Effect of Acid Rain on *Vigna radiata*

Nilima Gajbhiye

***Abstract*—**The acid rain causes change in pH level of soil it is directly influence on root and leaf growth. Yield of the crop was reduced if acidity of soil is more. Acid rain seeps into the earth and poisons plants and trees by dissolving toxic substances in the soil, such as aluminum, which get absorbed by the roots. In present investigation, effect of acid rain on crop *Vigna radiata* was studied. The effect of acid rain on change in soil fertility was detected in which pH of control sample was 6.5 and pH of 1% H_2SO_4 and 1% HNO^3 were 3.5. Nitrogen nitrate in soil was high in 1% HNO_3 treated soil & Control sample. Ammonium nitrogen in soil was low in 1% HNO_3 & H_2SO_4 treated soil. Ammonium nitrogen was medium in control and other samples. The effect of acid rain on seed germination on 3rd day of germination control sample growth was 6.1cm with plumule 0.001% HNO_3 & 0.001% H2SO4 was 5.5cm with plumule and 8cm with plumule. On 10th day fungal growth was observed in 1% and 0.1% H_2SO_4 concentrations when all plants were dead. The effect of acid rain on crop productivity was investigated on 3rd day roots were developed in plants. On 12th day *Vigna radiata* showed more growth in 0.1% HNO_3 and 0.1% H_2SO_4 treated plants as compare to control plants. On 20th day development of discoloration of plant pigments were observed on acid treated plants leaves. On 34th day *Vigna radiata* showed flower in 0.1% $HNO_{3,}$ 0.01% HNO_3 and 0.01% H_2SO_4treated plants and no flowers were observed on control plants. On 42th day 0.1% $HNO_{3,}$ 0.01% HNO and 0.01% H_2SO_4 treated *Vigna radiata* variety and control plants were showed seeds on plants. In *Vigna radiate* variety 0.1%, 0.01% $HNO_{3,}$ 0.01% H_2SO_4treated plants were dead on 46th day and fungal growth was observed. The toxicological study was carried out *on Vigna radiata* plants exposed to 1% HNO_3 cells were damaged more than 1% H_2SO_4. Leaf sections exposed to 0.001% HNO_3 & H_2SO_4 showed less damaged of cells and pigmentation observed in entire slide when compare with control plant.

***Keywords*—**Acid rain, pH, *Vigna radiate*, HNO_3 & H_2SO_4.

I. Introduction

ACID rain is a broad term referring to a mixture of wet and dry deposition from the atmosphere containing higher than normal amounts of nitric and sulfuric acids. The precursors of acid rain formation result from both natural sources, such as volcanoes and decaying vegetation, and man-made sources, primarily emissions of sulphur dioxide (SO_2) and nitrogen oxides (NO_X) resulting from fossil fuel combustion [16]. Acid rain occurs when these gases react in the atmosphere with water, oxygen, and other chemicals to form various acidic compounds. The result is a mild solution of sulfuric acid and nitric acid. When sulfur dioxide and nitrogen oxides are released from power plants and other sources, prevailing winds blow these compounds across state and national borders, sometimes over hundreds of miles [10], [15]. Acidic deposition has adverse effects on vegetation. This is mainly due to soil acidification and the uptake of substances, which disturb the pH levels within plant cells that may lead to the evolution of reactive radicals [13]. Pure water has a pH of 7.0. However, normal rain is slightly acidic because carbon dioxide (CO_2) dissolves into it forming weak carbonic acid, giving the resulting mixture a pH of approximately 5.6 at typical atmospheric concentrations of CO_2 [9]. Acid rain seeps into the earth and poisons plants and trees by dissolving toxic substances in the soil, such as aluminum, which get absorbed by the roots [10]. Acid rain also dissolves the beneficial minerals and nutrients in the soil, which are then washed away before the plants and trees have a chance of using them in order to grow. When there is frequent acid rain, it corrodes the waxy protective coating of the leaves. When this protective coating on the leaves is lost, it results in making the plant susceptible to disease. When the leaves are damaged, the plant loses its ability to produce sufficient amounts of nutrition for it to stay healthy. Once weakened, the plant becomes vulnerable to the cold weather, insects, and disease, which can lead to its death [6]. Seedlings of winter barley, perennial ryegrass and white clover were grown on a range of British soils for 21–24 weeks and exposed to simulated acid rainfall treatments of pHs 5·6, 4·5, 3·5 and 2·5. Whilst leaves of white clover developed leaf lesions after 18 weeks of exposure to the pH 2·5 treatments, there were no signs of visible injury to the other two species [1].

In experiments with radishes, leaf and root growth were consistently reduced when acidity was increased from pH 3·5 to 2·5, the effect on roots being accentuated by an interaction with sulphite [5]. For one year of the study, yield of B73 × Mo17 (corn) was reduced 3139kg ha^{-1} by the most severe drought, and an additional 1883kg ha^{-1} by acid rain of pH 3·0, as compared to the control (pH 5·6) [3]. The effects of simulated sulphuric acid rain were investigated, under controlled laboratory conditions, on the surface structure and *n*-alkane composition of the lichen *Pseudevernia furfuracea*. The response to simulated acid rain was a clear change in the quantitative alkane composition, with a decreasing trend observed for C_{28} and C_{30} with increasing sulphuric acid concentration [12]. The pH 2.0 treatment seemed to be a threshold level for inhibition of seed germination and seedling growth for all the treated species [6]. The growth of *Aspergillus niger*, *A. flavipes*, *Trichoderma viride* and *Penicillium brefeldianum* was reduced or completely inhibited in soils acidified below pH 3.5 [4].

Leaf beetle larvae were fed on foliage treated during 6–7 years with simulated acid rain of pH 3 (both H_2S0_4 and HNO_3) or with spring water of pH 6.The beetles reared on acid

Dr. Nilima D. Gajbhiye is with the Ramnarain Ruia College, Mumbai, 400019, INDIA (phone: 9221782379; e-mail: drneem@yahoo.com).

treated birches were more susceptible to predators than those reared on irrigated control trees [11]. In Rampur India rainwater pH was detected, Rainwater pH varied from 5.9 and 7.4 with volume weighted mean pH of 6.6 [7]. In this study, I the effects of simulated acid rain exposure and its effects on the germination and growth of the plant are examined.

II. Materials and Methods

A. To Detect the Effect of Acid on Soil Fertility

1.pH of the Soil

1gm soil sample was weighed and 2ml distilled water added to it in the test tube. The soil sample was mixed thoroughly. Then the soil sample was filtered through Whatmann paper 1 and pH of the sample was detected. The plastic pots containing approximately 750gms of soil to which separate concentration of acid samples were added and pH of the each soil sample were noted.

2.Ammonium Nitrogen in Soil and Nitrogen Nitrate in Soil

Soil analysis was done by kit method. Kits were procured from HiMedia, India.

B. To Detect the Effect of Acid Rain on Seed Germination

Soaked seeds of *Vigna radiata* were kept in a humidity chamber. The humidity chamber was made by keeping filter paper in petriplates. Different concentrations of acids were made and poured 5ml each day in respective petriplates. Observations were made every alternate day.

C. To Investigate the Effect of Acid Rain on Crop Productivity

Soaked seeds of *Vigna radiata* was ploughed in plastic 7-5 cm diameter pots containing 750gms of soil and allowed to become established. Supplementary watering of the plants was provided at the rate of 40ml/day' using simulated acid rain solution of pH2.5- 4.5 (Different concentrations of acids were made from sulphuric and nitric acids 1% to 0.001% concentrations) and poured every day in respective pots. Observations were made every alternate day. The pH of the rain solution was comparable with that for rainfall in upland areas of India [2].

D. To Detect the Effect of Acid Rain on Health of the Plants

Toxicological studies, on plant variety were carried out; Plant varieties were exposed to different concentration of HNO_3 and H_2SO_4 for 46th days. Leaves were removed by cutting the petiole near the base cut petioles were coated in petroleum jelly to prevent water loss from the open wounds and then the leaves were placed in open Petri dishes on a laboratory bench [2]. After 15th days of exposure of plants to different concentrations of acids showed decolouration of plant pigment on leaves so leaf sections were taken and observed under LOBO High magnification microscope.

III. Result and Discussion

A. The Effect of Acid Rain on Soil Fertility

TABLE I
pH of Soil Samples

	Sample	Observed pH of soil sample	Colour of Soil sample
1	1% H_2SO_4	3.5	Pink
2	0.1% H_2SO_4	4.0	Yellow
3	0.01% H_2SO_4	4.0	Pinkish Brown
4	0.001% H_2SO_4	4.5	Pink
5	Control	6.5	Yellow

TABLE II
pH of Soil Sample

	Sample	Observed pH of soil sample	Colour of Soil sample
1	1% HNO_3	3.5	Red
2	0.1% HNO_3	4.5	Yellow
3	0.01% HNO_3	4.5	Pinkish Brown
4	0.001% HNO_3	4.5	Pink
5	Control	6.5	Yellow

TABLE III
Ammonium Nitrogen in Soil

HNO_3	Ammonium nitrogen in soil	H_2SO_4	Ammonium nitrogen in soil
1%	Low about 15	1%	Low about 15
0.1%	Medium about 75	0.1%	Medium about 75
0.01%	Medium about 75	0.01%	Medium about 75
0.001%	Medium about 75	0.001%	Medium about 75
Control	Medium about 75	Control	Medium about 75

Ammonium nitrogen in soil was low in 1% HNO_3 & H_2SO_4 treated soil Ammonium nitrogen was medium in control and other samples.

TABLE IV
Nitrogen Nitrate in Soil

HNO_3	Nitrogen nitrate in soil	H_2SO_4	Nitrogen nitrate in soil
1%	High about 50	1%	Low about 10
0.1%	Medium about 30	0.1%	Very low about 4
0.01%	Low about 10	0.01%	Very low about 4
0.001%	Low about 10	0.001%	Medium about 20
Control	High about 50	Control	High about 50

Nitrogen nitrate in soil was high in 1% HNO_3 treated soil & Control sample. Nitrogen nitrate was low in lower concentration of HNO_3, medium in 0.001% H_2SO_4 treated soil and very low in other samples.

TABLE V
Available Phosphate in Soil Kg per Hectare as (P_2O_5)

HNO_3	Available phosphate in soil	H_2SO_4	Available phosphate in soil
1%	Medium about 22to 56.	1%	Medium about 22to 56.
0.1%	Medium high about 56 to 73	0.1%	Medium high about 56 to 73
0.01%	Low less than22	0.01%	Medium high about 56 to 73
0.001%	Medium about 22to 56.	0.001%	Medium about 22 to 56
Control	Low less than22	Control	Low less than 22

Available phosphate in soil was low in 0.01%HNO_3 treated sample and control sample. 0.1% HNO_3, 0.1% & 0.01% H_2SO_4 Medium high phosphate content was found but rest of the samples showed medium phosphate content.

B. The Effect of Acid Rain on Seed Germination

TABLE VI
3RD DAY READINGS FOR *VIGNA RADIATA*

HNO_3	Germination of seeds	H_2SO_4	Germination of seeds
1%	0.2cm	1%	0.6cm
0.1%	4.0cm	0.1%	2.5+0.2cm plumule
0.01%	6.0cm	0.01%	1.9+0.8cm
0.001%	5.5cm+ plumule 1cm	0.001%	8cm+1cm
Control	6.1cm+plumule1cmand roots developed	Control	6.1cm+plumule1cmand roots developed

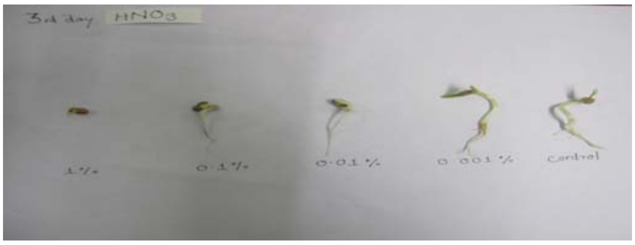


Fig. 1 Germination of *Vigna radiata* seeds treated with HNO_3 concentrations

Fig. 2 Germination of *Vigna radiata* seeds treated with H_2SO_4 concentrations

TABLE VII
5TH DAY READINGS FOR *VIGNA RADIATA*

HNO_3	Germination of seeds	H_2SO_4	Germination of seeds
1%	0.8cm	1%	0.6cm
0.1%	7.5cm	0.1%	3.5cm
0.01%	10cm	0.01%	2.5cm
0.001%	15 cm	0.001%	12.5cm
Control	15cm	Control	15cm

TABLE VIII
7TH DAY READINGS *VIGNA RADIATA*

HNO_3	Germination of seeds	H_2SO_4	Germination of seeds
1%	0.8cm	1%	Fungal growth observed
0.1%	9.0cm	0.1%	4.0cm with roots
0.01%	12cm	0.01%	2.5cm
0.001%	15 cm	0.001%	15cm
Control	18cm	Control	18cm

Fig. 3 Germination of *Vigna radiata* seeds treated with HNO_3 & H_2SO_4 concentrations on 7th day. On 10th day fungal growth was observed in 1% and 0.1%H_2SO_4 concentrations when all plants were dead

C. The Effect of Acid Rain on Crop Productivity

Fig. 4 H_2SO_4 treated *Vigna radiata* plants on 3rd day

TABLE IX
5TH DAY READINGS *VIGNA RADIATA*

HNO_3	Germination of seeds	H_2SO_4	Germination of seeds in pot
1%	3.5cm	1%	7cm
0.1%	9.5cm	0.1%	9.0cm
0.01%	9.5cm	0.01%	9.5cm
0.001%	9.0cm	0.001%	10cm
Control	9cm	Control	9cm

TABLE X
7TH DAY READINGS *VIGNA RADIATA*

HNO_3	Germination of seeds	H_2SO_4	Germination of seeds in pots
1%	4.5cm	1%	12cm
0.1%	16cm	0.1%	14cm
0.01%	14.5cm	0.01%	11.5cm
0.001%	14. 5cm	0.001%	13.5cm
Control	14.58cm	Control	14.5cm

Fig. 5 *Vigna radiata* showed more growth in 0.1% HNO_3 treated plants

Vigna radiata showed same length as control plant (24cm) in 0.1% $HNO_3$and 0.1% H_2SO_4treated plant showed 19cm length. 1% H_2SO_4 treated plant had died on 26th day after development of discoloration of plant pigments.

Fig. 6 Dicoloration of plant pigments were observed

Vigna radiata showed flower in 0.1% HNO_3, 0.01% HNO_3 and 0.01% H_2SO_4 treated plants and no flowers were observed on control plants on 34th day.

Fig. 7 Flowers were observed on 0.1% HNO_3 treated *Vigna radiate* plants

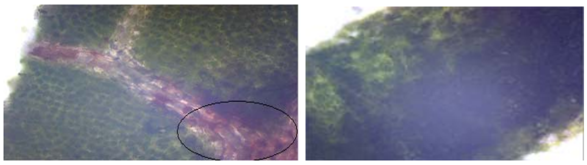
On 42th day, In *Vigna radiata* 0.1% HNO_3, 0.01%HNO and 0.01% H_2SO_4treated plants showed seeds on plants.

Fig. 8 *Vigna radiata* 0.1% HNO_3, 0.01% HNO_3 treated plants showed seed

Fig. 9 Dead plants of *Vigna radiata*

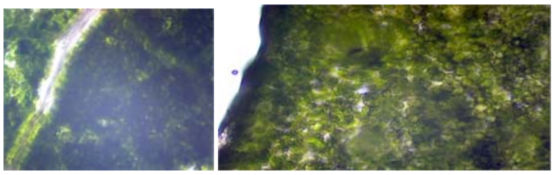
D.The Effect of Acid Rain on Health of the Plants

Main findings; plant cells were ruptured, uneven distribution of chlorophyll pigments were observed. *Vigna radiata* plants exposed to 1% HNO_3 cells were damaged more than 1% H_2SO_4. Leaf sections exposed to 0.001% HNO_3 & H_2SO_4 showed less damaged of cells and pigmentation observed in entire slide when compare with control plant.



(a) (b)

Fig. 10 (a) Leaf section of *Vigna radiata* plant exposed to 1% HNO_3 concentration (b) Leaf section of *Vigna radiata* plant exposed to 1% H_2SO_4 concentration



(a) (b)

Fig. 11 (a) Leaf section of *Vigna radiata* plant exposed to 0.001%HNO_3 concentration (b) Leaf section of *Vigna radiata* plant exposed to 0.001%H_2SO_4 concentration

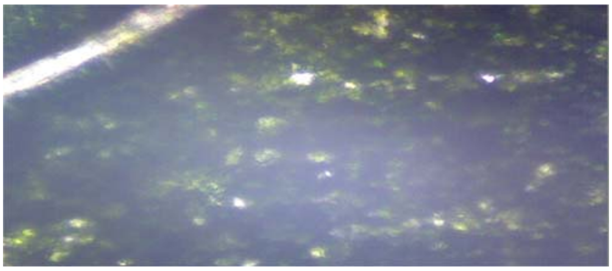


Fig. 12 Leaf sections of *Vigna radiata* control plant

Tables I and II showed the variation of pH to 0.1 to 0.001% HNO_3 and H_2SO_4 acid treated soil samples. The lowest rain pH values measured by [14] and [9] observed the acidity of the precipitation in Shanghai was considerably high with the annual mean pH value of 4.49 and the frequency of acid rain was 71%. The lowest pH of the rain event reached 2.95.

In present investigations, soil fertility was detected by checking essential parameters of the soil. Tables III-V listed Ammonium nitrogen was low at 1% HNO_3 and H_2SO_4 but medium in 0.1 to 0.001% HNO_3 and H_2SO_4 as well as control samples. Nitrogen nitrate was high in 1% HNO_3 and control sample but low in 1% to 0.001% H_2SO_4. Available nitrogen was less in control samples but increased in acid treated samples. The findings were different from [8] and [4]. The soil fertility was found increasing in higher concentrations of acid from 0.001 to 0.1% H_2SO_4 & HNO_3 in present investigations but acid rain dissolves the beneficial minerals and nutrients in the soil was reported by[6].

Germination of the seed and seedling growth was more in acid treated samples. As shown in Table VI at pH 2.5 the germination of seeds was fast and plumule developed early than control plants. *Vigna radiata* showed fast germination and seedling growth as compare to other crop. When pH was increased from 4.5 to 2.5 leaf and root growth was normal. This was in agreement to observations made by other workers [1].

Figs. 1 and 2 showed when pH was increased from 4.5 to2.5 seed germination and seedling growth was faster than controlled one. The similar results were reported by other workers that seedling growth was stimulated at pH levels between 3.5 and 5.0. [6]. In present investigations, Table VIII listed that when pH was increased from 4.5 to 3.5, crop varieties tolerated the increased pH and the growth of the plants was more than controlled plants. This was not in agreement with other workers [5] reported that. In experiments with radishes, leaf and root growth were consistently reduced when acidity was increased from pH 3·5 to 2·5.and [6] reported that the pH 2.0 treatment seemed to be a threshold level for inhibition of seed germination and seedling growth for all the treated species results were different in our study it was revealed that seed germination was found.

Figs. 4, 5 and Table IX, X showed that *Vigna radiata* showed more growth in 0.1% HNO_3 and 0.1% H_2SO_4 treated plants as compare to control plants.

In the present study surprisingly leaf and root growth of crop variety (*Vigna radiata)* was normal for 15 days. Though the growth was equally good in 44 days of the study, Fig 6 showed that the leaves were found dipigmented at a larger scale.

Fig. 7 showed *Vigna radiata* plants were germinated fast and growth of the plant was higher in increasing concentrations of HNO_3 and H_2SO_4. On 34th day of experiments flowers were on 0.1% and 001% HNO_3 and in H_2SO_4 treated *Vigna radiata* plants and In Fig. 8 observed seeds appeared on 42th day of experiments. Results were appeared much faster than gaseous pollutant and acid mist [2].

Fig. 9 showed that in *Vigna radiate* variety 0.1%, 0.01% HNO_3, 0.01% H_2SO_4treated plants were dead on 46th day and fungal growth was observed and showed fungal growth.

Fig. 10 showed the leaf sections with rupture of cells and brown pigments were observed in the sections of both crop varieties at 1% HNO_3 and H_2SO_4 concentrations but Fig. 11 showed less damage in 0.001% HNO_3 and H_2SO_4 concentration as compare to control plants as showed in Fig. 12. Plant cells were ruptured, uneven distribution of chlorophyll pigments were observed [6].

ACKNOWLEDGMENT

The Author would like to thanks sponsorship and financial support by University of Mumbai, INDIA and facilities and moral support by Ramnarain Ruia College Mumbai, 400 019, .Maharashtra, India.

REFERENCES

[1] T.W. Ashenden and S.A.Bell,"The effects of simulated acid rain on the growth of three herbaceous species grown on a range of British soils" Environmental pollution.,vol.48(4),pp295-310. 1987.

[2] T. W. Ashenden, S. A. Bell and C. R. Rafarel. "Responses of white clover to gaseous pollutants and acid mist: implications for setting critical levels and loads," New Phytol.vol.130,**pp.** 89-9, 1995.

[3] W.L., Banwart, "Field evaluation of an acid rain-drought stress interaction," Environmental pollution.,53(1-4),123-133,1998.

[4] R.J.F Bewley., and G**.**Stotzky, "Simulated acid rain (H_2SO_4) and microbial activity in soil" Soil Biology and Biochemistry.,vol.15(4),pp.425-429,1983.

[5] S.A Harcourt and J.F Farrar., Environmental Pollution Series A, Ecological and Biological.,vol. 22(1),pp. 69-73,1990.G. O. Young, "Synthetic structure of industrial plastics (Book style with paper title and editor)," in *Plastics*, 2nd ed. vol. 3, J. Peters, Ed. New York: McGraw-Hill, 1964, pp. 15–64.

[6] Hou Bao Fan and Yi Hong Wang**.**, **"**Effects of simulated acid rain on germination, foliar damage, chlorophyll contents and seedling growth of five hardwood species growing in China" *Forest Ecology and Management*, Vol. 132, issue 2-3, p. 285, July 1, 2000,ISSN: 0378-1127.

[7] N Das., R .Das., G.R Chaudhury. and S. N.Das**,** **"**Chemical Composition of Precipitation at Background Level," Atmospheric Research,vol.95(1),108-113,2010.

[8] JS Jacobson_, JJ Troiano, LI Heller , L Osmeloski . "Effect of fertilizer on the growth of radish plants exposed to simulated acidic rain containing different sulfate to nitrate ratios". Environ Pollut 44(1):71-9, 1987

[9] Kan Huang .,Guoshun Zhuang ., Chang Xu .,Ying Wang and Aohan Tang **.,** **"**The chemistry of the severe acidic precipitation in Shanghai, China" Atmospheric Research.,vol.89(1),pp.149-160,2008.

[10] Larssen, Thorjørn; Seip, Hans Martin; Anne G Semb,; Jan Mulder,; I.P Muniz,.; Vogt, David Rolf ; Lydersen, Espen et al. "Acid rain and its effects in China," Environmental Science and Policy. ISSN 1462-9011. Vol. 2, pp 9- 24,1999.

[11] Masahide Aikawa, Takatoshi Hiraki and Jiro Eiho., **"** Ambient nano and ultrafine particles from motor vehicle emissions: Characteristics, ambient processing and implications on human exposure," Atmospheric Environment,42(30),7043-7049,2008.

[12] R.Piervittori , L Usai., F .Alessio and M.Maffei**,** "The effect of simulated acid rain on surface morphology and***n***-alkane composition of ***Pseudevernia furfuracea."***.The Lichenologist,vol 29(2),pp191-198.1997.

[13] H,.Rennenberg , A .Gessler (2001) Acid rain. Nature encyclopedia of life sciences. Nature Publishing Group, London

[14] **R.**Tsitouridou and Ch Anatolaki **"**On the wet and dry deposition of ionic species in the vicinity of coal-fired power plants, northwestern Greece," Atmospheric Research, 83 (1), 93-105 (2007).

[15] Yoko Nagase and C .Emilson, .D. Silva **."**Acid rain in China and Japan: A game-theoretic analysis**"** Reginal Science and Urban Economics.,37(1),100-120,2007.

[16] Zhao Dianwu, Xiong Jiling, Xu Yu and Walter H. Chan., **"**Acid rain in southwestern China,"Atmospheric Environment (1967).,22(2), 349-358(1988).

Nilima D. Gajbhiye is Assistant professor, Department of Lifesciecnes, Ramnarain Ruia College, Mumbai, 400019 INDIA.DOB:28/11/1973. Qualifications: M.Sc., D.M.L.T., ph.D.(Lifesciences, University of Mumbai., March , 24th 2003).Member of Mumbai Immunology group 2011, Faculity member of Dept. of Lifesciences. Teaching and guiding students of B.Sc. and M.Sc. degree of Mumbai, University for ten years. Has research interest in field of microbiology, environmental science, genetic engineering and immunology. Guided more than 25short research projects, So for published five research papers out of which two were read in international conferences in Germany and Mumbai India.