Comparison of Phytochemical and Antibacterial Screening of Leaves and Latex of *Calotropis Procera*

Tripti Joshi, Student, Sardar Bhagwan Singh University, Dehradun, India, joshitripti98@gmail.com

Pooja Naudiyal, Assistant Professor, Sardar Bhagwan Singh University, Dehradun, India, poojabamrara_2007@yahoo.co.in

Alok Maithani, Associate Professor, Sardar Bhagwan Singh University, Dehradun, India, alokmaithani@gmail.com

***Neetu Pandey, Assistant Professor, Sardar Bhagwan Singh University, Dehradun, India, neetu_bhtt@yahoo.co.in**

Abstract:

Objective: To compare and investigate the antibacterial and phytochemical screening of leaves and latex extract of *Calotropis procera*

Method: Extract of petroleum ether, ethanol, chloroform and water was made and all the mentioned extract were than evaluated for the phytochemical test and the antibacterial activity.

Result: The antibacterial activity of leaf and latex extract has been reported with three bacteria's namely *Escheria coli, S.aureus, B.cereus* the results revealed that the best extraction solvent for antibacterial activity of leaf and latex extract is of alcohol followed by chloroform and water.

Conclusion: The results revealed that the latex extract of *Calotropis procera* has better antibacterial effect as compare to the leaf extract of *Calotropis procera.*

Keywords —antibacterial, activity, evaluation, latex, screening, phytochemical

I. INTRODUCTION

India has been known for its use on herbal drugs of the plant origin since the ancient past. Looking back in times we could find the people are very much dependent on sources as they are beneficial. The home remedies and DIY has been the most common and favourite way of dealing with diseases for us Indian as we've practiced this since ages and now it has been incorporated in our genes to search for these ingredients in our kitchen or in the nature. Research has explored the nature of the secondary metabolites that in various medicinal plants. Medicinal plants shows a precious, renewable source for new drugs. The world health organisation has claimed that 80% of the world's population rely on the herbal method of treatment of various diseases Around 500,000 plant spices were estimated but only a small amount has been investigated phyto-chemically. More than 130 drugs in the world's markets comes from higher plants either directly or synthetically [1], [2], [3], [4], [5]. Although hundreds of plants were tested for antifungal and antibacterial properties, the majority of them have not been adequately evaluated and processed well. [6].

The whole Himalayan belt of gods world is the home of several medicinal plants in India as not only the mythological facts says so but always the science has given approval to this very fact. Himalayan Region with background information on family, habit and nativity. A total of 190 invasive alien species under 112 genera, belonging to 47 families have been recorded. Among these, the dicotyledons represent by 40 families, 95 genera and 170 species; monocotyledons represent by 7 families, 17 genera and 20 species. The analysis of invasive species reveals that 18 species have been introduced intentionally,

 DOI : 10.35291/2454-9150.2022.0374

while the remaining species established unintentionally through trade [7].

Calotropis procera is a plant of Asclepiadaceae family and it is a large broad leaf evergreen plant with a strong odour, abundant in the tropical regions of Asia and Africa, which is commonly known as Milkweed Apple of seldom and many more names. *Calotropis procera* is used as a folk medicine and is not a new name in Indian household as it is used as ornamental plant due to beautiful white flower. It has been reported that the plant possess potential antimicrobial ,anthelmintic, anti-inflammatory, anticancer, purgative, anticoagulant, analgesic, and antipyretic characteristics and is also used in the treatment of leucoderma, leprosy, liver and abdomen diseases [8]. The latex of *Calotropis procera* has been known for important indigenous medicinal uses due to its laxative, antisyphilitic and analgesic action [9]. *Calotropis procera* flowers causes temporary paralysis of red stomach worm in sheep and notably reduces egg count percent of gastrointestinal nematodesin naturally infected sheep [10]. Dry latex of *Calotropis procera* has potential anti-cancer properties due to its differentiable targets and non-interference with regular pathway of apoptosis [11]. The pharmacological properties of *Calotropis procera* is a versatile plant for the pharmaceutical industry to develop new drugs [12]. Medicinal plants have no doubt remained the major sources of traditional medicine worldwide [13]. The main objective of this research work was to analyze the various solvent extracts obtained from the leaf, seed and stems bark of *Calotropis procera* and qualitatively screened them for phytochemicals using standard tests. Successful extraction, determination and isolation of biologically active components from plant material are largely dependent on the type of solvent [14].

II. MATERIALS AND METHODS

Collection: The leaves and latex of *Calotropis procera* has been collected from local areas of Balawala, Dehradun the plant is identified at that place by the means of standard key and description.

Preparation of plant extract: Leaves of *Calotropis procera* dried in shade and avoid direct contact of sunlight and pulverization method is used which is as follows.

The dried leaves were macerated in the liquid such as hexane, isopropyl, ethyl acetate, ethanol, methanol, acetone for 48 hours. The latex was collected in sterile plastic/glass bottle by squeezing the apex and tips of leaves and kept in refrigerator at 4 ˚Celsius. [15]

The latex was then dried under shade at ambient temperature with the yield of 20 gram/100 ml. To remove the chlorophyll content, the sample was extracted with petroleum ether. 20 ml of latex then further extracted with petroleum ether in the separating funnel after the formation of two separate layers of petroleum ether and residue the same is repeated with other solvents also.

Test organism-To study antibacterial effect of the very plant 3 bacteria's were taken namely,

Escheria coli, S.aureus, B.cereus from the department.

Photo Chemical Screening: [16], [17], [18], [19], [20], [21], [22], [23], [24], [25].

1. Test for carbohydrates:

Molisch's test: Take 2-3 ml of extract and added few drops of 95% napthol solution in alcohol. After shaking, conc. H_2SO_4, was added from the sides of the test tubes. Appearances of violet ring at the junction of two layers indicate the positive test for reducing sugar.

Benedict's solution test: Equal volume of Benedict's reagent and extract were mixed in the test tube. Heat it in boiling water bath for 5 mins. Appearance of red coloured solution indicates the positive test for reducing sugar.

2. Test for alkaloids:

Dried extract was dissolved in dilute HCl. Filtered and subjected the filtrate to the following tests.

Test with Dragendorff's reagent: Take 2-3 ml of filtrate add few drops of Dragendorff's reagent. Formation of orange brown precipitates reveals the positive test for alkaloids.

Test with Mayer's reagent: Take 2-3 ml filtrate, add few drops of Mayer's reagent. Formation of cream coloured precipitates reveals the positive test for alkaloids.

Test for Hager's reagent: Take 2-3 ml of filtrate, add few drops of Hager reagent. Formation of yellow coloured precipitates reveals the positive test for alkaloids.

3. Test for proteins and amino acids:

Biuret test: Take 2-3 ml of aqueous extract added 4% NaOH and few drops of 1% CuSO4 solution. Violet or pink colour is formed, if proteins are present.

Ninhydrin solution test: Heat 3 ml of extract and 3 drops of 5% ninhydrin solution in boiling water bath for 10 mins. The development of violet or purple colour shows the presence of amino acid.

4. Tests for steroids:

Liebermann-Burchard reaction: Mix 2 ml of extract with chloroform. Add 1-2 ml of acetic anhydride and 2 drops of conc, sulphuric acid from the sides of test tubes. Development of green colour reveals the positive test for the steroid.

Salkowaski reaction: Take 2 ml of extract, 2ml of chloroform and 2ml conc. sulphuric acid. After shaking appearance of red colour in chloroform layer and greenish

 DOI : 10.35291/2454-9150.2022.0374

yellow fluorescence in acid layer reveals the positive test for steroid moiety.

5. Test for flavonoids:

Shinoda's test: Take 2ml of extract, 2ml ethanol, few drops of conc. HCI and little amount of magnesium turning. Appearance of pink colour reveals the positive test for flavonoids.

Lead acetate solution test: Take small quantity of extract added lead acetate solution. Appearance of yellow colour precipitate reveals the positive test for flavonoids.

6. Test for glycosides:

Bontrager's test: Take 2-3 ml of extract, add dilute H_2SO_4 boil it and filter. Now add equal volume of chloroform to filtrate. After shaking chloroform layer was separated. Add ammonia, appearance of red colour in ammonia layer revealed the positive test for anthraquinone glycosides.

III. RESULTS AND DISCUSSION

Antibacterial test [26]

Antibacterial activity of chloroform, ethanol, aqueous extract *of Calotropis procera* over determined by Agar well diffusion method.

Phytochemistry [27]

Table-1: Phytochemical screening of *Calotropis procera* leaf extract

Solvent	%yield	Alkaloids	Flavnoids	Saponins	Tannin	Glycoside	Amino acid	Carbohydrate
Chloroform	2.1	+	_	++	+	_	_	++
Ethanol	6.15	+++	_	+++	+	++	_	+
Pet.Ether	1.31	_	_	_	_	+	+	_
Water	7.55	+	-	-	+	+	-	+

+ = Present - = Absent

Table-2: Phytochemical screening of *Calotropis procera* latex extract

Ingredient	Ethanol	Pet.ether	Aqueous	Chloroform
%Yield	1.66	0.87	0.32	0.56
Reducing sugar	++	+	++	-
Tannins	+++	+	++	+
Steroid glycoside	++	-	+++	+
Resin	_	+	+	+
Alkaliods	_	+	_	_
Saponins	+	+	+	+
Flavonoids	++	+	++	+

+ = Present - = Absent

From the above table 2 it has been observed that the ethanolic extract of the latex was obtained in maximum amount i.e 1.66% followed by Pet.ether(0.87%).The minimum % yield was obtained in aqueous extract i.e 0.32%.

When the leaf and latex extract of petroleum ether, chloroform, , ethanol, and water was analyse or tested for the presence of alkaloids flavonoids saponins, tannins, stair instance glycoside, carbohydrate and amino acids it was found that the different extract shows positive test for the certain constituent as shown in Table-1 and 2.

When the yield of extract of leaf was observed, it was found that the maximum yield was found in the water extract that is about 7.55%, followed by ethanol 6.15%. The minimum amount of extract was formed by the petroleum Ether that is 1.31%.

When ethanol, chloroform and aqueous extracts of *Calotropis procera*`s leaf was subjected for the phytochemical test, the results observed were that the petroleum ether extract is rich in glycoside and amino acid whereas ethanol and chloroform extracts are rich in tannins, carbohydrates, reducing sugar, alkaloids and saponins also some alkaloids found abundantly in aqueous extract.

When the latex extract of *Calotropis procera* was subjected to phytochemical investigation of mainly ethanol, chloroform and aqueous extract, it was found that the ethanol extract is rich in reducing sugar, tannins, steroids, flavonoids and loaded with saponins whereas chloroform is mainly rich in tennins and saponins. Aqueous extract has tannins, steroids, resin, saponins and flavonoids. [27, 28]

Zone of inhibition

In the leave and latex extract the zone of inhibition in *S.aureus* bacteria (Table-3) the concentration that we took were 50 mg per ml and 100 mg per ml of solvent extract for *S.aureus* bacteria only. It has been observed that the maximum zone of inhibition was found for the ethanol followed by petroleum ether latex extract when taken in 100 mg/ml concentration, followed by chloroform leaf extract when taken in 100mg per ml concentration which was 13mm, 11mm and 10mm respectively. When the results were compared to the standard that is

 DOI : 10.35291/2454-9150.2022.0374

chloramphenicol it has been observed that it shows a zone of inhibition of 15 mm for petroleum ether extract and 19 mm for ethanol where as it shows 15 mm of zone of inhibition for chloroform extract (Figure-1).

Similarly when zone of inhibition was observed for the bacteria *Bacillus cereus* (Table-4) the different extract of solvent of leaf and latex were treated in different concentration for the bacteria, the zone that has been observe for *Bacillus cereus* bacteria is very less as comparison to S.aureus the maximum zone of inhibition has been shown by the petroleum ether latex extract when taken in 50 mg per ml concentration that is 6 mm whereas the ethanol extract in 50 mg/ml shows of zone of inhibition of 5 mm. Whereas the petroleum ether latex extract when taken in 20 mg/ml concentration it shows zone of 4 mm other than that chloroform and petroleum ether leaf extract shows zone of negative the standard chloramphenicol shows zone of inhibition of 17 mm, 15 mm and 14 mm for petroleum ether, ethanol and chloroform respectively (Figure-2).

When the zone was observe for *E.coli* bacteria, (Table-5) extracts were taken in the concentration of 20 mg/ml and 50 mg per litre the maximum zone was found for ethanol leaf extract that is 6 mm at the concentration of 50mg/ml and ethanol leaf extract that is 5 mm for 50 mg per ml concentration. The chloroform extract of latex and leaf when taken in 20mg/ml concentration show no zone of inhibition. It has been seen that extract of *Calotropis procera* is less effective against *E.coli* bacteria by the observation (Figure-3).

The *S. aureus* shows zone of 11mm for both leave and latex ethanol extract, whereas the water extract for leaf and latex shows a zone of 11 and 12 mm while chloroform leaf extracts shows zero zone of inhibition while latex shoes 10 mm of zone. Chloroamphienicol shows zone of 14 mm when taken in 25 mg/ ml. [29]

The Zone of inhibition for *Bacillus cereus* when recorded by others was found that the zone of aqueous extract of leaf when taken in 30 mg/ml concentration is found to be 14.32mm and for methanol it is 18.24mm [30].

Similarly for *E.coli* bacteria the ethanol shows zone of 11mm and 7mm for leaf and latex extract. Water extract shows on of 10mm and 7 mm for both leaf and latex while the standard shows are zone of 13 mm [29]

Zone of Inhibition (Bacteria - S.aureus) - Petroleum ether extract of Leaves and Latex of *Calotropis procera* (Table-3)

	Bacteria - S.aureus											
	Pet.ether latex		Pet.ether leaf		Ethanol latex		Ethanol leaf		Chloroform latex		Chloroform leaf	
Con.(mg/ml)	50	100	50	100	50	100	50	100	50	100	50	100
ZOI in (mm)	-ve	11	4	5	5	8	-ve	8	2	6	7	10
Chloroamphenicol 25mg/ml	15mm				19mm				15mm			

Zone of Inhibition (Bacteria - Bacillus cereus) - Petroleum ether, chloroform and ethanol extract of Leaves and Latex of *Calotropis procera* (Table-4)

	Bacteria - Bacillus cereus											
	Pet.ether latex		Pet.ether leaf		Ethanol Latex		Ethanol leaf		Chloroform latex		Chloroform Leaf	
Con.(mg/ml)	20	50	20	50	20	50	20	50	20	50	20	50
ZOI in (mm)	4	6	-ve	3	2	5	1	2	1	2	-ve	1
Chloroamphenicol 25mg/ml	17mm				15mm				14mm			

Zone of Inhibition (Bacteria – E. coli) - Petroleum ether, chloroform and ethanol extract of Leaves and Latex of *Calotropis procera* (Table-5)

	Bacteria - E.coli											
	Pet.ether latex		Pet.ether leaf		Ethanol Latex		Ethanol leaf		Chloroform latex		Chloroform Leaf	
Con.(mg/ml)	20	50	20	50	20	50	20	50	20	50	20	50
ZOI in (mm)	1	4	-ve	2	3	5	2	6	-ve	5	-ve	1
Chloroamphenicol25mg/ml	15mm				17mm				13mm			

 DOI : 10.35291/2454-9150.2022.0374

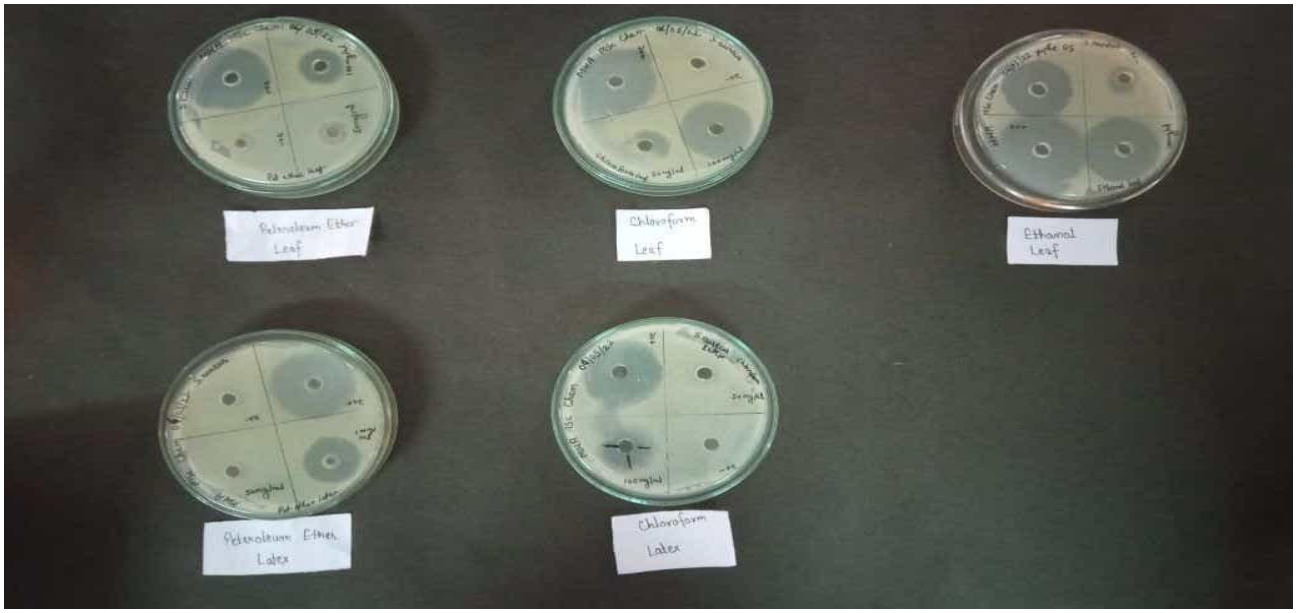

Figure-1 Zone of Inhibition For *Staphylococcus aureus*

Figure-2 Zone of inhibition for *Bacillus cereus*

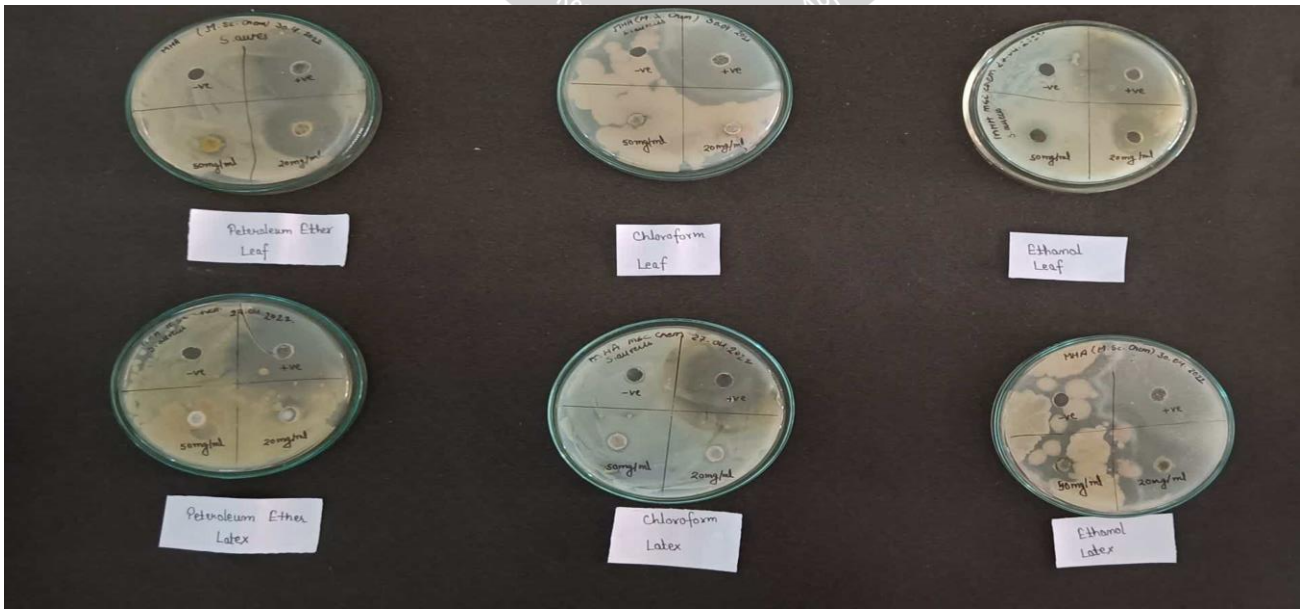

Figure-3 Zone of Inhibition for *E. Coli* bacteria

 DOI : 10.35291/2454-9150.2022.0374

IV CONCLUSION

80 grams of leaves were taken for the practical the whole 80 grams were macerated for 48 hours in the respective solvent to get the extract the solvent, where used according to the polarity .The percentage yield of leaf extract work petroleum Ether was found to be 1.31%t the alcohol was 6.15 %, chloroform was 2.1%, water extract was 7.5%.Water content has the maximum percentage yield. About 40 ml of latex is used and the percentage yield was found maximum for ethanol equals to 1.66%, followed by petroleum ether extract equals to 0.87%.

The zone of inhibition for *Staphylococcus aureus* for petroleum ether latex extract was shown as that 50 mg/ml then give no zone of inhibition whereas 100 mg/ml of Petroleum ether extract give a zone of inhibition of 11 mm, whereas has when petroleum leaf extract was taken in the concentration of 50 mg per ml and 100 mg per ml they give us only zone of inhibition of 4 and 5 mm respectively .The ethanol latex and ethanol leave extract of 50 mg per ml give a zone of inhibition of 5 and 7 mm while when taken in 100 mg per ml it gives the zone of inhibition of 8 mm and 10 mm respectively. Chloroform latex extract when taken in 50 mg per ml gives to a zone of inhibition of 2 mm, while when it is taken in 100 mg per ml it give us own of inhibition of 6 mm. Chloroform leaf extract when taken in 50 mg per ml gives a zone of inhibition of 7mm and while the 100 mg per ml concentration gives the zone of innovation of 10 mm.

For the *Bacillus cereus* bacteria it has been found that petroleum ether latex extract when taken in concentration of 20 MG per ml and 50 mg per ml gives a zone of any vision of 4 mm and 6 mm respectively, while when petroleum ether extract of leave was taken in the concentration 50 mg/ml gives the zone of 2 mm and 3 mm ,whereas ethanol latex extra when taken in 20 and 50 mg per zone of innovation of 2mm and 5mm respectively ,whereas for leaf it is negative and 2 mm for 20 mg/ml and 50 mg/ml respectively. For chloroform extract of latex when taken in 20 mg per ml gives 1mm of zone of inhibition while when taken in 50 mg per ml give 1 mm as zone of inhibition.

For *Escherichia coli* bacteria when zone of any inhibition was observed for petroleum ether latex when taken in concentration of 20 mg per ml and 50 mg per ml was 4 and 6 mm respectively, whereas for Petroleum Ether leaf extract it was found to be 2 mm and 8 mm for 20 mg/ ml and 50 mg/ ml respectively. The ethanol latex and ethanol leaf extract for 20 MG per ml and 50 mg per ml is 5 and 2 for 20 MG, 2 and 6 for 50 mg per ml respectively. The chloroform latex extract for 20 mg/ml shows no zone of inhibition whereas 50 mg /ml shows a 5 mm zone of inhibition, as the latex the latex when taken in 20mg/ml gives no zone of inhibition whereas when the amount is increased upto 50 mg/ml it gives a zone of 1mm.

From the above results we can finally conclude that the *S.aureus* bacteria stated showing zones at a concentration higher than 20mg/ml whereas for the other 2 bacteria smaller concentration was good enough to get a zone .Hence we can conclude that for *S.aureus* the smaller concentration of leaf and latex was not enough for antibacterial effect.

When the *B.cereus* and E.coli was observed for antibacterial activity it has been found that smaller concentration is effective for antibacterial activity as it started showing zone in 20mg/ml concentration.

It can be concluded from the above result that the leaf has greater antibacterial activity as it shows zone greater for the same concentration of latex.

REFERENCES

[1] R.P. Borris, Natural products research: Perspectives from a major pharmaceutical company, J Ethnopharmacol 51 (1996), 29–38.

[2] J.M. Kong, N.K. Goh, L.S. Chia and T.F. Chia, Recent advances in traditional plant drugs and orchids, Acta Pharmacol Sin 24 (2003), 7–21. N. Morsy et al. / Phytochemical analysis of Calotropis procera 273

[3] S.M.K. Rates, Plants as source of drugs, Toxicon 39 (2001), 603–613.

[4] A. Gurib-Fakim, Medicinal plants: Traditions of yesterday and drugs of tomorrow, Mol Aspects Med 27 (2006), 1–93.

[5] D.J. Newman, G.M. Cragg and K.M. Snader, The influence of natural products upon drug discovery, Nat Prod Res 17 (2000), 215–234.

[6] M.F. Balandrin, J.A. Klocke, E.S. Wurtele and W.H. Bollinger, Natural plant chemicals: Sources of industrial and medicinal materials, Science 228 (1985), 1154–1160

[8] S.C. Jain, R. Sharma, R. Jain and R.A. Sharma, Antimicrobial activity of Calotropis procera, Fitoterapia 67 (1996), 275–277.

[9] Z. Iqbal, M. Lateef, A. Jabbar, G. Muhammad and M.N. Khan, Anthelmintic activity of Calotropis procera (Ait.) Ait. F. flowers in sheep, J Ethnopharmacol 102 (2005), 256–277.

[10] M. Larhsini, M. Bousaid, H.B. Lazrek, M. Jana and H. Amarouch, Evaluation of antifungal and molluscicidal

 DOI : 10.35291/2454-9150.2022.0374

properties of extracts of Calotropis procera, Fitoterapia 68 (1997), 371–373.

[11] T. Choedon, G. Mathan S. Arya, V.L. Kumar and V. Kumar, Anticancer and cytotoxic properties of the latex ofCalotropis procera in a transgenic mousemodel of hepatocellular carcinoma, World J Gastroenterol 12 (2006), 2517–2522.

[12] M.C.C. Silva, A.B. da Silva, F.M. Teixeira, P.C.P. de Sousa, R.M.M. Rondon and J.E.R.H. Junior, Therapeutic and ´ biological activities of Calotropis procera (Ait.) R, Br Asian Pac J Trop Med 3 (2010), 332–336.

[13] Goyal, M. and Mathur, R. 2011. Antimicrobial potential and Phytochemical analysis of plant extracts of Calotropis procera. Int. J. of Drug Discovery and Herbal Research, 1(3):138-143.

[14] P. Tiwari, B. Kumar, M. Kaur, G. Kaur, H. Kaur, Int. Pharm. Sciencia, 2011, 1, 98-106.

[15] Verastegui MA,Sanchez CA,Heredia NL,Santos GAJ (1996)Antimicrobial activity of ectracts of three major plants from the Chihuahuan desert .J. Ethnopharmacol. 52:175-177.

[16] H. Doshi, H. Satodiya, M.C. Thakur and F. Parabia, A. Khan, Phytochemical Screening and Biological Activity of Calotropis procera (Ait). R. Br. (Asclepiadaceae) Against Selected Bacteria and Anopheles stephansi Larvae, Int J Plant Res 1 (2011) 29–33.

[17] J.S. Mossa, M. Tariq, A. Moshin, A.M. Angeel, M.A. Al-Yahya, M.S. Al-Said and S. Rafatullah, Pharmacological studies on aerial parts of Calotropis procera, Am J Chin Mesd 3 (1991), 223–231.

[18] M. Ramaprabha, and K. Vasantha, Phytochemical and antibacterial activity of Calotropis procera (Ait.) R.Br. flowers. Int J of Pharma and Biosci 3 (2012), 1–6.

[19] M.M. Mainsara, B.L. Aliero, A. A. Aliero and M. Yakubu, Phytochemical and Antibacterial properties of root and leaf extracts of Calotropis procera, NJBAS 20 (2012), 1–6.

[20] A. Mohsin, A.H. Shah, M.A. Alaha, M.O. Tariqi and A.M. Ageel, Analytic anti-pyretic activity and phytochemical

[21] N.R. Farnsworth, Biological and phytochemical screening of plants, J pharm Sci 55 (1996), 225–276.

[22] J.B. Harborne, Phytochemical Methods, A guide to modern techniques of plant analysis, Chapman and Hall: New York, 1998;1–150.

[23] A. Sofowora, Medicinal plants and traditional medicine in Africa, Spectrum books limited: Ibadan, Nigeria, 1993; 150–289.

[24]] K.R. Brain, T.D. Tuner, The practical evaluation of phytopharmaceuticals, Wright Scientectica Publishers: Bristol, 1975; 57–58.

[25] Trease GE, and Evans WC. A textbook of pharmacognosy, Academic press: London, (1989). 22–40.G.E

[26] Jorgensen JH, Turnidge JD and Washington JA.Antibacterial susceptibility tests: Dilution and disk diffusion methods. In: Manual of clinical microbiology, Washington, DC: ASM Press,1999;7: 1526–1543.

[27] Abegunde S. M.1 , Ayodele-Oduola R.O.2 1, 2 Department of Science Technology, The Federal Polytechnic, P.M.B. 5351, Ado Ekiti, Nigeria.

[28] Raginee Verma", G. P. Satsangi, and J. N. Shrivastava Microbiology Laboratory, Department of Botany, Faculty of Science, Dayalbagh Educational Institute, Dayalbagh, Agra.

[29] SalemWM., Sayed W F, HaridyM. and HassanNH.Department of Botany, Faculty of Science, South Valley University, Qena, Egypt Department of Pathology and Clinical Pathology, Faculty of Veterinary Medicine, South Valley University, Qena, Egypt

[30] Toheeb DY, Wahab O,Okunowo¹R I. Afolayan, Abdulakeem R. Agboola HY, Lukman, AS, Abafi JM. Department of Bochemistry, Faculty of Basic Medical Sciences, College of Medicine, University of Lagos, Akoka-Yaba, Lagos, Nigeria *Department of Medical Laboratory Sciences, Gbagada General Hospital, Lagos State, Nigeria. Department of Biochemistry, University of Calabar, Calabar Cross-river State Nigeria. "Department of Chemical Sidences, Biochemistry Unit, College of Natural and Applied Sciences, Summit University Offa, PMB 4412, Kwara State, Nigeria Department of Biological sciences, Federal polytechnic, PMB 55, Bida Niger State Department of chemical sciences. Federal polytechnic, PRE 55, Bida Niger State.1;(2):2022:36-46

 DOI : 10.35291/2454-9150.2022.0374