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ANDROGRAPHOLIDE BIOACTIVE COMPOUND OF *ANDROGRAPHIS PANICULATA* FROM DIFFERENT REGION OF NARSINGPUR (M.P.)

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ABSTRACT
Andrographis paniculata belonging to family Acanthaceae was analysed to evaluate the content of andrographolide. **Materials and methods**: Nine different samples were analysed from different sites of Narsingpur (Madhya Pradesh) region. Samples were analysed through HPLC method. **Results**: Highest percentage of Andrographolide in *Andrographis paniculata* plant were from Gudarai site of Narsingpur that accounts to 1.057%, followed by Kishlai 0.997%, Gorshi 0.646%, Barpani 0.196%, Gotitoria 0.107%, Gangai 0.102%, Gorshi 0.070%, Peeperpani 0.044% and Hirnapur 0.040%. **Conclusion**: There was considerable variation in the percentage of andrographolide from one site to another site of Narsingpur region.

KEYWORDS: Andrographolide, *Andrographis paniculata*, Narsingpur region, HPLC.

INTRODUCTION
Andrographis paniculata in Acanthaceae family is an important medicinal plant. It is an annual herb also called kalmegh in some parts of India. It is available in many countries but native to China, India and Srilanka. It is key material used by tribes of many communities in India using for treating general debility, fever, jaundice, skin diseases.[7,10]

In India, its forests are found in West Bengal, Madhya Pradesh, Assam, Chhattisgarh, Kerala, Karnataka and Uttar Pradesh. It is found in every type of environment in pine, evergreen, deciduous forest areas, along roads and villages. It grows in every type of soil.[8] It grows in every type of habitat but for the best production of *Andrgraphis paniculata* plain area best suitable for their production.[2]

It was studied that there was comparative variation in andrographolide from one region to another region.[6] Four lactones, like neoandrographolide deoxydidehydroandrographolide, deoxyandrographolide and andrographolide were found in *Andrographis paniculata*[11] and are mostly used in clinics.[13] Andrographolide was found in high concentration in leaves part than in any other part of the plant.[12]

Its hepatoprotective activity has been reported by many researchers due to their andrographolide content tested in different solvent systems.[1,14,15] Immunostimulant activity in ethanolic extract of plant.[9] Neoandrographolide (100-150 mg/kg) was found to possess antiinflammatory activity while it shows both antiinflamtentary[3] and antiulcerogenic[5] activity. Andrographolide was reported to possess antiallergic[4] and antidiabetic activity in animal models.[4]

MATERIALS AND METHODS
Plant material: *Andrographis paniculata* was collected from different sites of Narsingpur region. Shoot part of the plant was cleaned with tap water and was allowed to dry in shade for fifteen days so that constituents will not alter. Dried part of plant was grinded in mortar and pestle then were allowed to make powder with the help of electric mixer. The powdered part were used in methanol solvent and allowed to run in soxhlet apparatus to get semisolid paste. Semisolid paste of the plant part were analyzed by HPLC method to evaluate the content of *Andrographis paniculata*.

HPLC Instrument: HPLC instrument that was used for the estimation of andrographolide were of the following features, HPLC- grade waters, Pump - 515 Isocratic pump, Injector - Rheodyne injector with a 20-microlitre loop, Detector - UV Vis detector, Software - Data ace

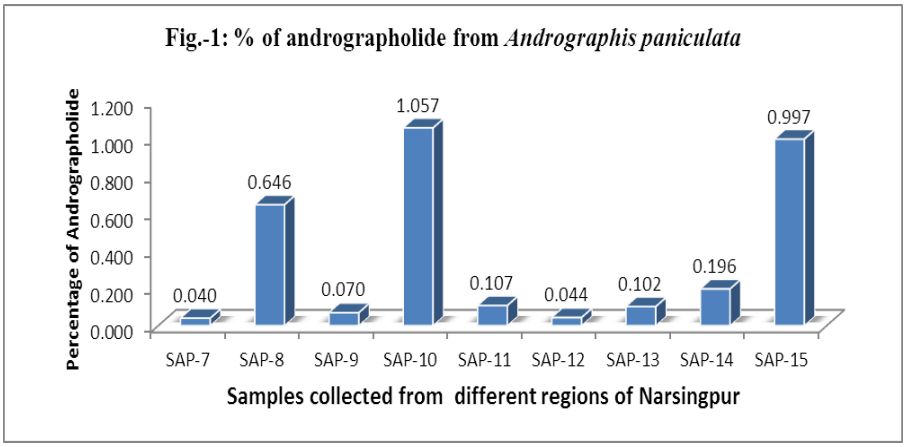
software, Column - Thermo C-18 column (4.6 x 250mm, 5µ particle size), sample size (20µl). Isocratic elution was carried out with methanol at a flow rate (1ml/min). The detection was performed with wavelength (230 nm) and column temperature was ambient (30^0C). Class VP software was used for integration and calibration. Evaluation was via peak areas with linear regression.

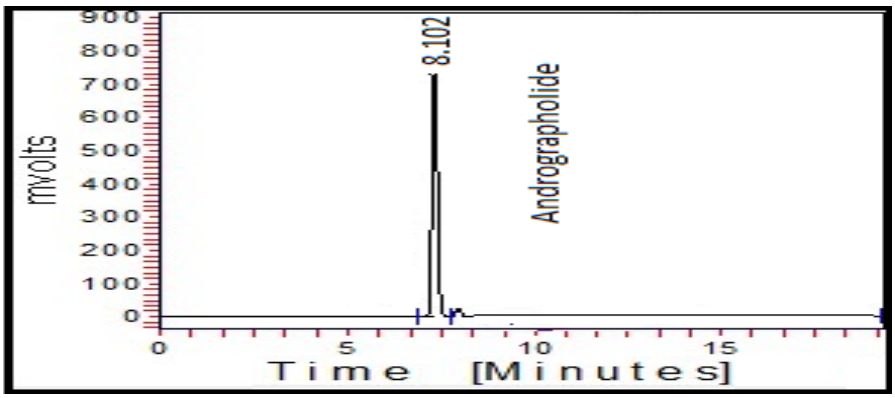
Preparation of herbal extract: Fresh aerial part of the plant sample collected from nine different sites of Narsingpur region. Reflux 1 gm dried powder along with 50 ml of methanol was kept in soxhlet for one hour. After one hour the refluxing load was subjected to Rota-vapor at 60 RPM and heated at 60^0C. Filter & subject the marc for another two cycles of refluxes (1 hrs. each) with methanol (50 ml) combine with the filtrate. Evaporate under vacuum to dryness Dissolve the residue 10 mg in methanol (10ml). Filter, Inject the solution in HPLC with the help of 20 µl fixed loop injector and percent content of andrographolide were estimated by counting the area of andrographolide peak in HPLC chromatogram in all sample.

RESULTS

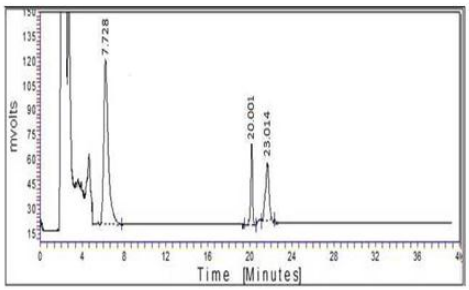
Table 1: Variations of andrographolide content from *andrographis paniculata*

Forest division	Forest range	Forest beats	Sample code number	Percent of andrographolide
Narsinghpur	Barman	Hirnapur	SAP-7	0.040
		Gorshi	SAP-8	0.646
			SAP-9	0.070
		Gudarai	SAP-10	**1.057**
	Gadarwara	Gotitoria	SAP-11	0.107
	Narsinghpur	Peeparpani	SAP-12	0.044
		Gangai	SAP-13	0.102
		Barpani	SAP-14	0.196
		Kishlai	SAP-15	0.997

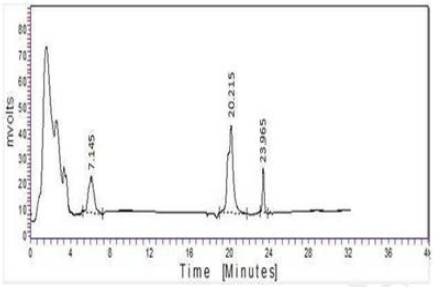




Graph. 1: Standard of andrographolide prepared by HPLC method.



Graph- 2: Maximum % andrographolide of SAP-10



Graph- 3: Minimum% andrographolide of SAP-07

Maximum % andrographolide of SAP-10			Minimum% andrographolide of SAP-07		
Peak name	**Retention time**	**Area**	**Peak name**	**Retention time**	**Area**
Andrographolide	7.728	5313.86	Andrographolide	7.145	201.657

DISCUSSION

The data pertaining to the percent concentration of andrographolide in plant samples of different locations from Narsingpur region of Madhya Pradesh were presented in Table 1. The data showed considerable variation in the concentration of andrographolide. Nine different samples were analysed from different sites of Narsingpur (Madhya Pradesh) region. Samples were analysed through HPLC method. Highest percentage of Andrographolide in *Andrographis paniculata* plant were from Gudarai site of Narsingpur that accounts to 1.057%, followed by Kishlai 0.997%, Gorshi 0.646%, Barpani 0.196%, Gotitoria 0.107%, Gangai 0.102%, Gorshi 0.070%, Peeperpani 0.044% and Hirnapur 0.040%. There was considerable variation in the percentage of andrographolide from one site to another site of Narsinpur region. Agroclimatic conditions also evident that there is considerable variation in the percent concentration of andrographolide. Considerable variation exists in the morphological traits in *Andrographis paniculata*. However information on the content of andrographolide from different sites of Narsingur is not available. Hence a study was under taken to evaluate the content of andrographolide and their considerable variation was analyzed by HPLC method.

CONCLUSION

In the present study, all the collected accessions were analyzed under controlled conditions and following variations were observed. Maximum andrographolide content was found in Gudarai site of Narsinpur and lowest was found in Hirnapur site. Documenting the biochemical variation is an efficient tool for identifying better accession could be used for better extraction of drugs from these medicinal valued plants.

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