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Effect of antioxidant activity of basella on convulsions mediated enzymes

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ABSTRACT

Seizures and epilepsy are inter-related conditions that are often seen in the patients with neurological conditions. In general there are various types of epilepsies like the grand mal, petit mal and clonic tonic types. Typically epilepsies are caused due to the oxidative free radicals that are generated in the brain. There are numerous investigations and researches that are carried out in this field to prove that the oxidative stress and free radicals are the primary etiology for the brain damage and epilepsy. The epilepsy and seizures are often evaluated with the elevation or variations in the brain enzymes. Herbs are few of those drugs that contain rich antioxidants and chemicals. There had been numerous investigations that were performed to prove the activities. They also contain chemicals like Vitamins, polyphenols, flavonoids, alkaloids etc. that posess the activity on brain. In this study, formulations were prepared using the extracts of Basella alba, Bacopa moneira. The effect of these formulations on the antioxidant levels in the brain are estimated and compared to the standard formulation Saraswathiaristam formulation. The formulations showed a better activity compared to the standard ayurvedic formulation. The prevention of tissue damage was estimated by determining the antioxidant enzyme levels.

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INTRODUCTION

Seizures and epilepsy are inter-related conditions that are often seen in the patients with neurological conditions. In general there are various types of epilepsies like the grand mal, petit mal and clonic tonic types. Typically epilepsies are caused due to the oxidative free radicals that are generated in the brain. Any physical damage or any psychological stress causes the liberation of free radicals. These free radicals cause the alteration of the enzymes in the brain. This intron causes the permanent or temporary damage to the brain tissues and nerves. It is clear that the brain tissue liberates free radicals and is prone to the same leading to further damage eventually causing seizures [1, 2].

There are numerous investigations and researches that are carried out in this field to prove that the oxidative stress and free radicals are the primary etiology for the brain damage and epilepsy. The epilepsy and seizures are often evaluated with the elevation or variations in the brain enzymes. Having this in mind, the antioxidants have been advocated to cause the normalization of the elevated brain enzymes and have a positive effect on the brain by controlling seizures. Instead of causing the brain damage or nerve damage, they will fight the generated free radicals and instantly prevent the brain damage. Brain enzymes like SOD, catalases, GPRs,

GPXs, GTs are significantly raised in the brain tissue. These drugs are known to cause the elevation in the protection from the tissue damage. These drugs are cause the elevation in the protective enzymes in the brain and normalize them to make sure the epilepsy is prevented [3, 4].

With an understanding the antioxidants combat the generated free radicals effectively and we can assume that the antioxidants can have a positive effect on the epilepsy too [5]. Apart from causing brain damamge or nervine damage, these free radicals adversely affect other tissues and cause damage to the body functions. Due to the generation of free radicals, the protective enzymes like superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase and glutathione transferase levels are significantly reduced [6]. These antioxidant drugs are known to elevate the protective enzyme levels in the body and restore them to ensure the proper functioning is done [7].

Herbs are few of those drugs that contain rich antioxidants and chemicals. There had been numerous investigations that were performed to prove the activities. They also contain chemicals like Vitamins, polyphenols, flavonoids, alkaloids etc. that posess the activity on brain [8, 9]. A poly herbal formulation was prepared using Basella alba and Bacopa moneira at various concentrations and prepared formulations were tested on laboratory animals to prove the antiepileptic activity on the brain enzymes. These formulations were compared to the standard ayurvedic formulation Saraswathiaristam [10–12].

MATERIALS AND METHODS

The plant parts that are used in preparation of formulations were collected from the local farm and authenticated. These were dried and extracted using ethanol using soxhlet apparatus. The extract was filtered and dried using a rotary evaporator and the yielded extract was 19.04 % w/w. these extracts were formulated in specified ratios as per [Table 1]. These formulations wer compared with the ayurvedic formulation.

Laboratory animals that are used to investigate for the activity are albino rats of wistar strain. They weighed about 140-160g in weight and both male and female rats were included in the study. They were bought from local supplier and were maintained in their own polypropylene cages for about 5 days to accilimatize to the laboratory conditions. The rats were divided into 5 groups which contained 6 animals in each batch. 1^{st} batch animals receive normal saline solution. This batch is considered as control group. This group of animals did not receive the induction of seizures. All the batches were induced with epilepsy and 2 weeks before the induction of seizures, formulations were administered to the rats of fourth and fifth group. Third group received standard ayurvedic formulation [2].

Electric shock Induced Convulsion method

In this method, the electric shock is used to induce the seizures. Animals were given electric shock using an electroconvulsiometer. With the induction of the seizures the animals were noticed and sacrificed. The brain tissue of the rats was isolated carefully and was dropped into formalin solution. This tissue was homogenized and subjected to centrifugation at 5000 rpm for 15 mins. The supernatant was collected and used to analyse for the brain antioxidant enzymes [13, 14]. The brain homogenate tissue was weighed and is mixed with Tris buffer solution at 4^0C. This was then estimated for enzymes using standard procedures.

Pentylene Tetrazole induced convulsions method

This method of induction of convulsions uses PTZ at a dose of 90mg/kg of rat and is administered via the subcutaneous route. 30 mins after administration of PTZ, the rats were kept under monitoring to identify the induction of seizures. With the induction of epilepsy, the animals were sacrificed and the brain tissue was carefully isolated. This tissue was homogenized and subjected to centrifugation at 5000 rpm for 15mins. The supernatant was collected and used to analyse for the brain antioxidant enzymes. The brain homogenate tissue was weighed and is mixed with Tris buffer solution at 4^0C. This was then estimated for enzymes using standard procedures.

RESULTS

The formulations showed a comparably better activity to the standard 'saraswathiaristam' in restoring the brain enzyme after seizures induction. The effect of formulation on catalases was clear by reduction the enzymes after seizure induction. The first formulation showed the restoration of the catalases. SOD protects the brain from the oxidation due to the free radicals that arises from the metal ions. With the induction of the seizures, the SOD levels are lowered significantly and are avoided from further reduction with the administration with formulationsand start ayurvedic formulation saraswathi aristam. This was evident from the data that is tabulated.

The GPXs are very helpful in the lowering of the peroxide free radicals with the lowering of reactions

Table 1: Ingredients of the formulations

Ingredient	Quantity	
	Formulation 1	Formulation 2
Bacilla alba	100	–
Bacopa moneira	–	100
Elettaria cardamomum	20	20
Tinospora cordifolia	20	20
Piper longum	20	20
Piper cubeba	10	10
Woodfordia fruticosa	500	500

Table 2: Formulationeffect on the antioxidant enzymes of brain in Electric Shock InducedConvulsions Method

Group treatment	Catalase (Units/mg)	Glutathione Peroxidase (Units/mg)	Superoxide Dismutase (Units/mg)	Lipid Peroxidation (Nmol/mg)	Glutathione Reductase (Units/mg)
Normal saline	24.21±0.07	26.45±0.99	13.72±0.32	2.34±0.73	27.02±0.84
EIC + saline	19.32±0.34*	18.13±0.62*	7.29±0.91*	5.08±0.23*	6.10±0.98*
EIC + Saraswathiaristam	20.45±0.06	20.63±0.74	9.03±0.48	4.57±0.69	4.61±0.53
EIC + Formulation 1	22.74±0.47^a	23.42±0.56^a	12.36±0.53^a	3.14±0.38^a	3.78±0.97^a
EIC + Formulation 2	23.58±0.03^a	25.91±0.74^a	13.14±0.67^a	2.87±0.99^a	3.35±0.86^a

Table 3: Formulation effect on the antioxidant enzymes of brain in PTZ induced convulsions method

Group treatment	Catalase Units/mg	Glutathione Peroxidase Units/mg	Superoxide Dismutase Units/mg	Lipid Peroxidation Nmol/mg	Glutathione Reductase Units/mg
Normal saline	23.79±0.14	26.58±0.23	14.46±0.68	3.15±0.49	33.72±0.75
EIC + saline	16.83±0.05*	17.34±0.11*	9.21±0.74*	6.10±0.98*	22.12±0.59*
EIC + Saraswathiaristam	18.15±0.07	19.52±0.72	10.34±0.83	4.63±0.54	25.01±0.73
EIC + Formulation 1	19.34±0.13^a	21.63±0.55^a	11.04±0.94	3.72±0.99^a	28.25±0.46^a
EIC + Formulation 2	21.27±0.06^a	24.02±0.94^a	14.52±0.59^a	3.34±0.85^a	31.89±0.91^a

and the reduction of the oxygen free radicals that are present in the brain. The induction of epilepsy in both methods resulted in the lowering of the GPX concentration in the brain tissue. With the administration of the formulations, the prominent lowering of the glutathione peroxidases and prevented the brain tissue from the prolongation of the oxidative damage [15]. [Table 2]

The values were represented as the mean and the standard error of mean; P<0.001* significant when compared to normal group. A - Significant when compared to EIC + saline group

The LPX levels in the brain are increased with the induction of epilepsy and the seizures. The peroxide levels prevent the brain damage as it reacts with the hydrogen peroxide. As there is a rise in peroxidation of the tissue there is a significant damage in the brain. This also lowers the GPXs in the brain. The data that resulted is due to the oxidative damage that is happening due to the generation of the free

radicals. The formulation prevented this damage in the brain and also this is presumed due the presence of the antioxidant compound and also the Bacosides that are known to be the brain tonics. These chemical constituents are known to lower the free radical generation and prevent the lipid peroxidation [16]. [Table 3]

The values were represented as the mean and the standard error of mean; P<0.001* significant when compared to normal group. A - Significant when compared to PTZ + saline group

CONCLUSION

In this study, formulations were prepared using the extracts of Basella alba, Bacopa moneira. The effect of these formulations on the antioxidant levels in the brain are estimated and compared to the standard formulation Saraswathiaristam formulation. The formulations showed a better activity compared to the standard ayurvedic formulation. The prevention of tissue damage was estimated by determining the antioxidant enzyme levels.

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Conflict of Interest

The authors declare that they have no conflict of interest for this study.

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