RESEARCH PAPER

Development and Standardization of a Polyherbal Formulation Containing Ashwagandha, Brahmi and Curcumin for Cognitive Dysfunction in Alzheimer's Disease

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ABSTRACT

Cognitive decline is acknowledged as a leading global health concern and is often connected to several brain-degenerative disorders such as epilepsy and Parkinson's disease, as well as aging, hypertension and increased chronic stress. Alzheimer's disease (AD) is the main focus of this review because it is the major contributor to the loss of mental function in these diseases. The illness is characterized by a progressive deterioration of language and memory skills accompanied by a variety of behavioral and psychological symptoms including emotional discomfort, rage, anxiety, mood instability, and psychotic symptoms.The illness is a progressively worsening neurological condition that is impacted by a number of variables and causes observable changes in personality and general mental capacities.A growing body of research suggests that a variety of factors, including genetic predisposition, environmental exposure, dietary habits, and lifestyle patterns, contribute to the development and progression of AD, even if the exact etiology of the condition is still unknown..Over the past few decades, numerous drug candidates have been evaluated; however, a large proportion have failed in clinical trials, reflecting the complexity of the disease. Recent research efforts have focused on the discovery of novel biomarkers, improved cognitive and behavioural science assessment tools, early-stage patient inclusion, and innovative clinical trial methodologies to enhance the effectiveness of future therapeutic interventions.The present Research focuses on the formulation and evaluation aspects of a polyherbal syrup used in the treatment of Alzheimer disease. Polyherbal formulations work on the principle of synergy, where different target multiple disease pathways together.various medicinal plants such as Withania somnifera (Ashwagandha), Bacopa monnieri(Brahmi), Curcuma longa (Curcumin) have been reported to possess neuroprotective, antioxidant,anti-inflammatoryand memory–enhancing properties.

KEYWORDS: Alzheimer disease, Neuroprotection, Herbal medicine, memory, cognitive impairment, Ashwagandha, Brahmi,Curcumin.

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INTRODUCTION

Alzheimer disease: -

Alzheimer disease is a progressive gradually worsening brain condition which slowly impairs memory, thinking capacity, and routine activities, making it the most common cause of dementia worldwide. The disease significantly interferes with daily activities and quality of life, ultimately leading to complete dependence on caregivers. Dementia is a long-term condition characterized by a gradual decline in cognitive functions that goes beyond the normal changes seen with aging[1].Nearly 50 different brain condition are known to contribute to the development of dementia, among which alzheimers disease is the leading cause, responsible for nearly 50-70% of all cases. the global population is expected to reach approximately 11.2 billion people by 2100 due to improvements in health indices and longer life expectancies brought about by advancements in medical sciences and healthcare. Forecasts indicate that by 2050, over 21% of the population would be over 60, creating a sizable old population of two billion. Illnesses associated with aging ,lncluding Alzheimer's disease (AD), have increased in frequency as the number of senior people rises. Memory loss, cognitive decline, and challenges with everyday tasks are the hallmarks of AD the most prevalent kind of dementia, a progressive and incurable neurological disease. Although the precise origin of AD is yet unknown, genetic factors are thought to be important. A smaller cerebral cortex and larger brain ventricles are the pathological results of neurofibrillary tangles and amyloid plaques spreading throughout the brain, which interferes with neuronal communication and causes nerve cell death [2].

Age, genetics, family history, smoking, drinking, and other variables are among the many risk factors for AD. Poor decision-making, inability to think clearly, misplacing items, difficulty moving, poor verbal

communication, strange emotions, and total memory loss are all signs of AD. According to a study by the Alzheimer's Association research group, 66% of people with dementia have AD, and only 10% receive a timely diagnosis. The remaining 90% receive no diagnosis at all. Government records show that 121,404 deaths in the US in 2017 were officially attributed to AD. AD is predicted to affect 60 million people globally in the next 20 years. According to the World Alzheimer's Report, there will be 152 million Alzheimer's sufferers globally by 2050[3]

Uses of Ashwagandha, Brahmi and curcumin as Alzheimer'sdisease:-

In Recent Years, Well-known medicinal plants including ashwagandha, Brahmi, and curcumin have demonstrated promise in Alzheimer's disease therapy. These herbs' neuroprotective, anti-inflammatory, and antioxidant properties may enhance memory andcognitive function. In those suffering from Alzheimer's, their combined use is thought to be a potential approach for lowering neuronal harm and promoting brain general condition[4].

2.1 Ashwagandha (Withania somnifera)

Ashwagandha is frequently used herbal plant in traditional medication. It is known because of its adaptogenic, neuroprotective, and antioxidant properties. Traditionally, it was used to promote memory, cognitive functions, and additionally brain health. The active constituents, mainly withanolides and alkaloids, are responsiblefor its advantages in treating neurological conditions, including cognitive disorder. The plant is frequently identified to as "Indian Ginseng" or "Indian Winter Cherry." It is widely recognized as a Rasayana in traditional ayurvedic practice due to its numerous well-being advantages, consequently, it holds a prominent position in ayurvedic practice, India's traditional healthcare system. A herbal or metallic mixture that promotes happiness and a youthful state of bodily and mental well-being is known as rasayana. These kinds of therapies are administered as supportive treatments to young children and consumed by middle adulthood and olderto support a longer life. Ashwagandha is the crucial of the Ayurvedic rejuvenating herbs. "Sattvic Kapha Rasayana" Herb is the name given to it. Most Rasayana herbs are stress-management and adaptogenic [5].

It has been used in traditional Indian medicine for almost three millennia. Its root has been used as a stimulant, tonic, anthelmintic, diuretic, narcotic, and aphrodisiac. Although it is native to India, Additionally, it is grown in Africa, the Canary Islands, the Cape of Good Hope, Australia, the Mediterranean region, and the Himalayan regions. Ashwagandha's potential health benefits have drawn more attention in recent years, especially in relation to stress reduction, cognitive function, and physical performance. According to multiple studies, ashwagandha supplements may help with obsessive-compulsive disorder and have neuroprotective, antibacterial, immunomodulatory, and inflammation-reducing effects [6].

There is also evidence that ashwagandha supplements help treat cancer, diabetes and infertility. While ashwagandha has a wide range of possible health benefits, further research is needed to better understand how it works and to evaluate its effectiveness in treating different health conditions. Research has shown that ashwagandha has cardioprotective qualities. It may also help sleep difficulties, increase stress resilience, decrease anxiety, and cure hypothyroidism. It may also help with muscle strength and recovery. Hence, the present study aims to examine the recent investigations on the activity of ashwagandha, focusing on the possible benefits for mental clarity, physical performance and stress reduction [7].

2.2 Brahmi (Bacopamonnieri)

Bacopa Monnieri or Brahmi is a popular herb in the medicinal world that has been used for a long time to improve memory and enhance cognitive function. The plant contains various phytoconstituents as Bacosides, Flavonoids, Saponins, Alkaloids and Sterols which contribute to its medicinal benefits. The main active ingredients responsible for the plant's neuroprotective, antioxidant, anti-inflammatory and memory boosting properties are believed to be bacosides. These pharmacological qualities make Bacopa monnieri widely utilized in the management of neurological illnesses including Alzheimer's disease [8].

The aging world population is leading to an increase in the prevalence of neurodegenerative diseases and, therefore, in the demand for suitable therapeutic approaches. Bacopa monnieri (BM) is a medicinal herb that could be used as a natural agent for brain health containing various bioactive compounds including bacosides A and B, quercetin, betulinic acid, asiatic acid and loliolide. This chemical has shown neuroprotective potential, however, its therapeutic importance in neurodegenerative diseases is poorly studied. BM has been demonstrated to be helpful for neuron repair, kinase activity, synaptic function, neural transmission and neuroprotection in studies. It also demonstrates antioxidant and antiapoptotic properties and decreases phosphorylation of Nuclear Factor-κB (NF-κB) and oxidative stress. Evidence from 22 clinical trials shows that BM enhances cognitive function, emotional well-being, memory retention, attentiveness, learning capacity, sleep quality, sorrow, impulsivity, hyperactivity, anhedonia and other psychiatric problems. Moreover, BM lowers the levels of pro-inflammatory markers and oxidative damage, indicating that it may be a possible adjuvant therapy for neurodegenerative illnesses. In conclusion, this review is devoted to the therapeutic potential of BM and its contribution to the development of new, natural therapies for neurodegenerative disorders of the brain and our understanding of the mechanisms of cognition and neurodegeneration [9].

Additionally, Brahmi is used to treat mental disease, epilepsy, joint pain, backache, hoarseness, and issues with both male and female sexual performance. It can

also be used as a "water pill. "Certain brain chemicals involved in thinking, learning, and memory may be elevated by Brahmi. According to some research, it may also shield brain tissue against substances linked to Alzheimer's [10].

2.3 C urcumin (curcuma longa)

Turmeric (Curcuma longa), belonging to the Zingiberaceae (ginger) family, has a long history of use in traditional Indian medicine for a wide range of therapeutic applications. The medically essential element of the plant is the underground rhizome, which supplies a valuable source of curcumin, a naturally occurring polyphenolic compound. Turmeric is a ubiquitous dietary spice but has also been used in traditional medicine as a natural dye and cosmetic and as a herbal remedy for inflammatory disorders, gastrointestinal disease, liver disease, infections and blood abnormalities. Its various biological qualities have been confirmed by current pharmacological research, including antioxidant, anti-inflammatory, antibacterial, hepatoprotective and anticancer activity. It has attracted a lot of attention in neuroscience research recently due to its ability to protect neuronal cells from harm . Experimental investigations have revealed that curcumin inhibits the oxidative damage, inhibits the amyloid-β aggregation and modulates the cellular signaling pathways that are important in neuronal survival thereby counteracting the numerous degenerative processes associated with dementia. In preclinical models of Alzheimer's disease, these activities have been associated with improvements in cognitive performance, memory retention and learning ability, suggesting curcumin as a potential candidate for the prevention and therapy of neurodegenerative diseases [11].

Curcumin has been attracted tremendous attention among phytochemicals for neurodegenerative disorders, as it affects on many pathogenic processes simultaneously. It is a naturally occurring substance, derived from the rhizome of Curcuma longa, a medicinal plant that has been utilized for ages in Asian health practices and food preparations. The pleiotropic effects of curcumin on oxidative stress, inflammatory signaling, immunological responses and neuronal survival imply its importance for disorders like Alzheimer's disease, Parkinson's disease, Huntington's disease, amyotrophic lateral sclerosis and prion-related neurodegeneration. Experimental studies have also shown that curcumin can inhibit protein aggregation and improve neuronal function, resulting in improved cognitive performance in preclinical studies. However, these interesting biological activities have not been converted into clinical therapy due to limited systemic availability of the molecule after oral administration. Rapid metabolism, poor water solubility, physiological instability and poor ability to cross blood–brain barrier of curcumin severely limit its distribution to neuronal sites. Therefore, its administration has been an important area of current research through enhanced formulations to improve its therapeutic efficacy against neurodegenerative diseases [12].

MATERIAL AND METHODS

The present study was carried out in the Pharmaceutics and Pharmacology Laboratory of Rungta Institute of Pharmaceutical Sciences and Research, Bhilai, Chhattisgarh, India.Using traditional pharmaceutical techniques, the Polyherbal formulation containing Ashwagandha, Brahmi, and Curcumin was developed and standardized in a lab setting.

3.1 Making a Polyherbal Syrup Using Ashwagandha, Brahmi and Curcumin:

Ashwagandha, Brahmi, and Curcumin extracts were added to an appropriate syrup foundation to create the polyherbal syrup. Purified water was combined with the necessary amount of materials after careful weighing and continuous stirring. To the herbal mixture, sucrose syrup was made separately and added gradually.Stirring the mixture resulted in a homogenous syrup. After adjusting the final volume with distilled water, it was filtered and put into sterile, airtight vials for additional analysis [13].

Tools:UV-visible spectrophotometer, Ostwald viscometer, glass rod, funnel, measuring cylinder, heating mantle, grinder, beaker, digital weighing balance, and specific gravity bottle.

Collection of Ingredients: We purchased curcumin, ashwagandha, and Brahmi from a local herbal vendor in Bhilai, Chhattisgarh. Every chemical and reagent used in the investigation was of analytical quality and came from reliable commercial vendors.

3.2 Step by Step Process for Preparation of Polyherbal Syrup

1.Ashwagandha, Brahmi, and curcumin were collected, dried, and ground separately.

2. The resulting extracts were filtered and concentrated following the ethanol extraction of the powdered constituents.

3. The concentrated extracts were carefully weighed and then kept for further use.

4. The syrup base was prepared with the appropriate ingredients and continuous stirring.

5. The extracts of curcumin, Brahmi, and ashwagandha were added to the syrup base and well mixed to produce a uniform formulation.

6. The proper excipients, including flavorings, pH adjusters, and preservatives, were added as needed.

7. The formulation was thoroughly blended and its final volume was adjusted using distilled water.

8. The resulting Polyherbal syrup was filtered and placed in sanitary, airtight containers for further evaluation and standardization[14].

Table.1 Ingredients of products

S.No.	**Ingredients**	**Amount (Quntity)**

1.	Dried Ashwagandha powder	50g
2.	Dried Brahmi powder	50g
3.	Curcumin powder	25g
4.	Sucrose syrup	60ml
5.	Glycerine	5ml
6.	Methyl paraben	0.1gm
7.	Propyl paraben	0.05gm
8.	Ethanol	250ml
9.	Flavouring agent	1 ml
10.	Distilled water	Q.s. to make 100ml
11.	Experimental Animal	8
12.	Alcl3	0.8 mg/kg
13.	donepezil	2.5 mg/kg

3.3 Packaging and Storage

To avoid contamination, the prepared Polyherbal syrup was put into sterile amber-colored bottles and carefully sealed. The formulation was stored in a cool, dry location at room temperature and out of direct sunlight until further evaluation.

EVALUATION OF POLYHERBAL SYRUP:

4.1Organoleptic Evaluation

The prepared polyherbal syrup was evaluated in terms of its color, flavor, smell, and look. The syrup had a pleasant smell, a consistent appearance, and a unique flavor.

4.2 Calculating pH

A digital pH meter was used to measure the syrup's pH to ensure it was stable and appropriate for oral use.

4.3 Viscosity Determination

Viscosity was measured with a viscometer to evaluate the syrup's pourability and flow properties.

4.4 Density Determination

To assess the physical characteristics of the syrup, the formulation's density was measured using a specific gravity bottle.

4.5 UV Spectrophotometric Analysis

Using UV spectroscopy, the phytoconstituents in the formulation were determined and quantified.

4.6 FTIR Analysis

FTIR analysis was used to identify the functional groups and possible interactions among herbal constituents.

4.7 In Vivo Assessment of Polyherbal Formulation

Swiss wistar rat were used for the in vivo assessment of the prepared Polyherbal syrup. The animals had unrestricted access to food and water and they kept in typical laboratory settings. The animals were split up into four groups, and the study methodology was used to evaluate the formulation's impact.

Table 8 :- Experimental Design and Treatment Schedule for In Vivo Study

Group	**Treatment**
Group I	Normal control group
Group II	Disease control group (aluminium chloride)
Group III	Disease+ treatment group (aluminium chloride + polyherbal syrup)
Group IV	Standard drug group

Fig5.10:- Initial Body Weight of Experimental animals

Fig 5.11-Grouping of Animals

4.10 Oral Administration of Aluminium chloride experimental animals

4.11 Administration of Polyherbalto Formulation

Fig .5.14-Elevated Zero Maze Test Showing Behavioural Response of Animal

Fig .5.15- Elevated Plus Maze Test Showing Behavioural Response of animal

RESULT

5.1 Organoleptic evaluation

Table 1. Organoleptic evaluation of Polyherbal syrup

Parameter	**Observation**
Color	Yellowish
Odor	Characteristic
Taste	Sweet
Appearance	Clear

5.2 pH determination

Table.2.pH of Polyherbal syrup

Parameter	**Result**
pH	5.38

5.3 Density/Specific Gravity

Table.5. Density of Polyherbal syrup

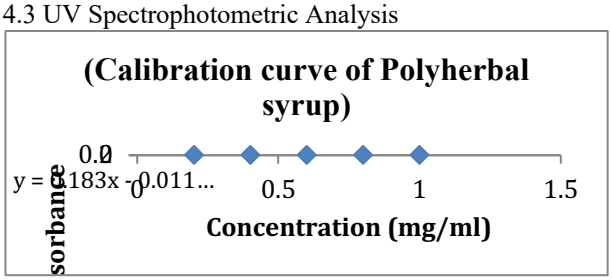
Parameter	Value
Density (g/mL)	1.20g/ml

5.4Viscosity Determination

Table.6.viscosity of Polyherbal syrup

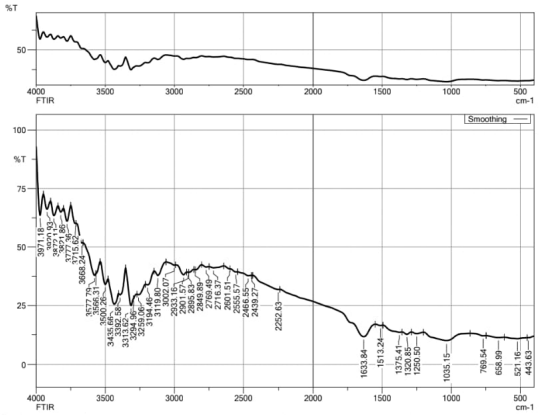
Sample	Viscosity (cP)
Polyherbal syrup	3.34 cP

4.3 UV Spectrophotometric Analysis



Graph 1 : Calibration curve of polyherbal syrup

4.6 FTIR Analysis



	Peak	Intensity	Corr. Intensity	Base (H)	Base (L)	Area	Corr. Area	Comment
1	443.63	11.24	0.10	450.81	400.56	4447.639	4.367	
2	521.16	10.81	0.51	617.36	450.81	14808.781	41.482	
3	658.99	11.30	0.35	748.01	617.36	11552.841	26.176	
4	769.54	12.07	0.25	864.30	748.01	10162.693	0.638	
5	1035.15	10.00	3.33	1201.69	864.30	29804.261	563.907	
6	1250.50	12.97	0.66	1287.83	1201.69	7471.051	30.407	
7	1320.85	12.69	0.99	1356.75	1287.83	5980.896	32.291	
8	1375.41	13.56	0.52	1494.58	1356.75	11783.099	87.305	
9	1513.24	16.46	0.27	1550.57	1494.58	4666.289	7.372	
10	1633.84	11.68	7.11	2241.14	1550.57	53080.102	905.161	
11	2252.63	31.86	0.40	2433.53	2241.14	12662.263	138.330	
12	2439.27	37.88	0.01	2442.14	2433.53	535.034	0.028	
13	2466.55	37.75	0.50	2542.64	2442.14	6183.224	17.520	
14	2555.57	39.36	0.36	2597.20	2542.64	3280.280	10.107	
15	2601.51	40.70	0.13	2641.71	2597.20	2614.686	3.593	
16	2716.37	41.10	0.66	2749.39	2641.71	6308.817	44.023	
17	2769.49	41.44	0.53	2802.51	2749.39	3093.264	15.513	
18	2849.89	40.32	0.03	2858.50	2847.01	685.000	0.151	
19	2895.83	39.20	0.05	2897.26	2858.50	2335.007	1.920	
20	2901.57	39.19	0.05	2911.62	2897.26	872.381	0.388	
21	2933.16	38.13	2.01	2992.02	2911.62	4816.152	59.531	
22	3002.07	42.25	0.29	3060.94	2992.02	3930.788	3.368	
23	3119.80	37.88	3.06	3147.08	3060.94	5138.080	113.758	
24	3194.46	33.82	1.65	3210.25	3147.08	4055.797	68.316	
25	3259.06	29.62	1.03	3270.55	3210.25	4148.834	46.013	
26	3294.96	27.90	0.15	3296.39	3270.55	1830.395	-7.060	
27	3313.62	24.97	6.92	3353.82	3296.39	3978.243	218.158	
28	3392.58	29.59	3.05	3404.07	3353.82	3386.790	151.423	
29	3435.66	25.54	7.17	3480.16	3404.07	5397.202	316.842	
30	3500.26	33.95	4.99	3537.59	3480.16	3590.479	147.518	
31	3566.31	38.67	0.25	3567.74	3537.59	1783.565	11.386	
32	3577.79	37.94	2.16	3661.06	3567.74	5144.033	35.830	
33	3668.24	51.72	1.30	3711.31	3661.06	2238.230	27.266	
34	3715.62	60.08	0.93	3750.08	3711.31	1437.679	42.181	
35	3777.36	61.08	6.08	3803.20	3750.08	1913.638	170.482	
36	3821.86	64.90	2.15	3841.96	3803.20	1320.428	43.659	
37	3872.11	63.45	5.29	3895.08	3841.96	1816.846	148.966	
38	3920.93	66.21	4.89	3948.21	3895.08	1671.307	138.229	
39	3971.18	63.56	18.06	3999.89	3948.21	1442.994	551.412	

Graph 2 : FTIR Analysis of Polyherbal syrup

Table.7. FTIR Peak Assignment of Polyherbal Syrup

Peak (cm-1)	Functional group	Assignment
3392.58	O-H stretching	Alcohols/Phenols
2933.16	C-H stretching	Alkanes
1633.84	C=C stretching	Aromatic compounds
1375.41	C-H bending	Alkanes
1035.15	C-O stretching	Alcohols/Ethers

DISCUSSION

Ashwagandha, Brahmi and Curcumin were included in the developed Polyherbal syrup, which demonstrated acceptable physicochemical and organoleptic properties. The syrup's appearance, pH, viscosity, d1ensity and stability were all satisfactory. UV examination revealed the existence of bioactive chemicals, whereas FTIR analysis verified the presence of distinctive functional groups of phytoconstituents. The behavioral investigation showed that the experimental animals' cognitive abilities were enhanced. Overall, the findings indicate that the prepared Polyherbal syrup may have health-promoting and memory-improving qualities.

CONCLUSION

The present work deals with the preparation and evaluation of polyherbal syrup using curcumin, Brahmi and ashwagandha. The pH, density, viscosity and organoleptic characteristics of the formulation were within acceptable limits The presence of required phytoconstituents was confirmed by UV and FTIR studies. The results imply that prepared Polyherbal syrup has adequate physicochemical properties and may be subjected to additional pharmacological evaluation. Moreover, the overall content of the syrup could be modified by the therapeutic potential of the plant chemicals featured in the formulation. But further study is needed to establish efficacy and acceptance.

RECOMMENDATION

1.Further pharmacological study is necessary to evaluate the therapeutic efficacy of the prepared Polyherbal syrup.

2. Extensive toxicity and safety studies are recommended to ascertain the formulation's safety profile.

3. Stability studies may be conducted to determine the shelf life and storage conditions of the syrup.

4. More extensive clinical studies are recommended to verify the formulation's effectiveness.

5. Further research could be conducted to enhance the formulation and boost its therapeutic potential.

APPRECIATION

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COMPETING INTERESTS

The author have declared that there are no competing interests associated with this Research work.

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