.9. **MS (EI)** ***m/z:*** 413.55 $(M)^+$

MTT ASSAY FOR ANTICANCER ACTIVITY

The cells were seeded at a density of approximately 5×10^3cells/well in a 96-well flat-bottom microplate and maintained at 37ºC in 95% humidity and 5% CO_2 overnight. Different concentration (500, 400, 300, 200, 100, 50 µg/ml) of samples was treated. The cells were incubated for another 48 hours. The cells in well were washed twice with phosphate buffer solution, and 20µL of the MTT staining solution (5mg/ml in phosphate buffer solution) was added to each well and plate was incubated at 37°C. After 4h, 100 µL of dimethyl sulfoxide (DMSO) was added to each well to dissolve the formazan crystals, and absorbance was recorded with a 570nm using a microplate reader (1, 2).

Surviving cells (%) = Mean OD of test compound / Mean OD of Negative control ×100

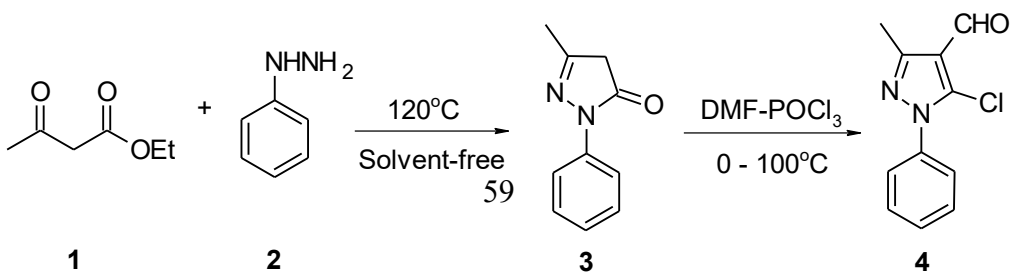
Using graph Pad Prism Version 5.1, we calculated the IC_{50} values of compounds.

Note: DMSO Concentration is less 1.5% in experiments. Concentrations are in duplicates.

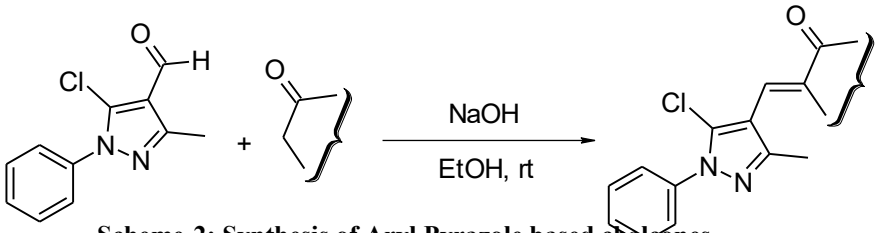
3. RESULTS AND DISCUSSION

CHEMISTRY

The synthesis of target molecules (**6a-l)** was achieved by the Claisen-Schmidt condensation of 5-chloro-3-methyl-1-phenyl-*1H*-pyrazole-4-carbaldehyde (**4**) with active hydrogen compound (**5**) in the presence of sodium hydroxide in ethanol in good to excellent yield (Scheme-2). The precursor 5-chloro-3-methyl-1-phenyl-*1H*-pyrazole-4-carbaldehyde (**4**) was synthesized by Vilsmeier-Hack formylation of 3-methyl-1-phenyl-2-pyrazolin-5-one (**3**). The synthesis of starting compound 3-methyl-1-phenyl-2-pyrazolin-5-one (**3**) was accomplished under solvent-free condition by the condensation of ethyl acetoacetate (**1**) and phenyl hydrazine (**2**) at 120ºC (Scheme-2). The structural investigation of the synthesized compounds was carried out using IR, ^{1}H NMR and mass spectral data. The structures of synthesized compounds are presented in Table-1.



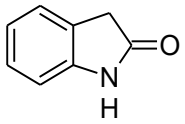
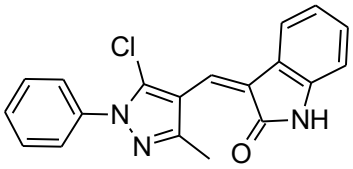
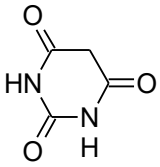
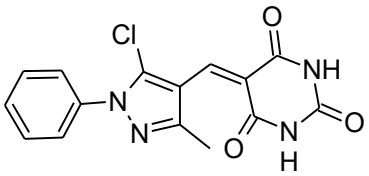
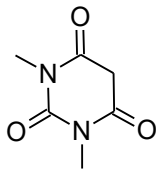
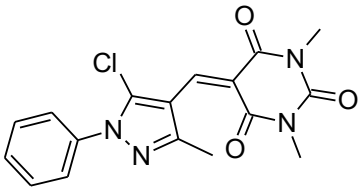
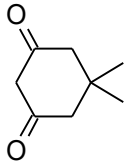
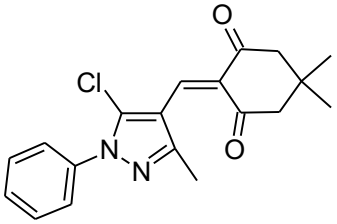
Scheme-1: Synthesis of Precursor Aryl Pyrazole Aldehyde, 4

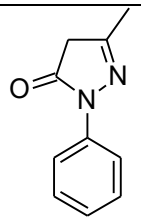
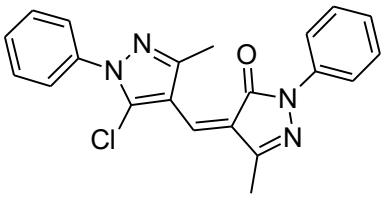
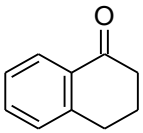
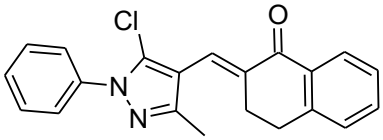
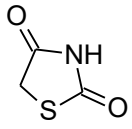
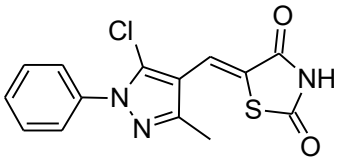
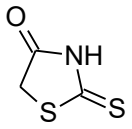
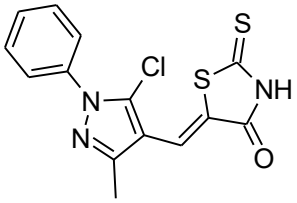
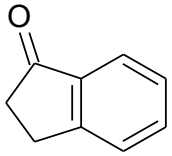
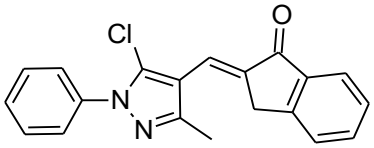
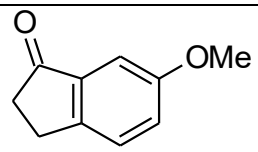
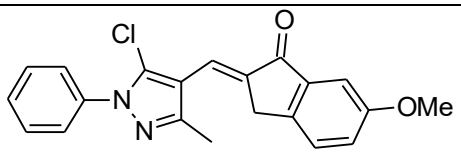
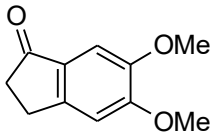
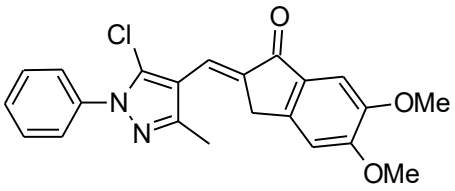
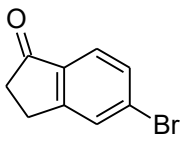


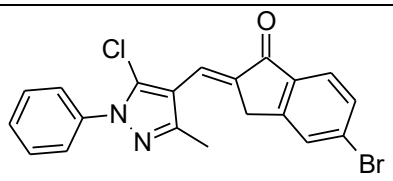
Scheme-2: Synthesis of Aryl Pyrazole based chalcones

4 **5** **6a-l**

Table-1: Structures of the Aryl Pyrazole based chalcones

Comp. Code	Active Hydrogen Compounds Entry	Product 6	Yield %	M.P. ^{0}C
6a			83	172-175
6b			72	190-192
6c			82	162-164
6d			83	188-191
				142-145

6e			74	
6f			71	176-178
6g			68	168-170
6h			68	118-120
6i			93	116-118
6j			78	140-142
6k			81	182-186
6l			87	170-172

BIOLOGICAL EVOLUATION

ANTICANCER ACTIVITY

From the synthesized compounds some selected structures were screened for their anticancer potential against breast carcinoma (MCF-7) using MTT assay method using paclitaxel as a reference standard drug. The results are summarized in Table-2 and 3. The IC_{50} values revels that compound **6l**, **6a** and **6e** have shown moderate anticancer activities (IC_{50} 47.01-62.5µM) and all other compounds displayed poor anticancer activities against MCF-7.

Table 2 Anti breast cancer activities of Aryl pyrazole based chalcones

(IC 50 values in µM)

Sr. No.	**Compound Code**	**MCF-7 IC_{50} values in µM**
1	Chalcone 6a	53.91
2	Chalcone 6e	62.5
3	Chalcone 6h	50.84
4	Chalcone 6i	383.7
5	Chalcone 6k	318.1
6	Chalcone 6l	47.01
7	**Paclitaxel**	**0.35**

Table 3 Cell Viability study (MCF-7)

Conc. µg/mL	Chalcone 6a	Chalcone 6e	Chalcone 6h	Chalcone 6i	Chalcone 6k	Chalcone 6l
500	24.86	30.00	26.14	37.29	32.00	33.00
400	31.29	35.14	29.14	55.71	44.29	37.71
300	33.86	40.29	38.57	57.43	55.00	38.57
200	36.36	40.29	39.43	58.71	63.00	42.00
100	39.86	46.29	44.57	60.71	81.79	44.57
50	52.50	51.00	48.00	68.14	95.07	49.29

NC		100

4. CONCLUSION

In conclusion, we have synthesized aryl pyrazolechalcone molecules by combining pyrazole aldehyde with various active hydrogen compounds under basic conditions. The results of the anticancer study reveal that compounds 6l, 6a and 6e showed moderate anticancer potential against MCF-7 with IC_{50} values 47.01-62.5µM. among tested compounds, chalcone6l with electron withdrawing bromo substituent on indanone (active hydrogen compound) showed potential anticancer activity.

ACKNOWLEDGEMENT

One of the authors, Dr. U. B. Chougale thanks to Principal, Karaveer Hire College, Gargoti for providing necessary laboratory facilities. Also thankful to NCL, Pune, Shivaji University, Kolhapur for providing characterization facilities and MarathaMandal's Central Research Laboratory, Belgum for screening of biological activities.

CONFLICT OF INTEREST

Authors have no conflict of interest.

REFERENCES

1. Abadi AH, Haleem EAA and Hassan GS. Synthesis of Novel 1,3,4-Trisubstituted Pyrazole Derivatives and Their Evaluation as Antitumor and Antiangiogenic Agents.*Chem. Pharm. Bull.,*2003;51(7):838-844.
2. Dabhi HR, Rana AK and Parmar KKH. Synthesis, Characterization and Antimicrobial Study of Some Pyrazole Compounds Derived from Chalcone.Arch. Appl. Sci. Res.,**2015**; 7(3): 1-5.
3. Thore SN, Gupta SV and Baheti KG. Novel ethyl-5-amino-3-methylhio-1H-pyrazole-4-carboxylates: Synthesis and Pharmacological Activity.J. Saudi Chem. Soc., 2016; 20: 259-264.
4. Parajuli R, Banerjee J and Khanal H. Synthesis of Some Pyrazolone Derivatives and Evaluation of its Antibacterial and Cytotoxic Activity. Orient. J. Chem.,**2015**; 31(4): 2099-2106.
5. Radi S, Tighadouini S, Feron O, Riant O,Bouakka M, Benabbes R and Mabkhot YN. Synthesis of Novel β-Keto-Enol Derivatives Tethered Pyrazole, Pyridine and Furan as

New Potential Antifungal and Anti-Breast Cancer Agents. Molecules,2015,; 20: 20186-20194.

6. Merugumolu VK and Chandrashekarappa RB. Synthesis and anti-depressant Evaluation of Novel Pyrazolone Derivatives.Bangladesh J. Pharmacol., 2016; 11:558-563.
7. BeyhanNagihan, Kocyigit-KaymakciogluB, Gumru S, Aricioglu F.Synthesis and anti-Convulsant Activity of Some 2-Pyrazolines Derived from Chalcones. Arab. J. Chem.,2017; 10:2073-2081.
8. Nitulescu GM, Draghici C and Olaru OT.New Potential Anti-tumor Pyrazole Derivatives: Synthesis and Cytotoxic Evaluation. Int. J. Mol. Sci.,2013; 14:21805-21818.
9. Mohareb RM, EI-Sayed NNE and Abdelaziz MA. Use of Cyanoacetylhydrazine in Heterocyclic Synthesis: Novel Synthesis of Pyrazole Derivatives with Anti-tumor Activities. Molecules,2012; 17:8449-8463.
10. Gouhar RS, Fathalla OA and Abd EI-Karim SS. Synthesis and Anticancer Screening of Some Novel Substituted Pyrazole Derivatives. Der. PharmaChemica,2013; 5(6): 225-233.
11. Puneeth HR, Ananda H, Sharath Kumar KS, Rangappa KS, Sharada AC.Synthesis and Anti-proliferative Studies of Curcumin Pyrazole Derivatives. Med. Chem. Res., 2016; 25: 1842-1851.
12. Gergo M, Laszlo M, Marton AK, Zsuzsanna S, Izabella S, Istav Z and Eva F.Microwave-assisted Synthesis of Biologically Relevant Steroidal 17-exo-Pyrazol-5-ones from a Norpregnene Precursor by a Side-chain Elongation/Heterocyclization Sequence. BeilsteinJ.Org. Chem., 2018; 14:2589-2596.
13. Beyzaei H, Motraghi Z, Aryan R, Zahedi MM and Samzadeh-Kermani A.Green One-pot Synthesis of Novel PolysubstitutedPyrazole Derivatives as Potential Antimicrobial Agents. Acta. Chim. Slov.,2017; 64:911-918.
14. Parmar N, Teraiya S, Patel R, Barad H, Jajda H, and Thakkar V. Synthesis, Antimicrobial and Antioxidant Activities of Some 5-Pyrazolone Based Schiff Bases.J. Saudi Chem. Soc.,2015; 19:36-41.
15. Reddy GM, Garcia JR, Zyryanov GV, Sravya G and Reddy NB.Pyranopyrazole as Efficient Antimicrobial Agents: Green One Pot and Multicomponent Approach. J. Bioorg. Chem.,2019; 82:324-331.

16. Patil PO and Bari SB. Synthesis, Characterization and Screening for Anti-Depressant and Anti-Convulsant Activity of 4,5-Dihydro-Pyrazole Bearing Indole Derivatives.Arab. J. Chem.,2016; 9:588-595.
17. Kumar RS, Arif IA, Ahamed A and Idhayadhulla A. Anti-Inflammatory and Anti-microbial Activities of Novel Pyrazole Analogues.Saudi J. Biol. Sci.,2016; 23:614-620.
18. Kenchappa R, Yadav DB, Chandrashekar A, Sindhe MA, Peethambar SK. Synthesis of Coumarin Derivatives Containing Pyrazole and Indenone Rings as Potent Anti-Oxidant and Anti-Hyperglycemic Agents. Arab. J. Chem.,2017; 10:3895-3906.
19. Dai H, Yao E, Fang Y, Sun S, Shi Y, Chen J, Jiang G, Shi J. Design, Synthesis and Bioactivities of Novel Isoxazole-Containing Pyrazole Oxime Derivatives.Molecule*s,* 2017; 22:2000-2014
20. Yousuf S, Khan KM, Salar U, Chigurupati S, Muuhammad MT, Wadood A, Aldubayan M, Vijayan V, Riaz M, Perveen S.Eur. 2-Aryl and 4-Arylidine Substituted Pyrazolones: As Potential α amylase Inhibitors. J. Med. Chem.,2018; 159:47-58.
21. Zhang CY, Liu XH, Wang SH, Li ZM. Synthesis and Antifungal Activities of New Pyrazole Derivatives via 1,3-Dipolar Cycloaddition Reaction.Chem. Biol. Drug Des.,2010; 75:489-493.
22. Patil A, Jadhav R, Raundal H, Sharma L, Badgujar R, Bobade V.Synthesis and Antifungal activities of DiarylPyrazolesCarboxamide Derivatives. J. Chem. Pharm. Res.,2014; 6(8):218-223.
23. Marappan G, SahaBp, Sutharson L, Singh A, Garg S, Pandey L, Kumar D. Analgesic, Anti-inflammatory, Anti-Pyretic and Toxicological Evaluation of Some newer 3-Methyl Pyrazolone Derivatives. Saudi Pharma. J., 2011; 19:115-122.
24. Thore SN, Gupta SV and Baheti KG. Novel Ethyl-5-amino-methylthio-1H-Pyrazole-4-Carboxylates: Synthesis and Pharmacological Activity. J. Saudi Chem.Soc., 2016; 20:259-264.
25. Selvam TP, Kumar PV, Saravanan G, Chinnasamy RP. Microwave Assisted Synthesis, Characterization and Biological Activity of Novel Pyrazole Derivatives. J. Saudi Chem.Soc., 2014; 18:1015-1021.

Review of Ethnofloristic diversity of Kharepatan village, Sindhudurg.

Prajyot S. Nalawade[1], Sharmin H. Kazi[1], Mangal A. Parab[1], Ruchi N. Teli[1], Kavita P. Amkar[1] and Pratik D. Natekar[2*]

1. Department of Botany, Arts, Commerce & Science College Kharepatan, Tal.-Kankavali, Dist.-Sindhudurg, Maharashtra, India.
2.Department of Botany,DattajiraoKadam Arts, Science & Commerce College, Ichalkaranji,Tal.-Hatkanangale, Dist.-Kolhapur, Maharashtra, India.

ABSTRACT: Kharepatan village is a historically and commercially important village in Sindhudurg district. The area of village is compactly covered with semi evergreen forest. People used variety of plants for the medicinal purpose and on other hand fluctuating global

environment is adversely affecting on plant treasure. Due to the lack of knowledge of medicinal properties of these plants, they are being neglected and overused. This study carries variable source of information for traditional medical experts and plant researchers. In this paper we have tried to enlist ethnofloristic diversity of Kharepatan village. Present investigation revealed that 71 Species of 70 Genera belonging from 46 families have been used as medicinal plants. Among this researchApocynaceae is more dominant family which comprises 8 genera & 8 species. In this point of view, we have try to document more medicinal plants with their medicinal properties from this village.

KEYWORDS: Diversity,Ethnofloristic, Kharepatan, Medicinal plants, Survey.

Corresponding Author:Pratik D. Natekar*
Department of Botany,DattajiraoKadam Arts, Science & Commerce College, Ichalkaranji, Tal.-Hatkanangale,Dist.-Kolhapur, Maharashtra, India.

1.INTRODUCTION

Sindhudurg is the last district on the southern coast of Maharashtra. Kharepatan was well known for its historical port and trade. This village is rich in nature and historical heritage, located at a distance of about 39 km from Kankavali tehsil. Kharepatan village is also known as the gateway of Sindhudurg district and national highway (NH-66) divided it into two large parts. Area of this village is 835 hectares and surrounded by mountains from all four sides. The village is endowed with natural beauty. Geological co-ordinates of Kharepatan are $16^0$55’69’’N, $73^0$62’57’’E and altitude is 85 feet above sea level. The climate is hot and humid with an average temperature up to 30^o C. This region experiences significant seasonal variations in rainfall [26].

The demand for medicinal plants is increasing day by day and this reflects the need to study and preserve diversity of medicinal plants [8]. It is cherished place for researchers and biologists as large number of medicinal plants, rare endemic plants and animals are found in this region [10]. Among the investigation so far we have registered the important families, number of the genera and species as well as medicinal importance of plants which are being used by the people of Kharepatan village. The study area is diverse with variety of plants and animals. Diversity of Western Ghats measures 4500 species of higher plants and about 2000 species are endemic to Western Ghats [7]. The combined topography and heavy rainfall helps to preserve its regional diversity. Therefore, it has become imperative to search, register medicinal plant and necessary to check their current status [15]. Local natural geographical constraints of Kharepatan provides unique favourable environment with regards to construct

diversity of plants [14]. Sindhudurg is one of the popular mega diversity zones in Maharashtra [3], [19]. In this paper we mainly focus on traditional use of medicinal plants by native peoples. The study delivers a veritable source of information for traditional medical experts and plant researchers.

2. MATERIALS AND METHODS

The first step studies assessed by field surveys were carried out for exploration of the study area. Intensive and extensive field tours conducting during different seasons in study area. The second step of study followed by, collected plant specimen was recorded with scientific information and proceeding for identification. With the help of regional floras and related published literature, plants were properly identified such as [5], [9], [13], [17], [18], [22], [24], [25].The nomenclature of plant species collected was updated by using IPNI, Tropicos and The Plant List available on websites. The aim of field visiting method is to attempts more field information with respect to the local names of plants and there medicinal values.The obtained data was cross checked with accessible literature about these medicinal plants and their Ethnobotany [1], [2], [4], [6], [11], [12], [21].

(Table 1). DetailedList of Ethnofloristic survey in Kharepatan region

Sr. No.	Botanical Name	Local Name	Family	Part used	Medicinal Use	Reference
1	*Andrographis paniculata* (Burm.f.) Nees	Bhuineem	Acanthaceae	Leaves, Stem	Fever, Antiseptic	[4], [12], [19]
2	*Barleriaprionitis* L.	Kate-Koranti	Acanthaceae	All plant parts	Strengthens Teeth, Toothache, joint pains, lung diseases, Fever,	[3], [6], [20]
3	*Justicia adhatoda* L.	Adulsa	Acanthaceae	Leaves, Bark	Cough, Other respiratory ailments like Asthma, Bronchitis	[6], [8], [21]
4	*Achyranthes aspera* L.	Aghada	Amaranthaceae	Root, Seed	Gynecological Disorders, Indigestion,	[4], [3], [8]

					Cough, Asthma, Anemia, Jaundice	
5	*Centella asiatica* (L.) Urb.	EkpaniBramhi	Apiaceae	Leaves, Stem	Anti-ulcer genic, Anxiolytic	[11], [8], [20]
6	*Alstonia scholaris* (L.) R. Br.	Saptaparni	Apocynaceae	Leaves, bark	Anti-diabetic, Fever, Skin ulcers, Increasing lactation	[6], [19], [21]
7	*Catharanthus roseus* (L.) G. Don	Sadafuli	Apocynaceae	Leaves, Roots	Anti-diabetic	[4], [10], [19]
8	*Cynanchumannularium* (Roxb.) Liede&Khanum	Utran	Apocynaceae	Root	Anti-diabetic	[11], [14], [10]
9	*Gymnemasylvestre* (Retz.) R. Br. ex Sm	Bedkicha Pala, Gudmar	Apocynaceae	Leaves	Anti-diabetic, hypertension	[12], [14], [21]
10	*Hemidesmus indicus* (L.) R. Br.	Anantmul	Apocynaceae	Root	Digestive	[6], [19], [21]
11	*Holarrhena pubescens* Wall. ex G. Don	Kuda	Apocynaceae	Bark	Diarrhea	[1], [3], [11]
12	*Plumeriarubra* L.	Chafa	Apocynaceae	Bark	Bark juice used on wound	[8], [12], [21]
13	*Rauvolfia serpentina* (L.) Benth. Ex Kurz	Sarpgandha	Apocynaceae	Root	Snake bites	[10], [14] [19]
14	*Asparagus racemosus*Willd.	Shatawari	Asparagaceae	Rhizome	Medicine for women, Infertility, Loss of Libido, Threatened Miscarriage	[4], [8], [20]
15	*Aloe vera* (L.) Burm. f.	Korphad	Asphodelaceae	Leaves	Skin Care, Ulcers, Burn Injuries,	[1], [10], [21]

					Acne, Jaundice	
16	*Chromolaenacorymbosa* (Aubl.) R. M. King & H. Rob	Ranmodi	Asteraceae	Leaves	Antiseptic	[6], [8], [14]
17	*Eclipta prostrata* (L.) L.	Maka	Asteraceae	Leaves, Stem	Hair treatment, Skin diseases	[11], [14], [19]
18	*Elephantopus scaber* L.	Bhamburda	Asteraceae	Whole plant	Kidney stone	[3], [6], [14]
19	*Tridax procumbens* L.	Dagadi Pala	Asteraceae	Leaves	Wound healing, Kidney stone	[4], [11], [19]
20	*Heterophragma quadriloculare* (Roxb.) K. Schum.	Varas	Bignoniaceae	Leaves	Anti-diabetic, Skin Diseases	[10], [14], [20]
21	*Celastrus paniculatus*Willd.	Malkamni	Celastraceae	Seed, Bark	Animal Bite, Muscle Pain	[3], [8], [19]
22	*Garcinia indica* (Thouars) Choisy	Kokam	Clusiaceae	Leaves, Fruit	Digestive	[4], [12], [21]
23	*Gloriosa superba* L.	KalLavi	Colchicaceae	Tuber, Leaves	Abortifacient, Spleen complaints, sores	[8], [11], [14]
24	*Terminalia bellirica* (Gaertn.) Roxb.	Behda	Combretaceae	Fruit	Expectorant, Stomachic	[12], [19], [20]
25	*Terminalia chebula* Retz.	Hirda	Combretaceae	Fruit	Cough, Stomachic	[1], [4], [21]
26	*Hellenia speciosa* (J. Koenig) S. R. Dutta	Jungliaal	Costaceae	Rhizome	Burns, Constipation, Skin diseases, Hyperlipidemia, Obesity, Diabetes	[6], [12], [14]
27	*Cyperus rotundus* L.	Nagarmotha	Cyperaceae	Tuber	Diarrheal Pathogenesis, Fever, Diabetes,	[1], [10], [21]

					Solar Dermatitis	
28	*Tacca leontopetaloides* (L.) Kuntze	Ransuran	Dioscoreaceae	Tuber	Body ache and Headache	[10], [12], [20]
29	*Dillenia pentagyna* Roxb.	Karmal	Dilleniaceae	Bark	Digestive	[4], [14], [19]
30	*Diospyros ferrea* (Willd.) Bakh.	Kaling	Ebenaceae	Fruits, Leaves, Stem	Antiseptic, Food	[6], [11], [20]
31	*Abrus precatorius* L.	Gunj	Fabaceae	Root, Leaves, Seeds	Skin disease, asthma, Stomatitis, Joint pains, Paralysis, Alopecia	[3], [11], [21]
32	*Cassia fistula* L.	Bahava	Fabaceae	Root, Fruit	Purgative, ulcers, wounds	[1], [8], [14]
33	*Clitoria ternatea* L.	Gokarn	Fabaceae	Leaves, Root	Nephron protective	[6], [20], [21]
34	*Mimosa pudica* L.	Lajalu	Fabaceae	Root	Insomnia, Inflammation	[1], [3], [11]
35	*Mucuna pruriens* (L.) DC.	Khajkuwali	Fabaceae	Leaves, Seeds	Deworming	[10], [14], [21]
36	*Pongamia pinnata* (L.) Pierre	Karanj	Fabaceae	Seeds	Treat wounds	[8], [12], [19]
37	*Tamarindus indica* L.	Chinch	Fabaceae	Fruits, Leaves	Dried fruits are taken orally to treat eye infection, Used to treat ulcer	[4], [6], [21]
38	*Rotheca serrata* (L.) Steane&Mabb.	Bharangi	Lamiaceae	Leaves, Roots	Inflammations, Anorexia, Flatulence, Common cold	[12], [14], [19]
39	*Vitex negundo* L.	Nirgundi	Lamiaceae	Leaves, Flower	Arthritis, Pesticide	[8], [10], [20]

40	*Careya arborea* Roxb.	Kumbha	Lecythidaceae	Root	Antiseptic	[10], [12], [19]
41	*Strychnosnux-vomica* L.	Kajara	Loganiaceae	Seeds, Bark	Digestive, Anti-diabetic	[6], [11], [21]
42	*Woodfordia fruticosa* (L.) Kurz	Dhayati	Lythraceae	Flower	Cytotoxic	[1], [8], [12]
43	*Grewia tiliifolia*Vahl.	Dhaman	Malvaceae	Stem bark	Pneumonia	[4], [8], [20]
44	*Helicteres isora* L.	Murudsheng	Malvaceae	Root, Pod	Anti-diabetic	[6], [11], [19]
45	*Thespesia populnea* (L.) Sol. ex Correa	Ranbhendi	Malvaceae	Root, Bark	Anti-inflammatory	[4], [10], [14]
46	*Urena lobata* L.	Caesar Gavat	Malvaceae	Leaves, Roots	Antioxidant, Antimicrobial	[1], [3], [8], [20]
47	*Memecylon umbellatum* Burm.f.	Anjan	Melastomataceae	Leaves	Anti-diabetic	[8], [11], [19]
48	*Azadirachta indica* A. Juss	Kadulimba	Meliaceae	Leaves, bark	Anti-diabetic, Blood purification, Skin diseases, Gums problem, Leprosy, Eye disorders, Bloody nose, Intestinal worms	[10], [12], [14]
49	*Tinospora cordifolia* (Willd.) Hook.f. & Thomson	Gulwel	Menispermaceae	Stem	Fever	[3], [[8], [20]
50	*Ficus racemosa* L.	Umber	Moraceae	Fruit, Latex	Food, Antiseptic	[11], [19], [21]
51	*Moringa oleifera* Lam.	Shevga	Moringaceae	Leaves	Reduces blood sugar level	[6], [8], [20]

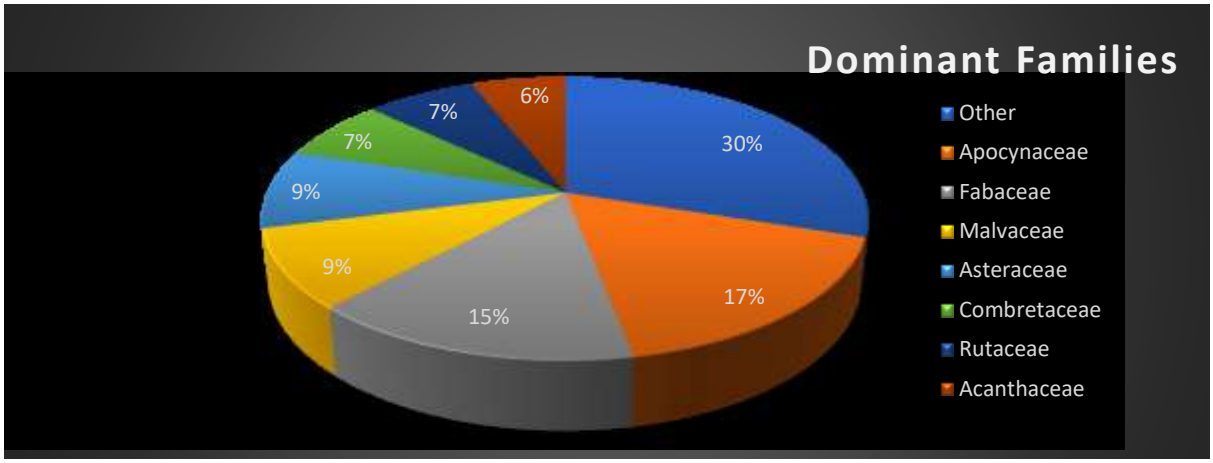
52	*Ensete superbum* (Roxb.) Cheesman	Rankeli	Musaceae	Flower, Seed	Urinary disorder and Kidney stone	[4], [14], [19]
53	*Nyctanthes arbor tristis* L.	Parijatak	Oleaceae	Bark	Used on cold & cough, Stops bleeding gums	[11], [14], [20]
54	*Argemone mexicana* L.	PiwalaDhotra	Papaveraceae	Leaves, Seeds, Roots, Flowers	Analgesic, antispasmodic	[3], [12], [19]
55	*Passiflora foetida* L.	Jungli Krishna-Kamal	Passifloraceae	Root	Antiseptic	[1], [8], [14]
56	*Phyllanthus urinaria* L.	BhuiAvala	Phyllanthaceae	All parts	Gonorrhea, Anti-diabetic, Flu	[4], [11], [20]
57	*Piper nigrum* L.	Kali Miri	Piperaceae	Fruit	Rheumatism, Appetizer	[10], [12], [19]
58	*Plumbago zeylanica* L.	Chitrak	Plubaginaceae	Leaves, Root, Bark	Rheumatism, Piles, Scabies, Menstrual Disorders, Obesity, Skin diseases, Arthritis	[10], [19], [21]
59	*Cymbopogon citratus* (DC.) Stapf	GavatiChaha	Poaceae	Leaves	Fever, Stomach cramps	[12], [14], [21]
60	*Embelia tsjeriamcottam*(Roem. &Schult.) A.DC.	Wawding	Primulaceae	Bark, Root	Piles, Sore throat, Dyspepsia	[1], [8], [11]
61	*Ixora coccinea* L.	Pendgul	Rubiaceae	Bark	Muscles Growth	[6], [20], [21]
62	*Aegle marmelos* (L.) Corrêa	Bel	Rutaceae	Leaves, Bark, Roots	Reduces cold & cough, Dysentery and	[4], [12], [20]

					Diabetes, Sun strokes, anti-cancer	
63	*Murraya koenigii* (L.) Spreng.	Kadipatta	Rutaceae	Leaves	Antioxidant	[1], [8], [11]
64	*Zanthoxylum rhetsa*(Roxb.) DC.	Tirphal	Rutaceae	Fruit	Stimulants, astringent	[4], [6], [19]
65	*Sapindus emarginatus*Vahl.	Ritha	Sapindaceae	Bark	Skin Diseases	[6], [20], [21]
66	*Manilkara kauki* (L.) Dubard	Bakuli	Sapotaceae	Bark, Seed, Flower, Fruit	Ulcers, Headache, Dental caries	[10], [12], [20]
67	*Smilax zeylanica* L.	Ghotwel	Smilacaceae	Leaves, Root	Antiseptic	[1], [8], [21]
68	*Solanum anguivi* Lam.	Chichardi	Solanaceae	Fruit	Digestive	[3], [11], [20]
69	*Lasiosiphon glaucus*Fresen.	Datpadi	Thymeleaceae	Leaves, Bark, Flower	Cancers, Sore throat, Wounds, Burns	[4], [10], [14]
70	*Leea indica* (Burm.f.) Merr.	Dinda	Vitaceae	Leaves	Antiseptic	[10], [8], [21]
71	*Curcuma pseudomontana* J. Graham	JungliHalad	Zingiberaceae	Rhizome	Ulcer, Antiseptic	[6],[19], [20]

3. RESULTS AND DISCUSSION

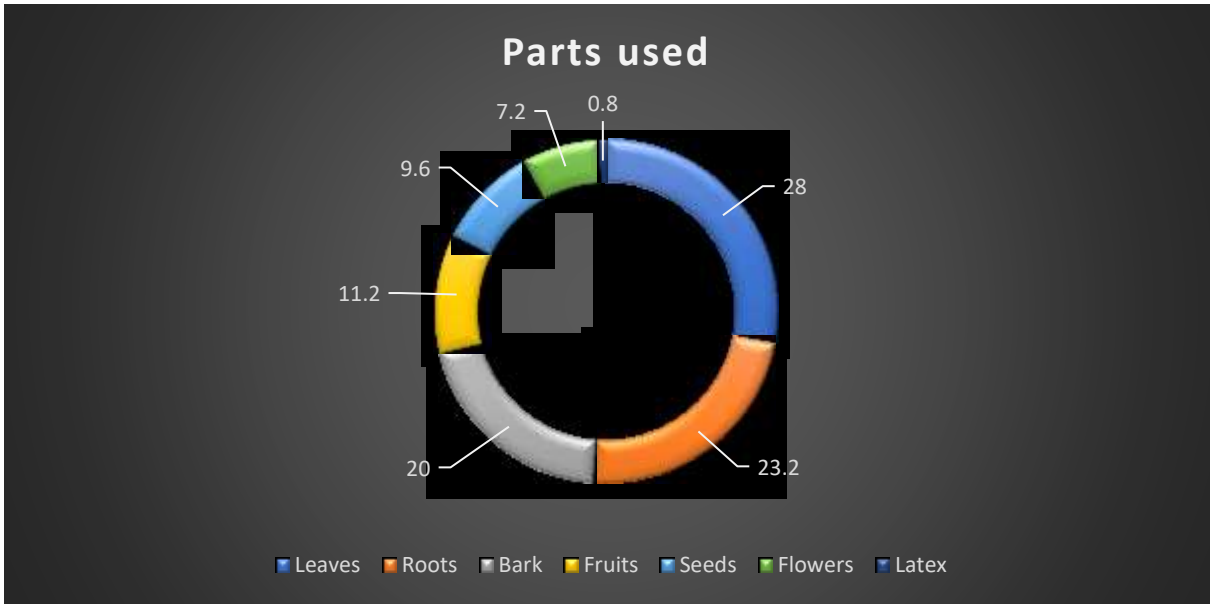
In the course of this survey, we recorded a total of 71 medicinal plants species belonging to 70 genus and 46 families. They were collected and identified (Table No.1) in the treatment of various ailments. For each species botanical name, family, local name, uses, parts used was discussed in detail (Table 1). The peak cited plant family was Apocynaceae (08 species) followed by Fabaceae (7 species), Malvaceae (04 species), Asteraceae (04 species), Acanthaceae (03 species), Rutaceae (03 species), Combretaceae (03 species).The study area is dominated by the trees like *Terminaliabellirica* (Gaertn.) Roxb, *Terminaliachebula*Retz. *DilleniapentagynaRoxb*. *Pongamiapinnata* (L.) Pierre, *Tamarindusindica* L., *Strychnosnux-vomica* L., *Grewiatiliifolia*Vahl, *Zanthoxylumrhetsa* (Roxb.). Kharepatan is situated near to the Sukh riverbank and the area is densely covered by trees and shrubs. The river basin is

dominated by species like *Ecliptaprostrata* (L.) L., *Cyperusrotundus* L., *Mimosapudica* L., *Thespesiapopulnea* (L.)Sol. ex Correa, *Phyllanthusurinaria* L.



4. CONCLUSION

It was seen that some plants were used in day to day life as food, spice and fruit. Variety of plant parts being used for their medicinal properties such as bark, fruits, leaves, rhizome, root, seed and stem; in some cases whole plants were used (Table 1). Leaf was the most widely used part for medicinal purpose. It was also seen that the availability of plants was decreasing at a startling rate. This observation also tells that habitat destruction, large scale cultivation of crop plants, over exploitation and environmental changes by human interference are the reason for decline the number of population of medicinal plants and their diversity too [16]. This area is undergoing rapid urbanization and fragmentation of forest patches [20]. Therefore, these plants need serious conservation measures because of their medicinal importance in conventional healthcare [23].



ACKNOWLEDGEMENT

Authors are thankful to Principal of Arts, Commerce & Science College Kharepatan, Tal.-Kankavali, Dist.- Sindhudurg and DattajiraoKadam Arts, Science & Commerce College Ichalkaranji, Dist.-Kolhapur for library and laboratory facilities. PDN is personally thankful to the SARTHI Institute, Pune for financial assistance. We also thankful to the Mr.SatyajitChavan (Engineer), Mr.VaseemSayyad, Mr.PrakashShinde, Mr.Gajanan Vhankali, Mr.Tanaji Godade, Mrs. Rutuja Karle and Mr. ShrikrishnaRanade for valuable guidance.

REFERENCES

1. Agharkar SP. Medicinal Plants of Bombay Presidency. Pbl," Scientific Publishers, Jodhpur. 1991;194-195.
2. Almeida SM. Flora of Sawantwadi. Scientific Publications, Jodhpur. 1990; Vol. 2.
3. Borate PP, Rao MS. An Ethnobotanical Study OfPinguli Village In Kudal Tehsil Of Sindhudurg Distrct Of Maharashtra. IJRBAT. 2018; 1:1-4.
4. Chopra RN, Nayar SL, Chopra IC. Glossary of Indian Medicinal Plants, New Delhi, Council of Scientific and Industrial Research. 1956.
5. Cooke T. The Flora of Presidency of Bombay. London reprinted ed., BSI Calcutta. 1901-1908; Vol. I and II.
6. Chopra IC, Verma BS. Supplement to the glossary of Indian medicinal plants, New Delhi, Council of Scientific and Industrial Research. 1968.
7. Daniel RJR. Taxonomic Uncertainties and Conservation Assessment of the Western Ghats, Current Science. 1997; 73(2):169-170.

8. Gogte VM. Ayurvedic Pharmacology and Therapeutic Uses of Medicinal Plants (Dravyagunavigyan), First ed. BharatiyaVidyaBhavan (SPARC), Mumbai Publications. 2000; 421-422.

9. Hooker JD. The Flora of British India. Reeves and company London. 1872:97.

10. Jain SK. Glimpses of Indian Ethnobotany, Oxford & IBH Publishing Co. New Delhi, Bombay, Calcutta. 1996.

11. Jain SK. Dictionary of Indian folk medicine and ethnobotany, New Delhi, Deep publications. 1991.

12. Kirtikar KR, Basu BD. Indian Medicinal Plants. Lalit Mohan Basu, Allahabad, Jayyed Press, New Delhi, India. 1987; 1-4:1313-1449.

13. Kothari MJ, Murthy JS. Flora of Raigad District, Maharashtra State. 1993; 403-404.

14. Natekar PD, Ugale NN, Indap SR, Parab MA, Pawar SS. Weed species diversity of Kharepatan village and Its nearby area. PLANTA. 2021;2:681–6.

15. Raza MA, Raza SS, Raza S, Raza TH, Raza SR, Raza MR, et al. An ethnobotanical study of medicinal plants in high mountainous region of Chail valley (District Swat-Pakistan). Journal of Ethnobiology and Ethnomedicine. 2014;10.

16. Scolozzi R, Geneletti D. Geneletti D. Environmental Impact Assessment Review. 2012; 36:9–22.

17. Sharma BD, Karthikeyan S, Singh NP. Flora of Maharashtra State, Monocotyledons, Botanical Survey of India, Flora of India. 1996;2.

18. Singh NP, Karthikeyan S. Flora of Maharashtra State. Botanical Survey of India. 2000;1.

19. Somkuwar SR, Chaudhary RR, Patil VN, Deokule SS. A Study of important medicinal plants of Savantwadi region, Western Ghats, (MS). Int J Cur Res. 2012; 4:154–9.

20. Somkuwar SR, Chaudhary RR, Chaturvedi AA. Ethnofloristic Diversity InDodamarg, Region (Ms) Central Western Ghats, India. Life Sciences Leaflets. 2013;55–71.

21. Vartak VD. Contribution to Indian Ethnobotany, 3rd Ed. Scientific Publishers, Jodhpur. 1997.

22. Yadav SR. Sardesai M M. Flora of Kolhapur District. 2002.

23. Yadav SR. Rare, Endangered and Threatened Plant species of Western Ghats and their conservation, Journal of Science Information/special issue-3. 2012; 01-04.

24. Tropicos [Internet]. Tropicos.org. [cited 2022 Feb 2]. Available from: http://www.tropicos.org

25. Home — the plant list [Internet]. Theplantlist.org. [cited 2022 Feb 2]. Available from: http://www.theplantlist.org

26. Worldweatheronline.com. [cited 2022 Feb 2]. Available from: http://www.worldweatheronline.com

Shivaji University, Kolhapur
Corresponding author nbmane123@gmail.com, tkhandagale98@gmail.com,

Abstract

The *Crozophora plicata* (Vahl) belongs to family euphorbiaceae from drought prone area Wathar station in Satara District were investigated for occurrence of arbuscular mycorrhizal fungal association. Collect the test plant and screened for qualitative determination and occurrence of (AM) fungi. The result were reported from rhizopsphere soil of test plant are two genera, Aculospora and Glomus. Glomus ((7) are maximum than Acaulospra (1). Qualitative analysis was carried out from fruit and leaf of selected plant. Carbohydrate, Phenol, Saponin, Flavonoid, were found more while Alkaloid, Tannin and Glycosides were recorded less.

Keywords: Arbuscular Mycorrhiza, *Crozophora plicata, Glomus, Acaulaspora,*

Introduction:

Crozophora *plicata* belongs to family euphorbiaceae from drought prone area Wathar station in Satara District were investigated for occurrence of arbuscular mycorrhizal fungal association. Soil microorganism can influence the soil structure and plays an important role for soil fertility. Arbuscular Mycorrhizal fungi are associated with roots with approximately 80% in terrestrial plants (Smith and Read 1997). Benefit from AMF plants consist higher nutrient uptake, great tolerance to some pathogens (Koide and Moss, 2004). Phytomedicine are used for the treatment of diseases. (Iwu M 1993). World health organization (WHO) also supported about Phytomedicines are safe, less toxic obtained easily from natural resourses. Plants contain some organic compounds which supply physiological actions on the human body and these bioactive constituents are alkaloids, carbohydrates, terpenoides and flavonoids Edoga, H. (2005), Mann, (1978). Phytochemicals are useful for to cure number of diseases. (Jawanmardi, Stushnoff and Locke et.al. 2003).

Vitamins are organic substances necessary for metabolism. In diet of Human being not contain required amount of Vitamins. These vitamins are present in fruits and vegetables are received from chemical constituents. Hussian et. al., 2006). Some plants have medicinal properties which are useful for physiological action on the human body and these bioactive substances are alkaloides, carbohydrates, phenol saponin etc. Phytochemical compounds are widely used in the human therapy, veterinary, agriculture and scientific research. Present

communication helps to assess the status of Arbuscular Mycorrhizal fungi and role of phytoconstituent to human being and agricultural purposes.

Materials and Methods:

Collection and identification of Plant material:

Crozophora *plicata*plant was collected from agricultural site of drought prone area of in Satara District. The test plant is identified in recognized research lab of Yashvantrao Chavan Institute of Science, Satara affiliated to Shivaji University, Kolhapur with the help of Flora of Cook. Herbarium of plant maintained in research lab for further study. The plant material was cut off and the plant was washed thoroughly under tap water to free from debris. The leaves and fruit of fresh plant material chopped in small segments and dried in shade, after drying, the plant material were ground well using mechanical blender into fine powder and stored in air tight container with proper labeling for future investigation.

Arbuscular Mycorrhizal Fungi isolation and its identification:

The soil samples were collected in sterile zip lock polythene bags. AMF were isolated from rhizosphere soil by wet sieving and decanting method of Gerdemann and Niccolson (1963). Intact AM spore were examined under binocular stereo microscope and identified spores with size shape and wall layers and hyphal attachments using the species descriptions given by INVAM and manual of Schenck and Peerez 1990.

Qualitative analysis:

Preparation of plant extract:

The dried leaf and fruit finely powdered material was taken in a beaker and 200ml of distilled water was added. The mixture was heated on a hot plate with continuous stirring at 30º-40ºC for 20 minutes. Then the water extract was filtered through filter paper and the filtrate was used for the phytochemical analysis. The extract was kept in cool condition for further analysis.

Qualitative phytochemical analysis:

The extract was tested for the presence of bioactive compounds by using standard methods of Horborne, Parekh and Sofowra.

Test for Alkaloids Wagner's test:

A fraction of extract was treated with Wagner's test reagent 1.27 g of iodine and 2 g of potassium iodide in 100 ml of water and observed for the formation of reddish brown colour precipitate.

Test for Flavonoids NaOH test:

A small amount of extract was treated with aqueous NaOH and HCl, observed for the formation of yellow orange colour.

H2SO4 test:

A fraction of extract was treated with concentrated H_2SO_4 and observed for the formation of orange colour.

Lead acetate test:

A small amount of extract was treated with lead acetate and observed for the formation of white precipitate.

Test for Saponins:

Foam test: A small amount of extract was shaken with water and observed for the formation of persistent foam.

Test for Glycosides:

Legals test:

Chloroform (3ml) and ammonia solution (10%) was added to 2ml plant extract. Formation of pink color indicated the presence of glycosides.

Test for Phenols:

Ferric chloride test: The fraction of extract was treated with 5 % ferric chloride and observed for formation of deep blue or black colour.

Liebermann's test:

The extract was heated with sodium nitrite, added H2SO4 solution diluted with water and excess of dilute NaOH was added and observed for the formation of deep red or green or blue colour.

Test for Anthraquinones

Borntrager's test:

About 50 mg of powdered extract was heated with 10% ferric chloride solution and 1ml concentrated HCl. The extract was cooled, filtered and the filtrate was shaken with diethyl

ether. The ether extract was further extracted with strong ammonia; pink or deep red colourations of aqueous layer indicate the presence of anthraquinone.

Test of Carbohydrates

Fehling's test:

Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and gently boiled. A brick red precipitate appeared at the bottom of the test tubeIndicated the presence of reducing sugars.

Benedict's test:

Crude extract when mixed with 2ml of Benedict's reagent and boiled, a reddish brown precipitate formed which indicated the presence of the carbohydrates.

Result and discussion:

The isolation and identification of Arbuscular Mycorrhizal Fungi was estimated from *Crozophora plicata.* The result were exhibited from rhizopsphere soil of test plant a total 8 AMF species belonging to two genera, Aculospora and Glomus. *Acaulosporalacuna*, *Glomus diaphanum, Glomus dimorphicum, Glomus fasciculatum, Glomus fistulosum, Glomus glabiferum, Glomus macrocarpum and Glomus macrolosum* were recorded from selected plant. Genera Glomus was recorded dominant than Aculospora.. The phytochemical analysis of *Crozophora plicata* were summarized in Table no 1. The result exhibited the presence of Carbohydrate, Alkaloide, Phenol, Tannin Saponin, Flavonoides and Glycosides etc. The Fruit indicates presence of Phenol, Saponin, Flavonoides. Tannins and Glycosides were reported very less in leaf of *Crozophora plicata* Table no 2. AMF species belonging to Aculospra and Glomus species were isolated from *Crozophora plicata.* Camprubi and Calvet (1996) attributed Glomus was most common and dominant found in Citrus soils. Klironomous and Hart (2002). AMF species individually compete for resourses through combination of stategis exhibiting in the maintainance of a diverse AMF Community. Alkaloides was benefitialfor the treatements of tumours and diarrhea. (G Visweswari 2013). Saponin were extracted from plants reports biological and pharmacological activities such as anti-inflammatory, wound healing . Antimicrobial and antiviral Rahman (2010).

Table no.1 Isolation of Arbuscular Mycorrhizal Fungi from *Crozophora plicata*

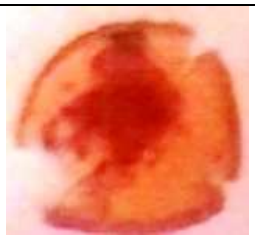
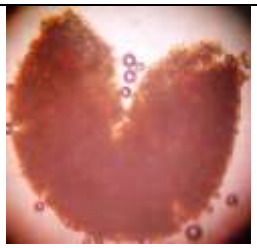
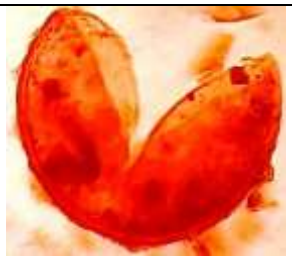
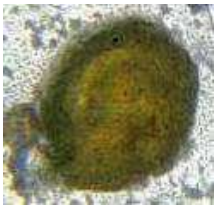
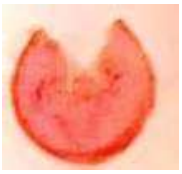
 *Aculospora lacunose* (Morten)	 *Glomus fistulosum* (Jacobson)	 *Glomus maculosum* (Miller and Walker)
 *Glomus macrocarpum* (Tulasne and Tulasne)	 *Glomus fasciculatum* (Thaxter)	 *Glomus dimorphicum* (Tewari)

Table no.2. Qualitative estimation of the crude extracts of fruit and leaves of *Crozophora plicata*

Phytochemical constituent	Aqueous	Ethyl acetate
Carbohydrate	++	++
Alkaloide	--	++
Phenol	++	++
Tannin	--	-
Saponin	++	
Flavonoides	++	++
Glycosides	--	--

++ indicates –Presence of Compounds, -- indicates absence of compounds

Conclusion:

Plants are rich source of phytochemicals are widely used in traditional medicine to cure various ailments. The different extract of plants part is contained Carbohydrate, Alkaloide, Phenol, Tannin Saponin, Flavonoides and Glycosides are used in high proportion as an aphrodisiac, neuroprotective, liver, tonic and astringent

Acknowledgement:

The authurs acknowledge the profound gratitude to the Principal Dr B. T. Jadhav, Head Department of Botany Yashvantrao Chavan Institute of Science, Satara for providing the laboratory facility

References:

Rahman Onike (2010). Phytochemical Screening tests and Medicinal Values of plants Active Properties.

Camprubí, A. and Calvet, C. (1996). Isolation and screening of mycorrhizal fungi from *Citrus* nurseries and orchards and inoculation studies. Hortscience 31(3): 366– 369.

Klironomos, J.N. and Hart, M. M.(2002). Colonization of roots by arbuscularmycorrhizal fungi using different sources of inoculum. Mycorrhiza 12: 181–184.

Vasu, K., Goud, J.V., Suryam, A., Singara, Chary, M.A. 2009. Biomolecular and phytochemical analyses of three aquatic angiosperms. Afr. J. Microbiol. Res., 3(8):418-421.

Smith, S. E. and Read, D. J. (1997). Mycorrhizal symbiosis. 2nd edn., Academic Press, San Diego, CA, USA.

Koide, R.T. and Mosse, B. (2004). A history of research on arbuscular mycorrhiza. Mycorrhiza 14: 145–163.

Javanmardi J, Stushnoff C, Locke E, Vivanco JM (2003). Antioxidant activity and total phenolic content of Iranian Ocimum accessions. J. Food Chem., 83: 547-550.

Hussian I, Saleem M, Iqbal Y, Khalil S. J. (2006). Comparison of Vitamin C Contents in Commercial Tea Brands and Fresh Tea Leaves. Jour. Chem. Soc. Pak, 28(5): 421-425.

Edoga, H.O., Okwu, D.E., Mbaebie, B.O. 2005. Phytochemicals constituents of some Nigerian medicinal plants. Afr. J. Biotechnol., 4(7): 685-688.

Mann, J.1978. Secondary Metabolism. Oxford University press,London, pp. 154.

Estimation of Phytoconstituents, Soil characterization and Isolation of Arbuscular Mycorrhizal fungi from *Curcuma longa* L.

N. B. Mane, R. S. Nikam

Department of Botany, YashavantraoChavan Institute of Science, Satara (Autonomous)

Shivaji University, Kolhapur
Corresponding author:nbmane123@gmail.com, nikamrajani1999@gmail.com

Abstract:

Present study was undertaken for the study of phytochemical analysis, soil characterization and isolation of ArbuscularMycorrhizal fungi in the Rhizosphere of *Curcuma longa* L. Collect the soil samples from Ambheri from Satara District of Maharashtra India. The soil samples of *Curcuma longa* screened for its physiochemical properties. The root powder of test plant showed maximum amount of Carbohydrate, Phenol, Saponin, Flavonoides while Glycosides, Tannin, alkaloids protein were recorded minimum. Isolation of AM fungi attributed with Glomus and Acaulospora.

Keywords: ArbuscularMycorrhiza, *Curcuma longa, Glomus, Acaulaspora,*Alkaloide

Introduction:

Curcuma longa L.is a flowering plant; it belongs to family Zingiberaceae commonly called as Turmeric. Rhizome of this medicinal plant used for safe and active drug for the treatment of different chronic diseases like diabetes. The turmeric is used as a traditional medicine and remedy for different diseases including a coughs, diabetes, dermatological conditions, respiratory problems, cardiovascular and hepatobiliary diseases, arthritis, irritable bowel disease peptic ulcers, psoriasis, and atherosclerosis.Angela Laguipo, (2022). It has universally as one of the most powerful herb for fighting various diseases. It decreases brain problems and heart problem. Curcumin is phenolic constituent and it is hydrophobic in nature. (Nelson et. Al, 2017). It is also used as remedy on different types of wounds and plays a role in delaying the wound healing process resulting wound infections (Bowler, 2001). Different plants secrete thousands of phytochemicals showing inhibitory effects against many types of micro-organism (Cowan, 1999). According to guidelines from World health organization

medicinal plants have best resource of medicine (Aggarwal, 2007). Herbal products are benefitial for treating a wide range of infections and diseases (Chattopadhyay et.al, 2004).

Material and Method:

Assessment of soil:

Analysis of soil was carried with the method given by Tan, (1996). Soil pH was determined by potentiometric method.

Collection and identification of Plant material:

Curcuma longa L. Plant was collected from Ambherivillage inSatara District of Maharashtra, India. The test plant is identified in recognized research lab of YashvantraoChavan Institute of Science, Satara affiliated to Shivaji University, Kolhapur with the help of Flora of Cook. Herbarium of plant maintained in research lab for further study. The plant material was washed thoroughly under tap water to free from debris. The root of plant material chopped in small segments and dried in shade, after drying, the plant material were ground well into fine powder and stored in air tight container with proper labeling for future investigation.

ArbuscularMycorrhizal Fungi isolation and its identification:

The soil samples of test plant were collected in sterile zip lock polythene bags. AMF were isolated from rhizosphere soil by wet sieving and decanting method of Gerdemann and Niccolson, (1963).Intact AM spore were examined under binocular stereo microscope and identified spores with size shape and wall layers and hyphal attachments using the species descriptions given by INVAM and manual of Schenck and Peerez, 1990. Blaszkowski, (1993).

Table no. 1. Physicochemical Properties of Soil:

Sr. No	Soil analysis	Study site	Limit	Suggestion
1.	PH	5.8	8-8.4	High
2.	EC	2.75	4.68	High
3.	Sodium (kg/ha)	16.49	16.01-16.04	High

4.	Calcium kg/ha	23.09	21.-22	Low
5.	Potassium kg/ha	16.36	16.35	Medium
6.	Temperature(°C).	28c	26.9	Medium

Qualitative analysis:

Preparation of plant extract:

The dried root of test plant finely powdered material was taken in a beaker and 200ml of distilled water was added. The mixture was heated on a hot plate with continuous stirring at 30º-40ºC for 20 minutes. Then the extract was filtered through filter paper and the filtrate was used for the phytochemical analysis. The extract was kept in cool condition for further analysis.

Qualitative phytochemical analysis:

The extract was tested for the presence of bioactive compounds by using standard methods of Horborne, Parekh and Sofowra.

Test for Alkaloids (Wagner's test):

A fraction of extract was treated with Wagner's test reagent 1.27 g of iodine and 2 g of potassium iodide in 100 ml of water and observed for the formation of reddish brown colour precipitate.

Test for Flavonoids NaOH test:

A small amount of extract was treated with aqueous NaOH and HCl, observed for the formation of yellow orange colour.

H2SO4 test:

A fraction of extract was treated with concentrated H_2SO_4 and observed for the formation of orange colour.

Lead acetate test:

A small amount of extract was treated with lead acetate and observed for the formation of white precipitate.

Test for Saponins (Foam test):

A small amount of extract was shaken with water and observed for the formation of persistent foam.

Test for Glycosides: (Legals test)

Chloroform (3ml) and ammonia solution (10%) was added to 2ml plant extract. Formation of pink color indicated the presence of glycosides.

Test for Phenols (Ferric chloride test):

The fraction of extract was treated with 5 % ferric chloride and observed for formation of deep blue or black colour.

Liebermann's test:

The extract was heated with sodium nitrite, added H2SO4 solution diluted with water and excess of dilute NaOH was added and observed for the formation of deep red or green or blue colour.

Test of Carbohydrates (Fehling's test):

Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and gently boiled. A brick red precipitate appeared at the bottom of the test tubeIndicated the presence of reducing sugars.

Benedict's test:

Crude extract when mixed with 2ml of Benedict's reagent and boiled, a reddish brown precipitate formed which indicated the presence of the carbohydrates.

Result and discussion:

The phytochemical analysis of *Curcuma longa* L. were summarized in Table no 2. The root powder of test plant showed maximum amount Carbohydrate, Phenol, Saponin, Flavonoides while Glycosides, Tannin, alkaloids protein were recorded minimum. Tannins, Protein and Glycosides were reported very less in root of *Curcuma longa*.

The isolation and identification of ArbuscularMycorrhizal Fungi was estimated from *Curcuma longa.* The result were exhibited from rhizopsphere soil of test plant a total 10 AMF species belonging to two genera, Aculospora and Glomus. *Acaulospora sporocarpa* (S.M.Berch),*A. appendicola*(Blume), *A. laevis*(Wiegand), *Glomus etunicatum* (Becker and Gerd), *G. arborense* (Mc. Gee), *G. fasciculatum*(Tul and Tul), *G. fistulosum* (Skou and Jacobson), *G. globiferum* (Dalpe and Declerk),*G. macrocarpum*(Tul and Tul) *and G. hoi* (Berck and Trappe) were recorded from selected plant. Genera Glomus was recorded dominant than Aculospora.

AMF species belonging to GeneraAcaulospora and Glomus were isolated from *Curcuma longa.* Camprubi and Calvet, (1996) attributed Glomus was most common and

dominant found in Citrus soils. Klironomous and Hart, (2002). AMF species individually compete for resources through combination of strategies exhibiting in the maintenance of a diverse AMF Community. Alkaloides was beneficialfor the treatments of tumors and diarrhea. (G.Visweswari, 2013). Brundrett et al., (1991) highlighted Variation of AM fungi in various localities could be due to the change in the habitat, environmental factor, soil fertility and acclimatization of a particular location. Similar results are highlighted by Muthukumar and Udaiyan, (2000), (2006) identified the Genera Glomus and Aculospora in grass, Bamboo. Kong, (2017) has attributed the Glomus and Acaulospora were maximum in *SasaKurilensis* in Japan. Das and Kayang, (2010) in *Phyllostachysmanti*. Jiyang et al, (2013) confirmed symbiotic association with inoculation of Glomus in *Bambusapervariabilis*due to absorption of Phoshorus.

Table no. 2. Qualitative estimation of the crude extracts of *Curcuma longa*.

Phytochemical constituent	**Aqueous**	**Ethyl acetate**
Carbohydrate	++	++
Alkaloide	--	++
Phenol	++	++
Tannin	--	-
Saponin	++	++
Flavonoides	++	++
Glycosides	--	--

++ indicates –Presence of Compounds, -- indicates absence of compounds

Acknowledgement:

The authors are Thankful to honorable Director Dr. B. T. Jadhav of Rayat Shikshan Sanstha's Yashvantrao Chavan Institute of Science, Satara (Autonomous) and Mr. H. L. Shinde Head, Department of Botany, for their constant support and facilities provided.

References:

Aggarwal BB, Sundarma C, Malini N & Ichikawa H. Adv. Exp. Med. Biol. 595:1-75 (2007).

Angela Betsaida and Laguipo, (2022). News life medical life Sciences.

Blaszkoski J. (1993). Comparative studies on the occurrence of arbuscular fungi and mycorrhizae (Glomales) in cultivated and uncultivated soils of Poland. Acta Mycol 28:93-140.

Chattopadhyay I, Biswas U, Badopadhyay and Banerjee R.K. Curr.Sci 87:44-53 (2004).

Cowan M. M: Plant products of antimicrobial agents. Clinical Microbial Review 1999; 12(1): 564-582.

Das P, Kayang H., (2010) Arbuscularmycorrhizal fungi and dark septateendophyte colonization in bamboo from Northeast India. Front Agric China 4:375–382.

Gerdemann J. W., Nicolson T. H., (1963) Spores of mycorrhizalEndogone species extracted from soil by wet sieving and decanting. Trans Br MycolSoc 46:235–244

INVAM(2018) International culture collection of (vesicular) arbuscularmycorrhizal fungi. West Virginia University, Morgantown

James D. Souza, Bernald Felinou Rodrigues, (2013). Biodivesity of Arbuscular Mycorrhizal (AM) Fungi in Mangroves of Goa in West India. Journal of Forestry and Research 24, 513-525.

Jiang W, Gou G, Ding Y., (2013) Influences of arbuscularmycorrhizal fungi on growth and mineral element absorption of chenglu hybrid bamboo seedlings. Pak J Bot 45:303–310.

Kong B (2017) Study of the microbial community structure in the rhizosphere of understory dwarf bamboo (Sasakurilensis) in a Betulaermanii forest, northern Japan. Doctoral Thesis, Hokkaido University, Japan.

M. K. Nelson, J. L. Dahlin, J. Bisson, J. Graham, G.F. Pauli, and M. A. Walters, 2017, Journal of Medicinal Chemistry, Vol. 60, pp. 1620.

Muthukumar T, Udaiyan K (2000) Arbuscularmycorrhizas of plants growing in the Western Ghats region, southern India. Mycorrhiza 9:297–313

Muthukumar T, Udaiyan K (2006) Growth of nursery-grown bamboo inoculated with arbuscularmycorrhizal fungi and plant growth promoting rhizobacteria in two tropical soil typeswith and without fertilizer application. NewFor 31:469–485.

P. G. Bowler, B. I. Duerden, and D. G. Armstrong, 2001, Clinical Microbiological Review, Vol.14, pp.244.

Diversity of marginal plants of some water bodies in Gadhinglaj Tahsil and their ecological significance.

Sawant R. S*., Patil A.P., Patel J. S. and Patil S.M.

Department of Botany,
Dr. Ghali College Gadhinglaj, 416502 (MS).
* Corresponding Author (rss.botany@gmail.com)

Abstract:

Gadhinglaj Tahsil (Kolhapur District) located at Northern Western region of Maharashtra. Forty four manmade water-bodies are recorded from 90 villages present in this Tahsil. Out of 44 water bodies, we studied 9 water-bodies. The plants present at peripheries of these water-bodies are enlisted here. These marginal plants are with all kinds of habits including herbs, shrubs, trees, climberand grasses. Marginal plants include plants of Reed-swamp stages and Sedge Marsh or Meadow stage. Majority of the vegetation studied was found to be naturally occurring. About 89 Angiosperms and one ferns were recorded around the water bodies. The present work reflects not only ecological role of marginal vegetation (as producer and habitat providers) but role in succession of the flora and fauna around the water-bodies.

Key words: Gadhinglaj, Water bodies, Angiosperms, Diversity, ecology

Introduction:

Since an ancient time, wetlands provide a better settlement of human and his activity, ultimately leads to modification of these wetlands. . These water reservoirs serves to peoples for domestic uses like cloth washing, animal washing, bathing, animal water drinking, agricultural irrigation etc. The water is also used for human consumption from some of the water bodies. Aquatic macrophytes are growing in or around water. The studies on aquatic macrophytes are important in order to understand the functioning of aquatic ecosystem. Macrophytes comprises and important component of aquatic life, especially in nutrient rich wyland ecosystem (Seabloom, 2003). contributing significantly towards primary production which influencing various hydrochemical processes. They also serve as complex habitat offers support , protection and food to aquatic fauna (Raspov et al. 2002; tessier et al. 2004; yousuf and Firdous, 2001 and Crouder and Cooper 1982).

Aquatic macrophytes are highly important as substrate for periphyton and epiphytic food, refuges from predetain, heterogeneous substrates for co- existence. Changes in water level and Depth affects on the distribution of species in a plant community in a habitat (Hudon et al., 2004). It also affects on substrate composition and interaction with other plant and animal communities (Leslie et al, 1988). Gadhinglaj village is located in Gadhinglaj Tahsil of Kolhapur district in Maharashtra, India. Gadhinglaj is at 16.23°N 74.35°E. It has an average

elevation of 623 meters (2043 feet). t has an average weather of clear sky and temperature of around 15 °C in winter and 24 °C in summer and has more rainfall than average in Kolhapur District. Sawant et al. (2014) has been made to reveal the status of fresh water reservoirs from Gadhinglaj Tahsil of Kolhapur District, Maharashtra, India by using Global Positioning System (GPS) with reference to survey and mapping.

Material and methods:

Study Area: The major water reservoirs of Gadhinglaj (16o 13' 26" N and l74o 26' 9" E) Tahsil of Kolhapur District from Maharashtra, Tahsil occupying 48094 ha of area. 9 water bodies were selected for marginal floristic study. Frequent visits were made to locate the water bodies of Gadhinglaj Tahsil. After preliminary survey, water bodies were identified and classified and accordingly, ecological observations were noted for individual water bodies and mapping of major water bodies was made (**Table 1 & fig. 1**).

Plant Identification: Frequent visits were made to enumerate floristic diversity around the all the water bodies under studied. Plants collected, characters ware noted in field and laboratory and pressed well to prepare herbarium. Plant identification was carried out with help of appropriate floras and other work including *The Flora of the Presidency of Bombay* Vol. 1, 2., *Flora of Kolhapur District*, *Flora Of Maharashtra state* vol. 1 and 2. (**Table 2.)**

Table 1. List of water bodies with some ecological and geographical parameters.

Sr. no.	Locality name with abbreviations written in bracket.	GPS	Rainfall (in mm)	Lake area (in Hectore)	Fig. no. In table
1	Karmabali **(Kr)**	16°11'51.4"N 74°17'48.1"E	1100	27.34	1A
2	Shendri **(Sd)**	16°16'14.2"N 74°21'01.6"E	1000	41.09	1B
3	Yenechanvandi **(Yn)**	16°10'29.1"N 74°25'51.5"E	1050	29.15	1C
4	Terani **(Tr)**	16°07'33.9"N 74°28'37.1"E	910	85.24	1D
5	Narevadi **(Nr)**	16°08'52.8"N 74°25'12.2"E	940	32	1E
6	Vairagvadi **(Vg)**	16°09'33.6"N 74°21'47.0"E	1000	29.87	1F
7	Kumari **(Ku)**	15°59'44.1"N 74°18'21.3"E	1250	38.43	1G
8	Kadgav **(Kd)**	16°15'13.8"N 74°18'02.5"E	980	3.25	1H

9	Mahagav **(Mh)**	16°08'38.5"N 74°20'05.8"E	1000	4.65	1I

Fig.1. Aerial views of Water bodies; 1A. Karambali, 1B. Shendri, 1C. Yenechavandi, 1 Terani, 1E. Narewadi, 1F. Vairagvadi, 1G. Kumari, 1H. Kadgav, 1I. Mahagav.

Result:

Table no. 2. Checklist of marginal plants species around all water bodies under study.

Sr. No.	Botanical Name	Kr	Sd	Yn	Tr	Nr	Vg	Ku	Kd	Mh
1	*Abutilon indicum* (L.) Sweet	+	+	-	-	-	-	+	+	+
2	*Acacia longifolia* (Andrews) Willd.	+	+	+	+	+	+	+	+	+
3	*Acacia nilotica* (L.) Delile	+	+	+	+	+	+	+	+	+
4	*Achyranthes aspera* L.	+	+	-	+	+	-	-	-	-
5	*Acilepis dendigulensis* (DC.) H.Rob.	+	+	-	-	-	-	+	+	+
6	*Aerva javanica* (Burm.f.) Juss. ex Schult.	-	-	+	-	-	-	-	-	-
7	*Ageratum conyzoides* (L.) L.	+	+	+	+	+	+	+	+	+
8	*Alternanthera sessilis* (L.) R.Br. ex DC.	+	+	+	+	+	+	+	+	+
9	*Alysicarpus vaginalis* (L.) DC.	+	+	-	+	+	+	-	-	-
10	*Amaranthus viridis* L.	+	+	+	+	+	+	+	+	+
11	*Apluda mutica* L.	+	+	+	+	+	+	+	+	+
12	*Argemone mexicana* L.	+	+	+	+	+	+	+	+	+
13	*Aristida funiculata* Trin. & Rupr.	+	-	-	+	+	-	+	-	+
14	*Arthraxon sp.*	+	+	-	-	-	-	+	+	+
15	*Arundinella sp.*	+	-	+	+	+	+	+	+	-
16	*Asparagus racemosus* Willd.	+	-	+	+	+	+	+	+	-
17	*Azadirachta indica* A.Juss.	+	+	+	+	+	+	+	+	+
18	*Boerhavia diffusa* L.	+	+	+	+	+	+	+	+	+
19	*Butea monosperma* (Lam.) Taub.	+	+	+	+	+	+	+	+	-
20	*Caesalpinia decapetala* (Roth) Alston	+	+	+	-	+	+	-	-	+
21	*Calotropis gigantea* (L.) Dryand.	+	+	-	-	-	-	-	+	+
22	*Calotropis procera* (Aiton) Dryand.									
23	*Carissa carandas* L.	+	+	-	+	-	+	+	-	-
24	*Cassia surattensis* Burm.f.									
25	*Celosia argentea* L.	+	+	+	+	+	+	+	+	+
26	*Chloris virgata* Sw.	+	+	+	+	+	+	+	+	+
27	*Chrozophora plicata* (Vahl) A.Juss. ex Spreng.	+	+	+	+	+	+	+	+	+
28	*Coix lacryma-jobi* L.	+	+	+	+	+	+	+	+	+

29	*Coldenia procumbens* L.	+	+	+	+	+	+	+	+	+
30	*Commelina benghalensis* L.	+	+	+	+	+	+	+	+	+
31	*Commelina diffusa* Burm.f.									
32	*Cynodon dactylon* (L.) Pers.	+	+	+	+	+	+	+	+	+
33	*Cyperus rotundus* L.									
34	*Cyperus sp.1*	+	+	+	+	+	+	+	+	+
35	*Dactyloctenium aegyptium* (L.)Willd	+	+	+	+	+	+	+	+	+
36	*Dendrocalamus strictus* (Roxb.) Nees	+	+	+	+	+	+	+	+	+
37	*Dichanthium annulatum* (Forssk.) Stapf	+	+	+	+	+	+	+	+	+
38	*Digera muricata* (L.) Mart.	-	+	+	+	+	+	-	-	-
39	*Digitaria stricta* Roth.	+	+	+	+	+	+	+	+	+
40	*Dimeria sp.*	+	+	+	+	+	+	+	+	+
41	*Dinebra retroflexa* (Vahl) Panz.	+	+	+	+	+	+	+	+	+
42	*Eclipta prostrata* (L.) L.	+	+	+	+	+	+	+	+	+
43	*Eleocharis sp.1*	+	+	+	+	+	+	+	+	+
44	*Eleusine indica* (L.) Gaertn.	+	+	+	+	+	+	+	+	+
45	*Eragrostis sp.*	+	+	+	+	+	+	+	+	+
46	*Eriocaulon sp.*	+	+	+	+	+	+	+	+	+
47	*Eucalyptus globulus*	+	+	+	+	+	+	+	+	+
48	*Euphorbia laciniata* Panigrahi.	+	+	+	+	+	+	+	+	+
49	*Euphorbia heterophylla* L.	+	+	+	+	+	+	+	+	+
50	*Euphorbia hirta* L.	+	+	+	+	+	+	+	+	+
51	*Ficus benghalensis* L.	+	+	-	-	+	+	+	-	-
52	*Ficus religiosa* L.	+	+	+	+	+	+	-	+	+
53	*Heliotropium sp.*	+	+	+	+	+	+	+	+	+
54	*Hygrophila auriculata* (Schumach.) Heine	+	+	+	+	+	+	+	+	+
55	*Impatiens balsamina* L.	+	+	+	+	+	+	+	+	+
56	*Indigofera sp.*	+	+	+	+	+	+	+	+	+
57	*Ipomoea cairica (*L.) Sweet	-	-	-	-	-	+	-	-	-
58	*Ipomoea carnea* Jacq.	+	+	+	+	+	+	+	+	-
59	*Jatropha curcus*	+	+	+	+	+	+	-	+	+
60	*Leucas aspera* (Willd.) Link	+	+	+	+	+	+	+	+	+
61	*Leucas stelligera* Wall. ex Benth.	+	+	+	-	-	-	+	+	+
62	*Ludwigia octovalvis* (Jacq.) P.H.Raven	+	+	+	+	+	+	+	+	+
63	*Lanatana camera* L.	+	+	+	+	+	+	+	+	+
64	*Mangifera indica* L.	+	-	-	-	+	-	-	+	+
65	***Marsilea minuta* L.**	+	+	+	+	+	+	+	+	+
66	*Mimosa pudica* L.	+	+	+	+	+	+	+	+	+
67	*Oxalis corniculata* L.	+	+	+	+	+	+	+	+	+

68	*Parthenium hysterophorus* L.	+	+	+	+	+	+	+	+	+
69	*Phyllanthus fraternus* G.L.Webster	+	+	-	-	-	-	+	+	+
70	*Phylla nodiflora* L.	+	+	+	+	+	+	+	+	+
71	*Pithocelobium dulce*	+	+	+	+	+	+	+	+	+
72	*Pleocaulus sessilis* (Nees) Bremek.	-	-	-	-	-	-	+	-	-
73	*Pogostemon deccanensis* (Panigrahi) Press	+	-	-	+	-	-	-	-	-
74	*Polygonum plebeium* R.Br.	+	+	+	+	+	+	+	+	+
75	*Pongamia pinnata* (L.) Pierre	+	+	+	+	+	+	+	+	+
76	*Prosopis cinerara*									
77	*Rungia repens* (L.) Nees	+	+	-	-	-	-	+	-	-
78	*Santalum album L.*	-	-	-	-	-	+	-	+	+
79	*Senna uniflora* (Mill.) H.S.Irwin & Barneby	+	+	+	+	+	+	+	+	+
80	*Sida rhombifolia* L.	+	+	-	-	+	-	-	-	-
81	*Sonchus oleraceus f. hydrophilus (*Boulos) J.Kost.	+	+	+	+	+	+	+	+	+
82	*Spilanthes acmella* (L.) L.	+	+	+	+	+	+	+	+	+
83	*Saccharum spontaneum* L.	+	+	+	-	-	-	-	-	-
84	*Tamarindus indica* L.	+	+	-	-	+	-	-	-	+
85	*Tephrosia sp.*	-	-	-	-	+	+	-	-	-
86	*Thevetia neriifolia* Juss. ex Steud.	+	+	+	+	+	+	+	+	+
87	*Tridax procumbens* (L.) L.	+	+	+	+	+	+	+	+	+
88	*Typha angustifolia* L.	+	+	+	+	+	+	+	+	+
89	*Vitex negundo* L.	+	+	+	+	+	+	+	+	+
90	*Zornia gibbosa Span.*	+	+	+	+	+	+	+	+	+

Result and discussion:

Hitherto we have studied 9 water bodies from Gadhinglaj Tahsil. Study including Diversity of marginal plants of some water bodies in Tahsil and their ecological significance. Marginal plants include plants of Reed-swamp stages and Sedge Marsh or Meadow stage. Majority of the vegetation studied was found to be naturally occurring. About 89 Angiosperms and one fern were recorded around the water bodies.

The plants can be classified in to herb, shrub and trees. The herbs and herbaceous shrub shows maximum diversity, while tree species are rare. The marginal tree species including B, *Butea monosperma, Cassia suratensis*, *Pongamia pinnata*, *Ficus racemosa* etc. shows large no. of presence of bird nest specifically of kites. As well as tree shade enable shadow area on lake-bank and also leaf litter promotes aggregation and growth of aquatic animals. Root cleft

of large trees often found to be home of water snakes and other aquatic animals. Grass Members of Cyperaceae and Poaceae are gregarious in marginal marshes. These seasonal temporary marshy grasss- lands favor the growth insects and extensive population of spiny *Hygrophila* shrub provides food & security for many migrating birds which lays eggs among these grasses. Rooted submerged flora other marginal flora provides substratum for growth of aquatic epiphytic algaes. Thus, a marginal plant also contributes biomass productivity of lakes. Many insects specifically dragonflys are most commonly occurring flying insects in marginal vegetation. Roots of Marginal plants also hold substratum firmly and checks soil erosion and keep water bodies undisturbed and as deep as original. Marginal flora enhances beauty of these water bodies. Only *Ipomoea carnea* found to be harmful to Lake Ecosystem because of extensive growth, and presence of poisonous latex which make it unsuitable as food for many organisms. Hence the marginal flora with some exception is essential for healthy lake ecosystem and to maintain good floral and faunal diversity.

Acknowledgement:

The authors are grateful and sincerely thanks to Principal Dr. M. R. Patil, Dr, Ghali College, Gadhinglaj, Kolhapur for inspiration. The authors are also thankful to Department of Botany, Dr, Ghali College, Gadhinglaj for facilities.

References:

- Sardesai M. M. 2002. 'Flora of Kolhapur district. Shivaji University. Kolhapur. PP 679.
- Yousuf., A. R. and firdous, G. 2001. Food spectrum of mirror carp in a deep mesotropic Himalayan lake. Journal Res. Dev. 1: 60-67.
- Tessier, Celine & Cattaneo, Antonia & Bernadette, Pinel-Alloul & Galanti, Gaetano & Morabito, Giuseppe. (2004). Biomass, composition and size structure of invertebrate communities associated to different types of aquatic vegetation during summer in Lago di Candia (Italy). J. Limnol. 63. 190-198.
- Seabloom, Eric & van der Valk, Arnold. (2003). The development of vegetative zonation patterns in restored prairie pothole wetlands. Journal of Applied Ecology. 40. 92 - 100. 10.1046/j.1365-2664.2003.00764.x.
- Raspopov I. M., Adamec L. & Husák Š. (2002): Influence of aquatic macrophytes on the littoral zone habitats of the Lake Ladoga, NW Russia. – Preslia, Praha, 74: 315–321.

- Jr, Andrew & Nall, Larry & Dyke, Jess. (2011). Effects of Vegetation Control by Grass Carp on Selected Water-Quality Variables in Four Florida Lakes. Transactions of the American Fisheries Society. 112. 777-787.
- Hudon, Christiane. (2004). Shift in wetland plant composition and biomass following low-level episodes in the St. Lawrence River: Looking into the future. Canadian Journal of Fisheries and Aquatic Sciences - CAN J FISHERIES AQUAT SCI. 61. 603-617.
- Crowder., L. B. and Cooper, W. E. 1982. Habitat Structural Complexity and the Interaction Between Bluegills and Their Prey. Ecology Vol. 63, No. 6 (Dec., 1982), pp. 1802-1813
- Chavan, Niranjana & Sawant, Rajaram & Patil, Sachinkumar. (2014). Survey and Mapping of Freshwater Bodies from Gadhinglaj Tahsil of Maharashtra (India) by using GPS. IOSR Journal of Environmental Science, Toxicology and Food Technology. 8. 17-22.

Phytoallelopathic effect of different concentration of *Vitex negundo* L leaf leachates on germination and growth of *Trigonella foenum-graecum* L c.v. Lam selection-1 and *Eleusine coracana* (L). c.v. Dapoli – 1

VIKRAM P. MASAL[a]

[a]P.G.Research Lab. Dapoli Urban Bank Senior Science College Dapoli. Dist:Ratnagiri. (M.S.) 415712, Email.- masalvikram@gmail.com

ABSTRACT:

It is now very well realized that the presence of neighboring plants species can have a significant influence on seed germination growth and yield of crop plant (Rice, 1974). The influence may be either positive or negative depending upon the nature of allelochemical released by the allelopathic plants such allelopathic effect will become more prominent to future agricultural systems because of decrease in farm size, intercropping and crop rotation and introduction of agro forestry. Hence it was though worthwhile to investigate influence of some common prominent plant species which have entered in the agriculture of konkan region, on the seed of germination and growth of seedling. In the present investigation deals with the study of significant Phytoallelopathic effect of different concentration of *Vitex negundo* L leaf leachates on germination and growth of *Trigonella foenum-graecum* L c.v. Lam selection-1 and *Eleusine coracana* (L). c.v. Dapoli – 1. During the experimental period Environmental temperature of Konkan region ranging from 12.02^0c to 34.87^0c and humidity 62% to 93.9%.

Keywords: Allelopathy, *Vitex negundo* L., *Trigonella foenum-graecum* L c.v. Lam selection-1 and *Eleusine coracana* (L). c.v. Dapoli - 1., Konkan.

INTRODUCTION:

Putnam (1985) considered the phenomenon of auto toxicity as a special form of allelopathy that occur when chemical substances released from one plant inhibit or delay germination and growth of same plant species.

Lovett (1990 and Lovett and Ryuntyu (1992) tried to broaden the concept of allelopathy as the complex of delicate communication between plants and also plants and other organisms

and adopted a picture the liberalizes the parameters of allelopathy to include some aspects of plant defense.

MATERIAL AND METHOD:

The Experiment was conducted in Department of Botany, D. U. B. Senior Science College Dapoli. The *Vitex negundo* L. leaf was collected from Dapoli. The plant materials were oven dried at 80 ^{0c} for 42 hrs and then ground to a fine powder. The extract was prepared by soaking 50 grams of dry ground Vitex negundo L. leaves powder in 200 ml Distilled water for 24 hours.

The leaf leachates were filtered and the filtrate was made up 200 ml volume by using distilled water. Which were considered as 100% and then diluted with distilled water and prepare solution of 20%, 40%, 60%, 80%, and 100%. The treatment was replicated four times by using R. B. D. design.

Trigonella foenum-graecum L c.v. Lam selection-1 and *Eleusine coracana* (L) seeds were treated with 0.1% mercuric chloride and washed thrice with distilled water and dried on sterile absorbent paper to avoid fungal attack. Twenty-five seeds of *Eleusine coracana* (L) were tested for germination in 20 cm diameter petridishes containing germinating paper saturated with above concentration of leaf leachates. The moistened petridishes was maintained by adding 2.5 ml leaf leachates solutions.

The percentage of germination, root and shoot length and biomass production of the seedling was recorded after 3 DAS, 5DAS and 7 Days after sowing.

RESULT AND DISCUSSION:

The water extract of *Vitex negundo* L leaves decreased the germination and seedling growth of *Eleusine coracana* (L) and *Trigonella foenum-graecum* L c.v. Lam selection-1 is depicted in Table. The 100% leaf extract of *Vitex negundo* L inhibit the germination percentage. In general, the length of radical and plumule were proportionately affected with increase in concentration of leaf leachates solution as well as with increase in the time of sowing. The radical and plumule length

Table. No. 1. Effect of different concentration of *Vitex negundo L.* leaf leachates on germination and growth of *Trigonella foenum graecum L.*

Treatment	Germination % Days after soaking					Length of Radicle cm Days after soaking					Length of plumule cm Days after soaking				
	3	4	5	6	7	3	4	5	6	7	3	4	5	6	7
T_0 control	94.66	97.33	97.33	97.33	97.33	0.83	2.06	3.16	4.06	4.70	-	2.06	2.93	4.06	4.56
T_1 (20%)	92.00	92.00	92.00	92.00	92.00	0.60	1.40	2.03	2.73	3.16	-	1.33	2.30	2.86	3.50
T_2 (40%)	82.66	82.66	83.00	83.00	83.00	0.50	1.30	1.83	2.43	2.83	-	1.23	2.16	2.10	2.53
T_3 (60%)	58.66	60.00	60.00	60.00	60.00	0.43	1.30	1.66	2.33	2.50	-	1.16	1.86	2.06	2.20
T_4 (80%)	37.33	40.00	40.00	40.00	40.00	0.26	0.86	1.50	1.56	1.60	-	0.80	1.26	1.30	1.33
T_5 (100%)	25.33	26.66	29.33	33.33	37.33	0.13	0.73	1.20	1.26	1.26	-	0.63	0.70	0.83	0.90
SE $\pm$ =	1.668	1.003	1.167	1.310	1.310	0.046	0.0416	0.0459	0.0235	0.0417	-	0.0663	0.0443	0.0512	0.0327
CD at 5%	5.254	3.159	3.676	4.127	4.127	0.144	0.1311	0.144	0.0741	0.1314	-	0.2091	0.1395	0.1615	0.1032

Table No. 2. Effect of different concentration of *Vitex negundo L.* leaf leachates on germination and growth of *Eleusine coracana (L).*

Treatment	Germination % Days after soaking					Length of Radicle cm Days after soaking					Length of plumule cm Days after soaking				
	3	4	5	6	7	3	4	5	6	7	3	4	5	6	7
T_0 control	98.66	100.00	100.00	100.00	100.00	0.57	1.12	1.49	1.76	2.50	0.33	0.56	1.27	1.89	2.15
T_1 (20%)	97.33	97.33	97.33	97.33	97.33	0.32	0.86	1.38	1.78	1.96	0.17	0.44	0.93	1.47	1.60
T_2 (40%)	96.00	96.00	96.00	96.00	96.00	0.30	0.59	0.80	1.20	1.26	0.16	0.36	0.90	1.20	1.16
T_3 (60%)	85.33	85.33	85.66	85.66	85.66	0.26	0.44	0.98	1.12	0.96	0.16	0.28	0.82	1.15	0.93
T_4 (80%)	73.33	78.66	78.66	78.66	78.66	0.10	0.40	0.63	0.91	0.80	0.13	0.20	0.67	1.01	0.93
T_5 (100%)	60.00	62.66	62.66	62.66	62.66	0.10	0.10	0.42	0.53	0.60	0.10	0.11	0.22	0.73	0.70
SE $\pm$ =	1.0036	1.115	1.115	1.115	1.115	1.144	1.5874	0.0112	0.0196	0.0232	0.0201	0.0129	0.0283	0.0719	0.025
CD at 5%	3.161	3.512	3.512	3.512	3.512	0.0256	5.001	0.0355	0.06184	0.0732	0.0633	0.0407	0.0893	0.226	0.0648

Singh and Bawa (1982) observed that the leaf leachates from *Eucalyptus globules* showed inhibitory effect on seed germination of *glaucium flavum*, Crants.

Bhatia and *et.al.* (2005) observed the germination percentage of wheat decreased with the increase in rice straw leachates concentration as compared to control. Rao and *et.al*. (1977) reported that aqueous extract of dry leaves of *Parthenium hysterophrus* L.inhibit the dry weight of plumule and radicals of *Triticum vulgare* L. Rai and Tripathi (1982) reported the leaf lechates from *Eupatorium riparium Regel.* significantly inhibited the radicle and plumule length of Eupatorium *adenophorum* and *Trifolium repens*. He observed that even at lowest concentration of leachates, there was considerable inhibition in radicle and plumule length. From above, our observations are similar line.

REFERENCES:

1.Bhatt, B.P., Chauhan D.S.and Todaria N.P. (1993) Phototoxic effect of tree crop on germination and radical extension of some food crop *Trop.sci.* **33**:69-73

2.Bhatia, R.K., Sukhpreet, Sindhu, M. S., Mehara, S.P., Singh Gurupratap. (2005) Allelopathic effect of rice crop residue of Wheat and Mustard . *Journal of research*. Volume, **42**: pp 412.

3.Hollis, C.A., Smith J.E.and Fisher R.F. (1982) Allelopathic effects of common under storey species on germination and growth of southern pines.*For.sci*.**28:**509-515.

4.Kendrick, R.E.and Frankland, B. (1969) Photo control of germination in *Amaranths caudatus. Planate* **85**:326-329.

5.Molisch H. (1937) Der Enflusslinear Pflanze auf die andere Allelopathic Germany: Gustav Fischer, Jena.

6.Muller C.H. (1969) Allelopathy as a factor in ecological process. *Vegetation* **18:**348-357.

7.Panse, N.G.and Suckhatme P.V. (1985) Statistical methods for Agricultural workers. I.C.A.R. New Delhi, Forth Edition.

8.Rao, J.V.S., K. Ram Mohan Rao and S.S. Murthy (1979) Allelopathic effect of some weed of vegetable crop on the germination and early seedling growth of bajara. *Trop. Eco.* **20(1**):5-8

9.Rao A.N., D.C. Dager and P.S. Dubey (1977) Phyto allelopathic potentials of *Parthenium hystrophorus* Linn. *Ind. j. weed science*. **9(1):**24-32.

10.Rice E.K. (1974) Allelopathy.Academic press, New York.

11. Rai, JPN and Tripathi, R.S. (1982) Allelopathy as a factor contributing to dominance of *Eupatorium riparium* Regal. *Indiand. J.Ecol*. **9 (01)**:14 – 20
12.Singh R and Bawa (1982) Effect of leaf leachates from *E. globules* L. and *Aesculus indico* C. on germination of *Glaucium flavum* Crantz. *Indian J. Ecology* 9(1): 21-28

GIANT AFRICAN SNAIL - *ACHATINA FULICA* (BOWDICH, 1822), A NURSERY PEST FROM KOLHAPUR DISTRICT (M.S.)

Mrunalini N. Desai[1] and Suryakant V. Maske[2]

[1]Department of Botany, Dr. Tanajirao Chorage Arts, Commerce and Science Senior College, Nandwal (Jaital Phata), Kolhapur - 416001.

[2]Department of Zoology, Shri Vijaysinha Yadav College, Peth Vadgaon, Kolhapur-416112

Corresponding Author: Dr. Suryakant V. Mask, Email – drsuryakant27vymp@gmail.com

ABSTRACT: *Achatina fulica* is invasive terrestrial snail can cause serious economic damage to different agricultural crops as well as nursery and garden plants. The extensive rasping, defoliation, slime trails, or ribbon like excrement is signs of infestation. The study was carried out in different nurseries from Kolhapur district. In recent times, severe outbreak of this pest has been noticed due to some desirable agricultural and gardening practices like minimum tillage practices and straw retention techniques which help in survival of snails and make seedlings more susceptible to damage. Present investigation aims to enlighten on taxonomy, appearance, behavior and habitat, dispersal, diet, reproduction pattern, nature of damage and to suggest management strategies.

KEY WORDS: Giant African Snail, *Achatina fulica*, Nursery plants, Management practices.

1. INTRODUCTION

Phylum *mollusca* is the second largest phylum of the animal kingdom [1]. Several species of snails and slugs are considered as notorious pests in agro-ecosystem in different parts of the world due to their rasping feeding behaviors [2]. The Giant African Snail (GAS) *Achatina fulica* (Bowdich, 1822) belongs to the Phylum Mollusca, Class Gastropoda, subclass– Pulmonata, and family– Achatinidae of order–Stylommatophora. This is the biggest and most damaging land snail pests having a protective shell, measuring about 19 cm. in length.

It is an exotic invasive pest introduced from East Africa to India in 1847. Now it is reported from all continents [3]. The World Conservation Union (IUCN) has listed *A. fulica* as one of the world's 100 most invasive species [4]. According to Nelson [5] it is very active during monsoon, nocturnal in behavior and damage 500 different crops like papaya, banana,

brinjal, beans, okra, cucumber, cabbage, cauliflower, pumpkin, ground nut, melon, areca nut, rubber buds, coffee seedlings, orchids, marigold etc.

In India, Reddy and Sreedharan [6] recorded *A. fulica* on coffee in Andhra Pradesh. Sridhar *et. al.,* [7]; Ravikumara *et. al.,* [8] and Mallappa and Patil [9] reported severe occurrence of the GAS in various districts of Karnataka. Badal *et. al.,* [10] focused on Bio ecology and management of GAS *A. fulica.* Avhad *et. al.,* [11] and Jadhav *et. al.,* [12] reported GAS as mulberry pest in Aurngabad district and Kolhapur district of Maharashtra respectively. More recently Pinku and Rafee [13] surveyed molluscan pests in Karnataka. Lenin and Ummer [14] enlightened GAS menace in crops and management in Kerala. Bishal *et. al.,* [15] studied population density and damage in organic farm in east Sikkim. Pradeep Kumar [16] suggested some management strategies for control of GAS in Uttar Pradesh.

GAS is now widely distributed and no longer limited to their region of origin due to several factors viz., high reproductive capacity, voracious feeding habit, inadequate quarantine management and human aided dispersal. It is known for its destructive nature on cultivated crops and garden plants wherever it occurs. The information regarding incidence of molluscan pests in nurseries are lacking. There is little information available on the management of the Giant African Snail at Kolhapur district. The present investigation will help to take steps to eradicate or control snail infestations from Kolhapur district as early as possible.

2. MATERIAL AND METHODS

Survey was carried out for the collection and observation on infestation of *Achatina fulica* in nurseries. Several visits were made to 22 nurseries viz Sajiv Nursery (Kolhapur City), Shinde Nursery (Kolhapur City), Sai Prasad Biotech (Karvir Tahsil), Rushi & Kunal Nursery, Kasaba Bawda (Karvir Tahsil), Kamddhenu Ropvatika , Kothali (Karvir Tahsil), Om Agro Services (Karvir Tahsil), Green Earth Services (Karvir Tahsil), Yashraj Nursery (Karvir Tahsil), Akshay Nursery, Kagal (Kagal Tahsil)Plant library Nursery, Kagal (Kagal Tahsil), Palavi Nursery, Shiye (Hatkanagle Tahsil), Shetkari Nursery, Minache (Hatkanagle Tahsil), Shri Ambika Nursery, Kondigre (Shirol Tahsil), Warana Nursery, Warananagar (Panhala Tahsil) and Ankur Nursery (Radhanagari Tahsil).

Distribution and abundance of *Achatina fulica* were recorded in various nurseries from in and around Kolhapur city. On the basis of infestation four categories of nursery plants like

Ornamental, Flowering, Vegetable and Fruit were made. The data obtained and management strategies are discussed in detail in result.

3. RESULTS AND DISCUSSION

The occurrence and infestation of GAS *Achatina fulica* was observed in various nurseries in Kolhapur district. The infestation was observed on total 22 plants from all four categories of nursery plants like Ornamental, Flowering, Vegetable and Fruit. The list of plants is provided in Table no. 1. In ornamental category maximum infestation was observed on *Syngonium, Spathyphyllum* and *Diefenbachia*; in Flowering plants Hibiscus was infested mostly, in Vegetable category infestation was observed on 6 plants out of these Cabbage and Cauliflower were infested badly. In fruit plant category Banana and Papaya were infested mostly. Nature of infestation, feeding nature, excrement pattern, clinging behavior is as shown in Plate 1.

GAS feeds on leaves, stems, fruits, flowers of the host plants and leafy vegetables causing severe damage especially in nurseries as well as plants of horticultural and medicinal value [17], [18], [13]. It affects the aesthetic value of kitchen garden and roof gardens and nurseries too. Snails and slugs (molluscs) are hermaphrodites, but there is reciprocal exchange of spermatozoa as they mature before development of eggs [19]. Due to the high reproductive potential, a single snail can multiply in the field and it is very difficult to control their population.

a. Appearance: Adults usually around 7-8 cm tall, but may reach 20 cm or more. Shell has rounded conical shape, being about twice as high as it is broad. Shell is generally brown in color with irregular darker streaks running transversely across the whorls [20]. Adult size is reached in 4 months, but may continue slowly up to 1 ½ years. It is cross-fertilizing, egg-laying hermaphrodite [17]. Number of eggs per clutch averages around 200 with 5-6 clutches per year. Hatching viability is about 90%. Locally, the eggs and snails are readily transported in garden waste.

b. Behavior & Habitat: The giant African snail commonly is found in warm, humid climates. They can be found in coastal areas, shrub lands, plantation habitats and forests. The snail prefers temperatures that are well above freezing [21], [22]. It is nocturnal and spends most of

the day underground. These snails produce a slime that reduces friction and allows them to move along many ground surfaces.

c. Diet: GASs are herbivores. They typically feed on leaves, wood, bark, seeds, grains and nuts. Older snails can become carnivorous, however, also feed on living plants or other snails, fungi or animal matter. Their tongue having radula that allows to scrape or cut food.

d. Reproduction: The typical life span of the GAS is 3-5 years, but they have been known to live as long as 9 years. They are hermaphrodites. Young African snails only produce sperm, but adults are able to produce both sperm and eggs. Even though they have both male and female reproductive parts, they still have to mate with another snail because their sperm cannot fertilize their own eggs.

When two snails mate, they exchange sperm. The sperm may immediately fertilize the eggs, or it can be stored inside the body for up to two years before fertilizing any eggs. Once fertilized, the snail does not lay the eggs for 8 to 20 days. They typically hatch 11 to 15 days later. The snail can lay up to 100 eggs in its first year and up to 500 in the second year. After six months, the young reach adult size.

MANAGEMENT STRATEGIES:

Eradication of GAS is difficult and costly. It is literally impossible for well established populations in agricultural field. The effective control of pests involves a combination of measures, including physical, cultural, biological and chemical methods so it is best not to rely on just one method. The different management practices are discussed below:

a. Physical Method: Hand picking of snails and eggs on daily basis after sunset and destroy or incinerate them with a flame proves best eradication practice in heavily infested areas. Food baits (over-ripe papaya fruit pieces) can be used for easy collection and removal of snails from any field. International quarantine and surveillance practices are necessary to avoid their entry in any new geographical area.

b. Cultural control: One can reduce the infestation and population of GAS by practicing good field sanitation. Good hygiene, weed control and removal of refuges can reduce the problem over time. Regular Monitoring is essential for the pest in the nursery or in garden. Abundant ground cover and vegetation growth provide favorable conditions like ideal moisture levels,

shelter and harborage where snails thrive and can be a problem. Avoidant of minimum tillage and straw-retention techniques in *A. fulica* endemic areas are effective since these practices not only help the snails to survive but also make the seedlings more susceptible to damage. Soils with more organic matter content are more attractive to the snails.

Unnecessary growing of plants between trees and vines can also act as shelter belt for the snails. Sprinkling of table salt around the crop-base in dry season is one of the best preventive measures. Prasad *et.al.,*[3] suggested and experimentally proved *Annona glabra* softwood cutting fence is a feasible and practical alternative to protect nursery beds from *Achatina fulica*.

c. Biological control: Since *A. fulica* is an alien pest therefore there are limited natural enemies that control this pest. Some predatory beetles, lizards, birds and rats can feed on them. Ducks and chickens can provide effective, long-term control in orchards and vineyards, if an appropriate breed is chosen and properly cared for. Khaki Campbell or Indian runner ducks are best breed to be used in snail control [23], [24], [25] and [26].

Use of predatory snails and worms in *A. fulica* management has also been implicated in the decline of native snails in many countries. Some of the predatory snails which can predate and feed on *A. fulica* include *Euglandina rosea, Gonaxis kibweziensis, Gonaxis quadrilateralis, Edentulina ovoidea and Edentulina affinis. Platydemus manokwari, a turbellarian* flat worm, has also been used to control the GAS in Guam, Philippines and Maldives.

d. Chemical control: Some of the chemicals are effective to control this species. However, it should be advisable to use the chemicals judiciously. Lime or bleaching powder may be sprinkled in the infested area was effective. Common salt may also be spread on the snail infested area.

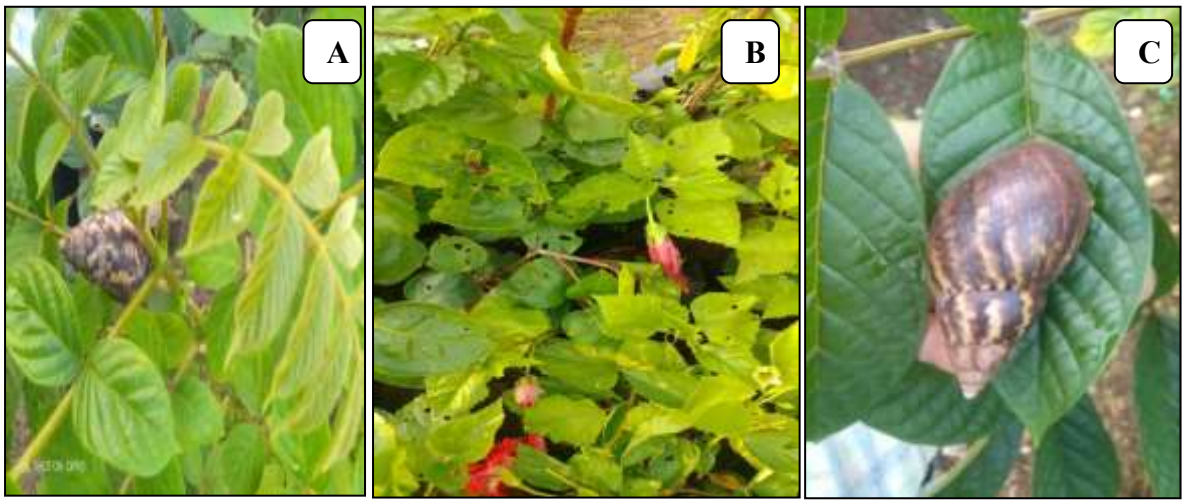
Few chemicals which are effective to control the snails are methiocarb, metaldehyde and EDTA. The bait materials such as dicholorvos bait (Wheat flour- 1kg + Jaggery- 0.2 kg + Dicholorvos 76EC- 250ml) and methomyl bait (Rice bran 1kg + Jaggery 0.2 kg + Methomyl 40 SP- 100 g) are suitable to control the infestation of the species. The bait preparation should be carried out prior to application of molluscicides. The bait should be prepared by heating the

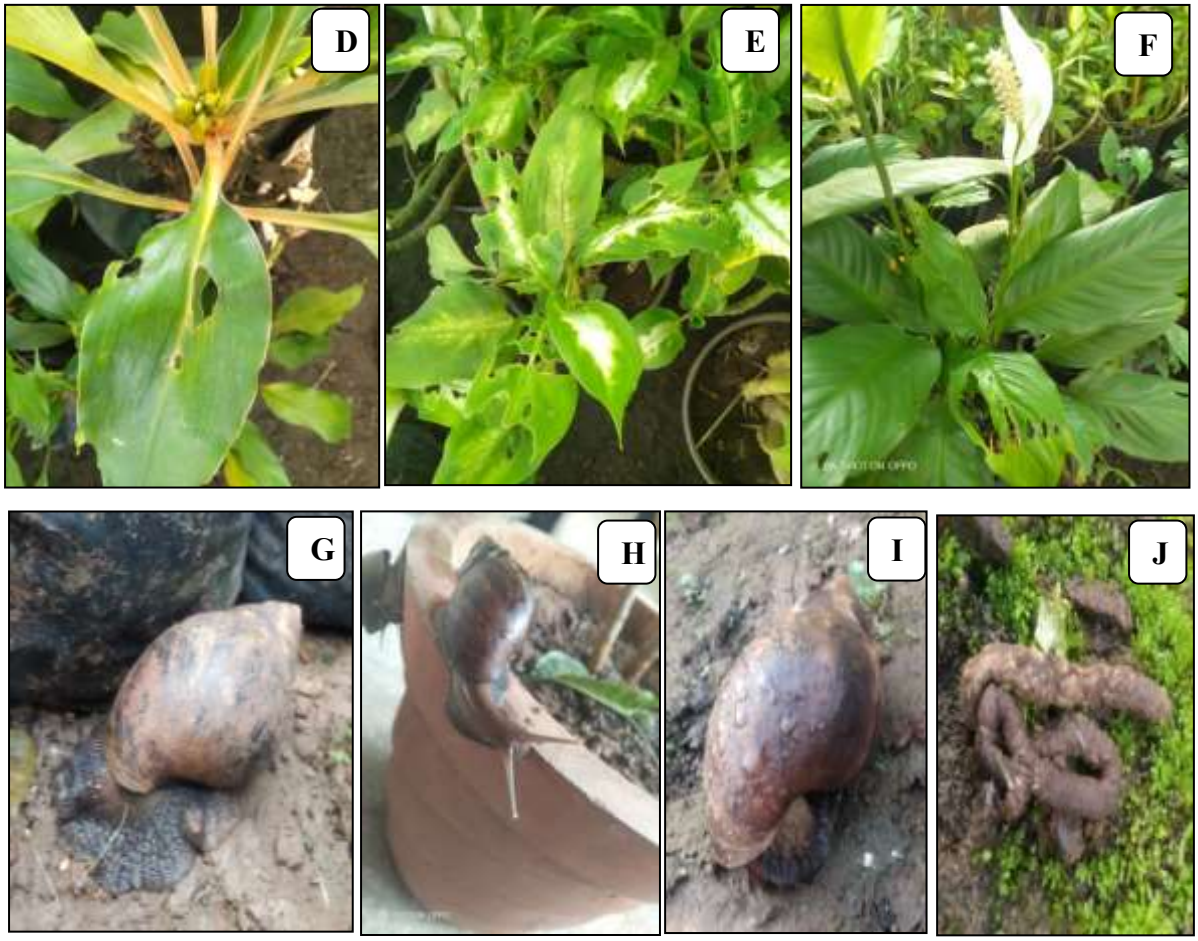
jaggery with wheat flour/ rice bran along with the poison. Hand gloves should be used to make small balls and keep it in 10 places in the field.

Table No. 1. List of Nursery Plants infested by *A. fulica.*

	Ornamental		**Flowering**		**Vegetable**		**Fruit**
1.	*Aglonema*	1.	Hibiscus	1.	Brinjal	1.	Banana
2.	*Syngonium*	2.	Marigold	2.	Tomato	2.	Jamun
3.	*Spathiphyllum*	3.	Aster	3.	Cauliflower	3.	Papaya
4.	*Dieffenbachia*	4.	Petunia	4.	Cabbage	4.	Mango
5.	*Chlorophytum*			5.	Capsicum	5.	Moringa
6.	Bird Cherry			6.	Curry Leaf	6.	Pumpkin

Plate 1. Giant African Snail *Achatina fulica* and its infestation on various nursery plants





A- GAS on Bird cherry plant **B- Infestation on Hibiscus** **C- GAS clinging to host plant**
D- Infestation on *Chlorophytum* **E- Infestation on *Dieffenbachia*** **F- Infestation on *Spathiphyllum***
G- GAS hiding under bags **H- GAS on Nursery Pots** **I- GAS in moist places**
J- Excrement of GAS

4. CONCLUSION

Invasive species are one of the top threats to biodiversity. Once an invasive species establishes its population in a new vulnerable area, it is very difficult to check its growth, spread and damages. In case of Giant African Snails, several eradication measures have already been found unsuccessful. Some of the management options also have lots of indirect issues related to environment, biodiversity and health hazards. Biological control in the form of introducing the rosy wolf snail proved disastrous and caused even more damage, razing of an entire ecosystem in the pursuit of eradicating only one species. Use of toxic baits targeted for *A. fulica* also victimized indigenous as well as other invasive snails. As regards to chemical control,

various molluscicides like metaldehyde are non-selective, thus their use has a chance of endangering the survival of non-target organisms.

However, some easy techniques like collection and destruction of the snails and their eggs are recommended as a form of physical control. Guarding pathways through which Giant African Snails can pass is much cheaper than pursuing them through biological or chemical control. Moreover, to be effective, the molluscicides should be such that it may not get dissolved and washed away by rain because snails are normally active during the rainy season. Therefore, an effective eco-friendly management strategy is needed to keep the pest below economic injury level. Holistic efforts in and around Kolhapur city at the regional level are not only needed to prevent further spread of *A. fulica* but also required to formulate an effective and environmentally sustainable management strategy.

REFERENCES:

1. Zala, M. B., Sipai, S. A., Bharpoda, T. M., and Patel, B. N. Molluscan pests and their management: A review. An International e. Journal, 2018; 7(2): 126-132.

2. Das, P. P. G., Bhattacharyya, B., Bhagawnti, S., Dev, E. B., Manpoong, N. S., and Bhairavi, K. S. Slug: A menace in agriculture: A review. Journal of Entomology and Zoology Studies; 2020; 8(4): 01-06.

3. Prasad, G.S., Singh, D.R., Senani, S. and Medhi, R.P. Eco-friendly way to keep away pestiferous giant African snail, *Achatina fulica* Bowdich from nursery beds. *Curr. Sci.,* 2004; 87: 1657– 1659.

4. Invasive Species Specialist Group. Global Invasive Species Database. Version 2012. 2. *Achatina fulica* (mollusc). http://www.issg.org/database/species/ecol ogy.asp? si=64&fr=1&sts=&lang=EN. 2012

5. Nelson Scot. Injuries Caused by the Giant African Snail to Papaya Miscellaneous Pests Published by Department of Plant and Environmental Protection Sciences, Hawaii University, 2012: 1-7.

6. Reddy KB and Sreedharan K. Record of giant African snail, *Achatina fulica* Bowdich on coffee in Visakha agency areas, Andhra Pradesh. *Indian Coffee.* 2006; 70: 17-19.

7. Sridhar V, Jayashankar M, Vinesh LS and Verghese. A Severe occurrence of the giant African snail, *Achatina fulica* (Bowdich) (Stylommatophora: Achatinidae) in Kolar district, Karnataka.*Pest Management in Horticultural Ecosystem.* 2013; 18: 228-230.

8. Ravikumara, N. M. I., Manjunath, M., and Pradeep, S. Seasonal incidence of giant African snail, *Achatina fulica* Bowdich (Gastropoda: Achatinidae) in areca ecosystem. Karnataka Journal of Agricultural Science. 2007; 20(1):157-158.

9. Mallappa, C., and Patil, R. K. Population dynamics of giant African snail, *Achatina fulica* Ferussac (Stylommataphora: Achatinidae) in betelvine ecosystem. Journal of Experimental Zoology. 2014; 17(1):285-288.

10. Badal Bhattacharyya, Mrinmoy Das, Himangshu Mishra, D.J. Nath and Sudhansu Bhagawati. Bioecology and management of giant African snail, *Achatina fulica* (Bowdich) International Journal of Plant Protection. 2014; 7 (2) : 476-481.

11. Avhad S.B., Shinde K. S. and C. J. Hiware. Record of molluscan pest in mulberry Aurangabad District of Maharashtra. Indian J. Seric. 2013; 52 (1): 29 - 33.

12. Jadhav, A. D., Dubal, R.S., Bagade R. P., Sanadi Reshma A., Kamble, P.L. Belgumpe S. and Sathe, T.V. Giant African Snail, *Achatina fulica* Bowdich a destructive pest of V1 mulberry (*Morus alba* L.) by - A new report and control strategies from Kolhapur, Maharashtra, India. 2016.

13. Pinku Paul and C.M. Rafee. A Survey on Molluscs Pests in Karnataka, India *Int.J.Curr.Microbiol.App.Sci.* 2017; 6(9): 3123-3132

14. Lenin Neena and Ummer Najitha. Giant african snail menace in crops and its Management. Ph. D. Thesis submitted to Kerala Agricultural University, Thiruvananthapuram, Kerala, India. 2018.

15. Bishal Thakuri, Bhoj Kumar Acharya and Ghanashyam Sharma. Population Density and Damage of Invasive Giant African Snail *Achatina fulica* in Organic Farm in East Sikkim, India *Indian journal of Ecology.* 2016; 46(3):631-635

16. Pradeep Kumar. A Review- On Molluscs as an Agricultural Pest and Their Control. International Journal of Food Science and Agriculture. 20202; 4(4).

17. Mead, A.R. *The Giant African Snail: A problem in economic malacology.* University of Chicago Press, Chigo, 1961: 257.

18. Muniappan, R., Duhamel, G., Santiago, R.M. and Acay, D.R. Giant African snail control in Bugsuk Island, Philippines, by *Platydemus manokwari. Oleagineux,* 1986; 41:183-186.
19. Routray, S. and Dey, D. Snails and slugs as crop pests. Rashtriya Krishi, 2016; 11(1): 40-41.
20. Skelley, P.E. Dixon, W.N. and Hodges, G. Giant African land snail and giant South American snails: field recognition. Florida Department of Agriculture and Consumer Services. Gainesville, Florida (U.S.A.). 2011.
21. Raut, S.K. and Barker, G.M. *Achatina fulica* Bowdich and other Achatinidae as pests in tropical agriculture. In: G.M. Barker (ed.), Molluscs as crop pests. CABI Publishing, Hamilton, New Zealand, 2002 : 55-114.
22. Smith, J.W. and Fowler, G. Pathway risk assessment for Achatinidae with emphasis on the giant African land snail, *Achatina fulica* (Bowdich) and *Limicolaria aurora* (Jay) from the Caribbean and Brazil, with comments on related taxa *Achatina achatina* (Linne) and *Archachatina marginata* (Swainson) intercepted by PPQ. USDA, APHIS, *Center for Plant Health Science and Technology* (Internal Report), Raleigh (N.C.). 2003.
23. Howlett, S. A. Terrestrial slug problems: Classical biological control and beyond. CAB Review, 2012; 7(051): 1-10.
24. Peter, D., Widmer, M. and Craven, T. Control of pest snail and slugs. Western Australian Agriculture Authority, *Garden note,* 2012; 12: 530.
25. Sallam, A. and Wakeil, N. E. Biological and Ecological Studies on Land Snails and Their Control. In: Integrated management and pest control-Current and Future Tactics, (Eds. Larramendy, ML. and Soloneski, S. Published online web.org. pp. 2009; 413-444.
26. Schuder, I. Integrated control of slug and snail pest in hardy nursery stock (Ph.D. thesis). Newcastle upon Ty and Wear, U.K. 2004.