



## Comparative Account of Amino Acids in Healthy as Well as Infected Leaves of *Tinospora Cordifolia* (Willd). Miers (Menispermaceae) by Using HPTLC

MISS V. S. BURE and A.U. PACHKHEDE

Department of Botany, Brijlal Biyani Science College, Amaravati, Maharashtra, India  
Corresponding author: burevaishali@gmail.com

### Introduction

*Tinospora cordifolia* (Willd.) Miers which is also known as Giloe belongs to the family Menispermaceae. It is an important medicinal plant used in Ayurved System of Medicine. Traditionally the plant has been in used as on anti-spasmodic, anti inflammatory, Janundice, diabetes, Seminal weakness, urinary tract infection, fever, general debility, skin diseases, and expectorant, carminative, digestive, antis tress and aphrordsisc. Piles problem can be controlled by eating this plant mixed with milk or water and thus, preventing the bleeding and constipation (Kritikar K. R. et al., 1987). Numerous constituents belonging to different classes such as carbohydrate, amino acids alkaloid, from different glycoside, Steroid have been isolated and characterised from different parts of *T. cordifolia*. Leaves are rich in protein calcium and phosphorus (Singh SS et al., 2003, Sinha K 2004)

Amino acids are organic substances containing both amino and acid group. They are building blocks of proteins and required by the human body. Essential aminoacids are not synthesised in the body and their exogenous supply is useful in a variety of metabolic activities. Weak immune system, nausea, dizziness, stomach problems and loss of antibody production are sometimes due to deficiency of amino acids. They play vital role in the metabolism of secondary metablites. They are used in industry Such as, in the synthesis of cosmetic and drugs and for the production of bio-degradable plastic. In the recent researches, HPTLC plays an important role in quantification of plant products because of its low cost, simplicity, rapidity and need of very small sample. For quantification of some amino acids, HPTLC method has been developed. The present investigation on *Tinospora cordifolia* (willd.) Miers provides valuble information about the presence or absence of amino acids that gives on indication of potential medicinal and nutritive value for human welfare.

### Materials and methods.

#### Collection of plant material

Plant materials i.e. healthy as well as infected leaves were collected from garden of Brijlal Biyani Science College, Amravati (M.S.). The Pathogens were isolated from infected leaves on Asthama and Hawker's medium 'A' and pathogensity test was carried out.

### Preparation of Extract

The dried and powdered plant material (200 mg) both healthy and infected leaves along with pathogens i.e. *Phoma sorghina* (Sacc. and *Colletotrichum capsici* (Syd.) was incubated for 24 hour at 4°C. The obtained supernatant was mixed again

The obtained supernatant was mixed again with 0.5 ml chlorform and 0.75 ml of Distilled water and centrifuged again. Supernatant so obtained was maintained at 38°C for about two hours and then subjected to freeze drying for further analysis.

### Mobile Phased used

Mobile Phases of different composition were tested for HPTLC analysis of amino acids. The desired objective was achieved by use of n Butanol: Acetic acid: Water (4: 1:1) for the separation of Arginine, Aspartick and, Serine, Threonine and tryptophan.

### Chromatography

TLC aluminum percolated plate with Silica gel 60 F 254 (6.0 x 10.0 cm) was used with mobile phase.

### Calibration of Curve of amino acids

A stock solution of standard amino acids (100µg/Ml) was prepared in distrilled water. The different volumes of the stock solution were spotted on the plate in triplicate to obtained concentration 400,600 and 800 hg / spot. Peaks of amino acids (standard and samples) were obtained ohen the plate was scanned at wavelength 350 nm, 400 nm and 450 nm



Fig.1

Healthy Leaves of  
*Tinospora cordifolia*



Fig.2

Infected leaves of  
*Tinospora cordifolia*

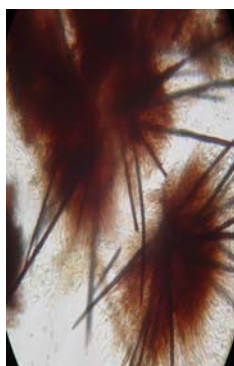


Fig.3

*Colletotrichum*  
*capsici*



Fig.4

*Phoma sorghina*

### Result and Discussion

In present investigation, We have reported five essential amino acids (Argenine, threonine, tryptophan, phenylalanine, histidine) and five non essential amino acids (Alanine, glycine, serine, glutamic acid.)are investigated. All the studied ten amino acids, the maximum essential amino acids was found in healthy leaves and minimum essential amino acids was found in infected leaves.

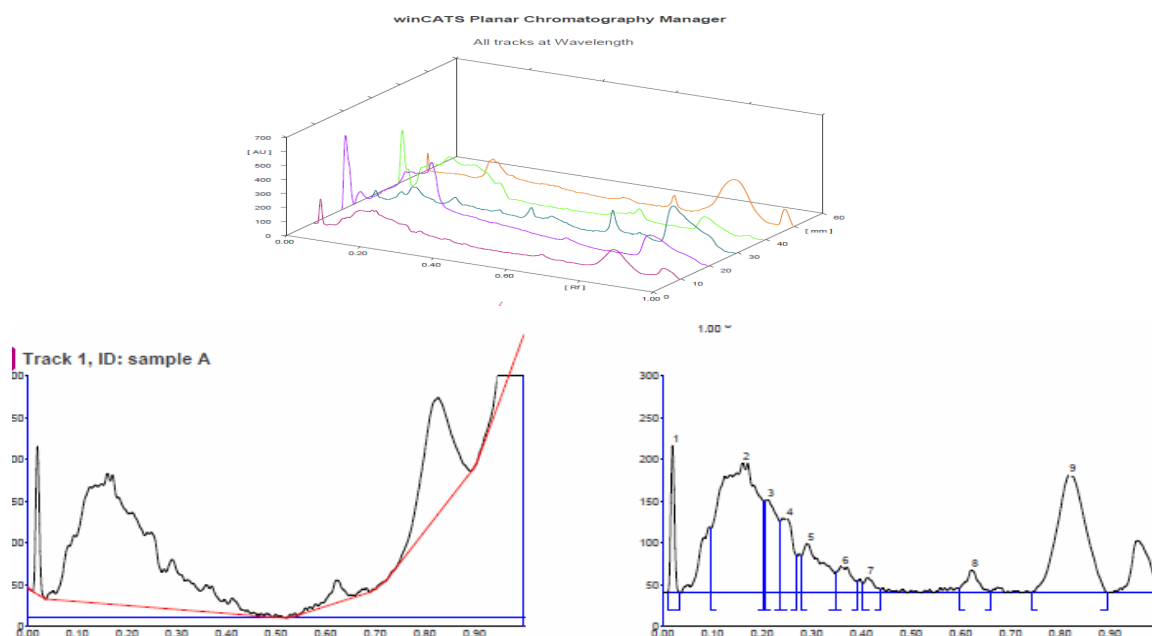


Fig.1 Chromatogram of healthy leaves of *Tinospora cordifolia*(Willd.)Miers

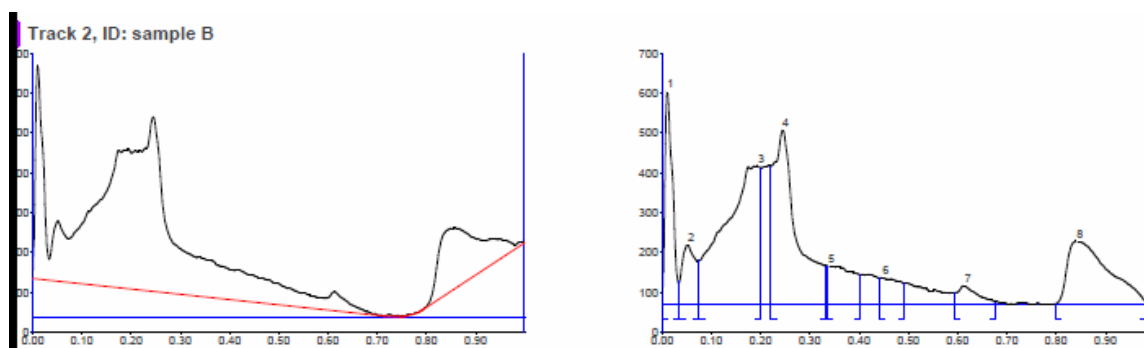


Fig.2 Chromatogram of dry infected leaves of *Tinospora cordifolia*(Willd.)Miers

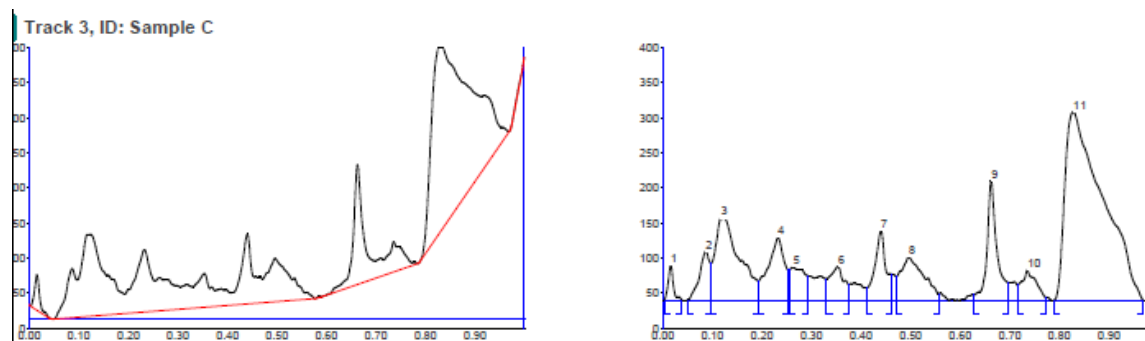


Fig.3 Chromatogram of healthy infected leaves of *Tinospora cordifolia* (Willd.)Miers

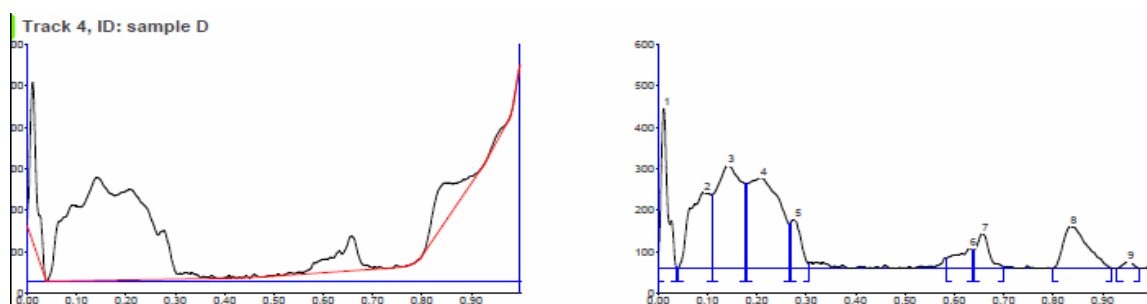


Fig. 4 chromatogram of *Phoma sorghina* (Sacc.)

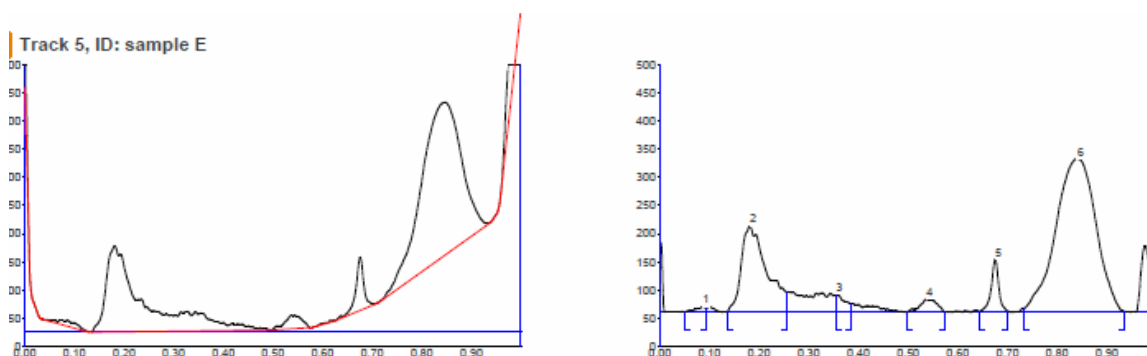


Fig.5 Chromatogram of *Colletotrichum capsici* (Syd.)

The pathogens i. e. *Phoma sorghina* (Sacc.) and *Colletotrichum capsici* (Syd.) lack the essential amino acids as they contain the only non essential amino acid. Arginine is involved in many metabolic processes and important in the treatment of heart diseases and high blood pressure. Arginine supports the growth of the osteoblasts which form the bone mass. Due to deficiency especially in older women, can be the cause of the emergence of osteoporosis.

Threonine – It supports the immune system by aiding in the production of antibodies. Due to deficiency it causes emotional agitation, confusion, digestion difficulties and fatty liver.

Tryptophan – it is needed for normal growth in infant and for nitrogen balance in adults.

## Conclusions

Arginine, threonine and tryptophan are essential amino acids for human so their presence in *Tinospora cordifolia* (Willd.) Miers indicates their use in Pharmaceutical industries. As the plant gets infected by pathogens they lose their property which was not useful for human beings.

A TLC method for the quantification of amino acids from healthy as well as infected leaves along with their pathogens like *Phoma sorghina* (Sacc) & *Colletotrichum capsici* (Syd.) using HPTLC was established. This method is simple, precise, rapid, accurate and economic and can also be used for the estimation of amino acid content for other medicinal plants.



## References

- [1]. Devi K., Kapila S and Rao A: High performance Thin layer chromatography (HPTLC) method for the determination of some Amino Acids from two species of *Marchantia*. Int J Pharm Sci Res 2014; 5 (12) : 5328 – 33, doi : 10. 13040 / IJPSR. 0975 – 823. 5 (12). 5328 – 33.
- [2]. Gaikwad S A, Kale A. Mundhe K. Deshpande NR and Salvekar J.P. : Detection of amino acids present in the leaves of *Cassia auriculata* L. International Journal of Pharm Tech Research 2010 ; 2 .1092 – 1094
- [3]. Hassan L G, Umar K4, Dangogo SM and Landan MJ: Protein and amino acid composition of African locust bean ( *Parkia biglobosa* L.) Journal of Tropical and Subtropical Agroecosystem S. 2005, 5 : 522 – 529
- [4]. Jana U, Chattopadhyay RN, Shw B.P. Preliminary studies on anti – Inflammatory activity of *Zingiber officinale* Rosc., *Vitex negundo* Linn. and *Tinospora cordifolia* (Willd.) Miers in albino rat India J Pharmacol., 31:232–3.
- [5]. Kirtikar K. R, Basu B.D. 1987. Indian Medicinal Plants, Vol . 1, International Book Distributors, Dehradun, India, pp 77-80
- [6]. Mehrotra R., Katiyar CK, Gupta A.P. 1999. Hepato-protective compositions and composition for treatment of conditions related to hepatitis B and E infection U.S. Patent 749296;
- [7]. Singh S S Pandey SC Srivastava S. Gupta VS, Patro B. Ghosh A. C. 2003 Chemistry and medicinal properties of *Tinospora cordifolia* (Guduchi) Indian Journal of Pharmacology ; 35 : 83 - 91
- [8]. Swati M.D, Gayatri S. K. Rasika CT, Asha AK, Nirmala RD and Joyti PS Preliminary Screening of amino acids from a medicinal plant: *Morinda pubescence*. Journal of Chemical and Pharmaceutical Research 2011 ; 3 : 192 – 195
- [9]. Williams, J. Z., Abumard, N.and Barbul, A (2002). Effect of a specialized Amino Acid mixture on human collagen Deposition, Annals of surgery, volume 236, issues, ( P.P. 369 – 375)
- [10]. Young V. R: Adult amino and requirements the case for a major revision in current recommendations. Journal of Nutrition 1994 ; 124 – 511 7S – 1523