

AMINO ACID ANALYSIS OF INTELLAN, A HERBAL PRODUCT USED IN ENHANCING BRAIN FUNCTION

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ABSTRACT:

Analysis of the drug "*INTELLAN*" and two reputed plants i.e. *Centella asiatica* and *Herpestis monniera* used in the preparation of the drug reveals that glutamic and aspartic acids are present in high concentration in the drug and are also required by brain in high concentration to keep itself in higher gears. Both these amino acids are used as neurotransmitters and also in stabilizing and stimulating the activities of brain, thus resulting in better performance.

Introduction

Mental disorders/diseases are of growing importance in the future health programme of the world. Morbidity due to mental disorders and diseases are very high day by day. Furthermore, the monopolar depression and bipolar effective disorders are highly recurrent (Angst 1976).

"*INTELLAN*" a herbal product is a compound formulation, prepared by Messers Herbion Pakistan (Pvt) Ltd. According to the manufacturers, the product is used to enhance and improve the mental performance, memory function, alertness, allay anxiety and depression. It also stimulates metabolism of cerebal cortex and activates neurotransmitting centres (Zafar 1992). Each capsule is based on the aqueous extract of organic and inorganic compounds from *Centella asiatica* Linn, *Herpestis monniera* H.B.K., *Corriander sativum* Linn., *Amomum aromaticum*, Roxb., *Emblica officinalis*, Linn. and *glycyrrhiza glabra*, Linn.

Folklore Literature reveals that these plants are used as tonic, (nerve, brain, cardiac, restorative, nutrient), to allay pain (cephalgic, neuralgic, muscular and headache) and also in cases of insanity. (Nadkarni 1954), Chopra 1980, ICR 1987). The plants *Centella asiatica* and *Herpestis monniera*, used in the preparation of the drug, i.e. "*INTELLAN*", have a well documented record of improving general ability, behavioral pattern, increasing intelligence quotient and improving impaired memory function in mentally retarded children (Ganguly 1969, Appa 1973). Chemical investigations of the plant reveal

the presence of bioactive alkaloids, glycosides, tannins, saponins, fatty acids, essential oils, phenolic compounds resins, vitamins especially Vit. C and amino acids (Dhar 1956, Dutta 1962, Singh 1968, Al-Saadi, 1984, Zamarish 1986, Mushtaq 1987).

High claims have been made by the manufacturer regarding the therapeutic efficacy of the drug i.e. "INTELLAN", which is based on the amino acid contents. The present work is an attempt to identify amino acids present in the drug and in two main plants i.e. *Centella asiatica* and *Herpestis monniera* and to correlate the role of amino acids as neuroenergizer.

Material and Method

The material i.e. drug and the plants were obtained from Messers nation Pakistan (Pvt) Ltd, the manufacturer, in the form of coarse dried powder.

Protein estimation

Protein estimation of the drug was done by Microkjeldahls method, as described in the method of AOAC 1984.

Amino acid analysis

Amino acids were analysed both by quantitative and qualitative method (Black-burn 1968), i.e. the plants and the drug respectively.

Preparation of the sample for amino acid analysis (Hydrolysis)

250 gms of drug (INTELLAN) and dried powdered plant material of *Centella asiatica* Linn and *Herpestis monniera* H.B.K respectively were taken in individual flasks, hydrolysed by 5.7N HCl. and freed from acid. The acid freed samples were then subjected to Dowex 50 column (10 x 1.2 Cms) and eluted with 10% ammonia. The eluent were then evaporated to dryness and taken in 10.5 ml of distilled water, 50.0/ul of the solution from each flask was then lyophilized and analysed for their amino-acids content quantitatively and qualitatively.

Quantitative analysis

Plants, i.e. *Centella asiatica* and *Herpestis monniera* were analysed by means of amino acids analyzer and amino acids analysed were expressed as mg/100 gms of dried plants material.

The quantitative aminoacid analysis was carried out on Biochrome automatic amino acid analyser by cation exchange chromatography process using speckman 1958 method.

Estimation/analysis of each amino acid was performed by determining the area under each peak on a chart recorder. The instrument was calibrated first by analysing the standard mixture of known amount of amino acids.

Qualitative analysis

The hydrolysed sample of the drug was analysed by means of paper chromatography, using Butanol : Acetic acid : Water (63:37:10) as solvent and nin-hydrin as spray reagent. Authentic amino acids as standard were also run with the sample.

Results

Protein

Approximately 11.0% was found to be present in the drug.

Amino acid analysis

1/- amino acids (Aspartic acid, Glutamic acid, γ -amino butyric acid, Leucine, Isoleucine, Valine, Methionine, Arginine, Lysine, Histidine, Proline, Phenylalanine, Tyrosine, Serine, Alanine, Threonine, and Glycine) were detected qualitatively, while quantitative analysis reveals the presence of 14 amino acids in *Centella asiatica* and 15 in *Herpestis monniera*. γ -aminobutyric acid and arginine were absent from both the plants and serine from *Centella asiatica* only.

Glutamic and Aspartic acids were present in higher concentration in both plants, (Table-I). The difference in the number of the amino acid contents exhibited by the drug and plants was due to the presence of other plants used in the preparation of the drug. Furthermore quantitative analysis of the plants reveals that *Herpestis monniera* is richer in amino acid contents as compared to *Centella asiatica*.

Discussion

Folklore reputed plants i.e. *Centella asiatica* and *Herpestis monniera* used as a neuroenergizer and brain tonic (Nadkarni 1954, Chopra 1980 & ICR 1987), exhibited a rich profile of amino acid contents both in quality and quantity. It was also observed that *H. monniera* exhibited a maximum quantity of amino acid contents as compared to *C. asiatica*. Furthermore the amino acid profile exhibited by these plants shown a good correlation with the requirement of amino acids for brain (Ansell et al, 1954). Comparative study of the both plants with plants not used as a brain energizer showed no correlation (Table-I). The minor and the main amino acids i.e. glutamic and aspartic acids responsible for the activation of brain were found to be totally absent from *Achyranthus biodenta* (Bisht et al., 1990), and in a very low quantity in *cucumis melo* (Ikram et al., 1978). (Table-1).

Literature survey has revealed that the therapeutic efficacy of the herbal products/drugs are not only based on the chemical and proximal composition but also on the protein and amino acid content. An ample data is available which depicts that proteins and amino acid are used in the therapy of many diseased conditions of mental disorders (Sourkes 1969, Andress 1978, James 1980, Meldrum 1987). A short disturbance in their composition and amount may alter or abolish the physiological and pharmacological activities of the body (West, 1984).

The drug "*INTELLAN*", contains 11.0% protein. Thus presence of protein in the drug tries to provide not only the amino acid units needed for the synthesis of new tissues but also compensates the system which results in loss of weight, marked weakness, strain, stress, mental emotional changes (Keele 1961, Chatterjee 1977). Amino acid analysis of the two plants correlates with the finding of Zamarish and Mushtaq, which reveals the presence of essential key amino acids (EAA) as well as non-essential amino acids (NEAA). It has been an established fact, that amino acids, specially the NEAA along with other chemical mediators, act as neurotransmitters within the central nervous system (CNS) involved in memory and learning abilities of brain (Olten 1988, Rairorodsky, 1990). In the CNS, amino acids act either by excitation, inhibition or by depressant action. Glutamic and aspartic acids are the only NEAA, which under certain conditions become contingency nutrient and thus essential, and are used in higher concentration, to stimulate and to stabilize the brain activities (Leon, 1986). Literature citation reveals that deficiency of amino acids specially glutamic and aspartic acids lead to decreased brain function, including number of abnormalities, such as impaired memory function, intelligence quotient and mental performance, abnormal behavioural pattern, autism, lack of concentration and decreased cognitive ability (Sourkes 1968, James, 1980, Leon 1986). The characteristic requirements of amino acids for brain reveal that glutamic and aspartic acids required in higher concentration to keep the brain in proper functioning order (Ansel) 1954, Sourkes 1968). Glutamic acid is also known as "Brain fuel" (Leon 1986). It is the only amino acid which can easily pass the blood brain barrier and can stimulate the cell in the CNS by increased membrane permeability in the same way as does an excitatory impulse (Streit, 1988). Furthermore these two amino acids i.e. glutamic and aspartic acids also take part and act as donor and precursor for the synthesis of other amino acids which are required for the proper functioning and improving the mental abilities of the brain. Glutamic and aspartic acids take part in transamination reaction. Both these amino acids combine with ammonia and get converted into glutamic and asparagine respectively. Glutamine is a major excitatory neurotransmitter in the brain and spinal cord (Goodman 1991). Glutamine acts as a detoxifier of ammonia for the brain and along with glucose it acts as a fuel for brain cells, which results in the improvement of the nutritional status of numerous nerve cells and removal of toxic metabolites resulting in enhanced functioning of the brain. While asparagine takes part in

the metabolic control of the brain and nervous system (Chatterjee 1977, James 1980). Glutamic and aspartic acids act on the excitatory receptors in the hippocampus and thereby enhance the discharge of neural pathways. These receptors seem to be involved in memory and learning ability of the brain (Olten 1988, Rairorodsky 1990). Glutamate and aspartate are also present at intraneurons at all levels, increase the cationic conductance and depolarize sulphur rich amino acids which are further utilized in the formation of number of essential compounds such as Co-enzyme-A, Heparin, biotin, Lipoic acid and glutathione (Sourkes 1968, West 1984, Leon 1986). Amino acids act directly on the receptors of the myocardium, cells of the pace makers and conducting tissues which results in increase of contractile force, enhances the rate of relaxation and decreases the time of peak tension. Increased excitation enhance the rate of O₂ consumption, spontaneous beating and induction of automatically in quiescent muscles. Thus resulting in strengthening of stamina and endurance (Ganong 1989).

It has been amply proved that the excitatory amino acids have many functional applications. Therapeutically they are used in the treatment of nutritional conditions (James 1980, Leon 1986), to improve negative nitrogen balance, loss of appetite, fatigue, nervous irritability, (Aley 1989), despaired behaviour, mental apathy, depressive anxiety state (Olten 1988, Tanaka 1988), sub-optimal development of brain, anorexia, ataxia, nervosa, analgesia (Ninomiya 1990), to enhance intelligence quotient in mentally retarded children (Leon 1986, Olten 1988), on cases of epilepsy (Meldrum 1987), schizophrenia (Deakin 1989), senility (Murayamakoichiro 1990), in controlling cerebrovascular tone and in regulating locomotor activity (Shreve 1981).

Analysis of the drug "INTELLAN" and individual plants i.e. *Centella asiatica* and *Herpestis momiera* reveal that glutamic acid and aspartic acids are present in high concentration in the drug and are also required by brain in high concentration to keep himself in higher gears. Both these amino acids have a well documented record of their use as neurotransmitter and also in stabilizing and stimulating the effect of metabolism of cerebral cortex, thus resulting in better performance (Sourkes 1968, Leon 1986). Furthermore, as supported by the literature that protein and amino-acids are used in the treatment of many diseased conditions and disorders of brain and central nervous system.

TABLE I
Amino acid detected qualitatively, quantitatively and comparative study, with other plants not used as neuro energizer

S.No.	Amino acid Estimated	Qualitative analysis of drug (Intellan)	Quantitative analysis mg/100 gms		S.D. $\pm$	Achyranthus biodenta ta mg/ 100 mgs	Cucumis melo mg/100 gms
			C. Asiatica Mean	H. monniera Mean			
1.	Aspartic acid	+	3.37	*13.7	0.03	0.05	--
2.	Glutamic acid	+	2.18	11.13	0.01	0.92	--
3.	γ -Aminobutyric acid	+	-	-	-	-	--
4.	Leucine	+	0.50	5.0	0.01	-	4.9
5.	Iso-leucine	+	2.00	2.29	0.01	-	4.2
6.	Valine	+	1.21	4.62	0.02	0.65	6.5
7.	Methionine	+	0.43	0.63	0.02	-	2.9
8.	Arginine	+	-	-	-	0.72	--
9.	Lysine	+	2.22	7.29	0.01	-	3.4
10.	Histidine	+	0.45	2.47	0.02	-	1.8
11.	Proline	+	-	3.64	0.01	0.86	2.3
12.	Tryptophane	-	-	-	-	0.76	1.0
13.	Tyrosine	+	0.35	6.93	0.01	0.82	5.6
14.	Phenylalanine	+	0.29	2.69	0.01	0.96	9.9-10.5
15.	Serine	+	-	5.35	0.01	0.17	-
16.	Alanine	+	0.92	6.82	0.02	-	-
17.	Threonine	+	0.43	5.24	0.01	0.53	1.9
18.	Glycine	+	0.82	6.13	0.01	0.72	-

*Value estimated are average of three readings.

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