Double-Blind Comparative Trial of Herbomineral Antioxidant Formulation with Ubiquinone (Coenzyme Q10) in Oligoasthenospermia

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ABSTRACT

Aim: To evaluate the efficacy of an antioxidant herbomineral formulation, Addyzoa (Charak Pharma Pvt. Ltd., Mumbai, India) in oligospermia in comparison with Ubiquinone (Coenzyme Q10).

Methods: A double-blind, randomised controlled trial was conducted at Dr Vaze's clinic, Mumbai. A total of sixty patients, 30 in each group, were studied. Cap Addyzoa was administered in a dose of two capsules twice a day for three months and Cap Ubiquinone (Coenzyme Q10) 30 mg, one capsule thrice a day. The main study outcomes were an increase in sperm count and improvement in motility and morphology of sperms.

Results: In the Addyzoa group, an average increase of 2.74 million sperm was observed, whereas in the Ubiquinone (Coenzyme Q10) group sperm count increased by 2.27 million. In both groups, the increase in motility was on average 1.5 to 2 times. Though

the observed difference between the two groups is not statistically significant ($p > 0.1$), the 0.47 million difference appears to be clinically significant.

Conclusion: The study concluded that treatment with Addyzoa is effective in improving the semen quality by increasing the sperm count and sperm motility. The results are comparable with that of Ubiquinone (Coenzyme Q10) on semen parameters.

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INTRODUCTION

Male infertility, with its clinical and psychosocial implications, poses a significant challenge to the physician and to the society as a whole. Recent studies have indicated that the prevalence of oligospermia is extremely high in metropolis as well as in smaller towns of India. According to the results of one such study, in Kurnool in Andhra Pradesh (a state in India), the prevalence of oligospermia was 51%; it was 31% in Mumbai and between 15% and 30% in other three cities like Jalandhar, Jodhpur and Bangalore (1). The alarmingly high prevalence of oligospermia has resulted in an increase in male infertility. Almost in 50% of couple infertility, male partner suffers from oligospermia. In 15% of subfertile couples, in approximately 30% of the cases, abnormality is identified in the male partner and in another 20%, abnormalities are detected in both the partners. For the majority of men with infertility, no specific causal factor can be identified.

In majority of cases, defective sperm function is the commonest cause for male infertility (2). Defects in sperms are associated with low sperm concentration (oligospermia), poor sperm motility (asthenospermia) or abnormal sperm morphology (teratospermia). These defects are collectively called as oligoasthenoteratospermia (3). Recent research suggests that large proportion of infertile men have elevated levels of seminal reactive oxygen species (ROS). Various ROS like superoxide, hydroxyl, nitric oxide, peroxide, and peroxynitrile are generated in excess by immature and abnormal spermatozoa and by contaminating leukocytes associated with genitourinary tract inflammation. These ROS have been associated with oligoasthenoteratospermia. As spermatozoa membranes are rich in poly-unsaturated fatty acids (PUFA), sperms are more susceptible to the damage caused by ROS by lipid peroxidation. Environmental factors like pesticides, exogenous estrogens and the process of aging itself, further induce oxidative stress (4, 5, 6). Free radical-induced sperm damage can occur within the semen, in the epididymis or in the testis. By damaging the membrane integrity, free radicals impair sperm motility as well as sperm viability.

Normally, semen contains antioxidant mechanisms, which protect spermatozoa from oxidative stress by neutralizing ROS. However, when there is an improper balance between ROS and antioxidant mechanisms, spermatozoa are damaged leading to male infertility.

In addition, the intracellular antioxidant enzymes cannot protect the plasma membrane that surrounds the acrosome and the tail, forcing spermatozoa to supplement their limited

intrinsic antioxidant defenses by depending on the protection afforded by the seminal plasma (7).

Oxidative stress attacks not only the fluidity of the sperm plasma membrane, but also the integrity of DNA in the sperm nucleus.

Since ROS induced lipid peroxidation in sperm cells is an important factor affecting male infertility, antioxidants could assume a significant therapeutic role in these patients.

Therefore, the present study was carried out to evaluate efficacy of Addyzoa (Charak Pharma Pvt. Ltd., Mumbai, India), a herbomineral formulation (Table 1), in comparison with Ubiquinone (Coenzyme Q10) 30 mg in oligospermia.

2. Materials and methods

2.1 Study protocol

This clinical study was undertaken at Dr. Vaze's clinic, Mumbai. This was a double blind, randomized, comparative study. The protocol was approved by the ethics committee. Before enrolling patients in the study, written informed consent was obtained after explaining the nature of the study.

Patients with infertility persisting longer than 2 years were examined twice before recruitment in the study. The main inclusion criteria were oligospermia (less than 20 million sperm counts) and asthenospermia (less than 50% motile sperms).

Complete physical examination and biochemical tests like CBC, ESR, blood sugar, renal function tests, liver function tests and semen examinations were performed in the screening examinations. Two semen samples were collected after abstinence of 4 days at baseline and at the end of the trial

2.2 Patients: Sixty patients in the age group of 25-42 were included in the trial.

2.3 Dose and Duration*:* Thirty patients received Addyzoa, two capsules twice a day and 30 patients received Ubiquinone (Coenzyme Q10), one capsule three times a day for 3 months.

2.4 Semen analysis: Semen analysis was performed according to WHO guidelines and included physical parameters as well as sperm count, motility and morphology. Semen analysis (and all the other biochemical investigations) was performed at the same centralized laboratory.

2.5 Statistical evaluation: Student's t-test was applied to evaluate the average sperm count increase from the baseline at the end of the trial.

3. Results

Of the 60 patients, 30 received Addyzoa and 30 patients received Ubiquinone (Coenzyme Q10). The sperm count at baseline and on completion of the therapy with corresponding increasing sperm count is shown in Table No. 2. The main expected improvement was the sperm count increase. No adverse effects were reported during the treatment.

In the Addyzoa group, an average increase of 2.74 million in sperm count was observed, whereas in Ubiquinone (Coenzyme Q10) group, sperm count increased by 2.27 million. Though the observed difference between the two is not statistically significant ($p > 0.1$), the 0.47 million difference in sperm count appears to be clinically significant (Figure 2).

3.1 Motility

Overall, the motility showed a significant increase in both groups. The increase in motility was on average 1.5 to 2 times.

3.2 Morphology

Marginal improvement in morphology was observed in both groups.

Discussion: Advances in medical sciences for the last three decades have identified the presence of excessive oxidative stress responsible for male infertility. Sperms are made up of unsaturated fatty acids and are very sensitive to free radical damage by lipid peroxidation. Many environmental, physiological and genetic factors have been identified as a cause of poor sperm function and infertility. All these factors lead to reactive oxygen

species (ROS). Therefore, the role of anti-oxidants in the treatment of male infertility is becoming popular.

In view of free radical damage to the sperms, anti-oxidant mechanisms are important in the maintenance of sperm motility, the spermatogenesis and the ability of sperms to fertilize the ovum.

The ingredients of Addyzoa such as Amalaki (Emblica officinalis) (9), Ashwagandha (Withania somnifera) (10), Guduchi (Tinospora cordifolia) (11,12), Shatavari (Asparagus racemosus) (13), Safed Musli (Chlorophytum tuberosum) (14), Kapikachhu (Mucuna pruriens) (15), Balamool (Sida cordifolia) (16) and Vidarikand (Ipomoea digitata) (17) are all reputed anti-oxidants. These ingredients are used in a number of antiageing, rejuvenating and virilizing formulations.

Various studies suggest that these medicinal plants might be potent and novel therapeutic agents for scavenging of NO and the regulation of pathological conditions caused by excessive generation of NO and its oxidation product, peroxynitrite.

The individual constituent herbs in Addyzoa have been extensively studied for their spermatogenic and antioxidant effects. For example, the antioxidant activity of tannoid active principles of Amalaki (E. officinalis) consisting of emblicanin A (37%), emblicanin B (33%), punigluconin (12%) and pedunculagin (14%), was investigated on the basis of their effects on rat brain frontal cortical and striatal concentrations of the

oxidative free radical scavenging enzymes, superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX), and lipid peroxidation, in terms of thiobarbituric acid-reactive products. The results also indicate that the antioxidant activity of E. officinalis may reside in the tannoids of the fruits of the plant, which have vitamin C-like properties, rather than vitamin C itself (9).

Ashwagandha (W. somnifera), the other important ingredient, has direct spermatogenic effect. The effect of lyophilized aqueous extract of Withania somnifera on testicular development and on serum levels of testosterone, ICSH and FSH has been studied in immature male Wistar rats. A notable increase was observed in the testicular weight of animals treated with the extract. Histological examination revealed an apparent increase in the diameter of seminiferous tubules and the number of seminiferous tubular cell layers in the testes of treated rats as compared with control ones. The extract of Withania somnifera elicited notable spermatogenesis in immature rats. Serum testosterone and FSH levels were lower in animals treated with the plant extract than controls, whereas ICSH level was higher in treated animals. It was concluded that the extract of Withania somnifera has a direct spermatogenic influence on the seminiferous tubules of immature rats presumably by exerting a testosterone-like effect (18).

In previous studies, the efficacy and safety of Addyzoa in improving seminal parameters has been studied extensively. The effect of 6 month's therapy with Addyzoa on sperm parameters was investigated in a randomized, placebo-controlled study of 200 men with idiopathic subfertility. The results indicated that Addyzoa resulted in statistically

significant improvements in sperm density (368%), motility (197%) and morphology (183%) as well as other routine sperm tests. Pregnancy rates among the subfertile couples, to which these men belonged, were found to be 25% with Addyzoa over a two-year follow-up period as compared to 2% with placebo (19).

The effects of Addyzoa on male factor subfertility have also been compared with clomiphene citrate. In a 6-month, randomized study of Addyzoa in 100 men in comparison with clomiphene, treatment with Addyzoa resulted in statistically significant improvements in sperm density, motility, morphology as well as other routine sperm tests. Pregnancy rates among the subfertile couples, to which these men belonged, were 22% and 19% in the Addyzoa and clomiphene groups, respectively, over a one-year follow-up period (20).

As such, significantly high proportion of male subfertility is unexplained. In such a scenario, infertility can significantly affect their relationship as it often becomes the central issue in their lives. Even in the era of assisted reproductive techniques (ART), therapy with anti-oxidants like Addyzoa helps in improving the semen parameters. Further, Addyzoa also provides an advantage in terms of cost of therapy compared to Coenzyme Q10.

Recent advances in cellular biology have indicated the presence of the facilitative glucose transporter GLUT-5 in sperms. GLUT-5 helps in transporting glucose into the sperm cell, which is then used as a substrate for energy production. Expression of GLUT-5 is susceptible to hormonal and environmental factors such as oxidative stress. Differential

expression of this molecule is associated with several physiological abnormalities (21). We now know that ingredients of Addyzoa such as Shilajit, Amalaki and Guduchi have a significant role in the regulation of glucose metabolism. It is postulated that by virtue of these ingredients, Addyzoa regulates the expression of GLUT-5 in sperm cells, thereby suitably modulating their motility.

The positive effect of anti-oxidants in improving the sperm quality and quantity will restrict indiscriminative use of assisted fertility treatments like intra-cytoplasmic sperm injections (ICSI).

CONCLUSION: This double-blind, comparative trial of the herbomineral antioxidant formulation Addyzoa in oligoasthenospermia concludes that treatment with Addyzoa is effective in improving the semen quality by increasing the sperm count and enhancing sperm motility and morphology. Treatment with Addyzoa resulted in a larger increase in sperm count as compared to Ubiquinone (Coenzyme Q10), though the difference was not statistically significant. In addition, a significant increase of 1.5 to 2 times in motility was observed with Addyzoa and Ubiquinone (Coenzyme Q10) treatment. Improvement in morphology was also observed in both groups. Thus, Addyzoa is a better option for treating male infertility as it also provides an advantage over Ubiquinone (Coenzyme Q10) in terms of cost of therapy. Further trials in a larger number of patients are required to corroborate the findings of this study.

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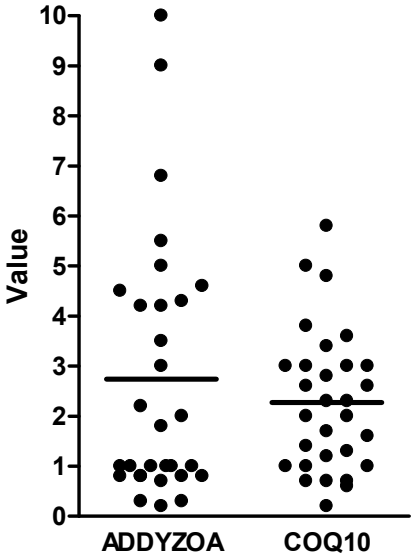
Sperm count in individual patients at baseline and on completion of trial.

ADDYZOA GROUP			UBIQUINONE (COENZYME Q10) GROUP		
Baseline	End point	Increase in sperm count (Millions)	Baseline	End point	Increase in sperm count (Millions)
0.1	0.4	0.3	0.1	3.1	3
9.3	14.3	5	0.1	1.1	1
1.3	3.5	2.2	0.1	1.3	1.2
3.1	4.9	1.8	11.3	11.9	0.6
1.1	1.9	0.8	1.4	2.1	0.7
4.9	5.7	0.8	3.1	3.3	0.2
7.3	17.3	10	7	12	5
11.9	16.2	4.3	5.2	11	5.8
1.1	1.3	0.2	3.7	7.3	3.6
1.1	2.1	1	0.1	1.7	1.6
5.3	14.3	9	6.3	8.9	2.6
3.1	7.3	4.2	3.1	6.1	3
21	23	2	0.1	2.1	2
7.8	12.4	4.6	0.3	1.3	1
1.5	1.8	0.3	13.1	15.9	2.8

	0.1	1.1	1	2.8	5.1	2.3
	2.1	3.1	1	7.2	8.6	1.4
	0.3	1.1	0.8	6	9	3
	2.1	3.1	1	7.2	11	3.8
	0.3	1.1	0.8	4.7	7.3	2.6
	2.1	3.1	1	2.5	5.9	3.4
	0.3	1.1	0.8	7.4	9.1	1.7
	4.1	8.3	4.2	2.1	3.1	1
	0.1	0.8	0.7	3.1	5.1	2
	4.8	8.3	3.5	0.1	1.4	1.3
	11.3	15.8	4.5	11.3	13.6	2.3
	11.3	12.3	1	1.4	2.1	0.7
	7.8	14.6	6.8	5.1	5.8	0.7
	15.3	20.8	5.5	8.3	11.3	3
	9.3	12.3	3	6.2	11	4.8
Averages	**5.04**	**7.776**	**2.736**	**4.346**	**6.616**	**2.27**
	Average sperm count improvement/increase at the end of the trial		**2.74**		**2.27**	

Figure 1 Comparison of the mean sperm count (in millions) between Addyzoa and Ubiquinone (Coenzyme Q10) group after 3 months of therapy

Figure 1



	ADDYZOA	COQ10
Mean	2.737	2.270
Std. Deviation	2.601	1.407

Mean ± SEM of column A	2.737 ± 0.4749 N=30
Mean ± SEM of column B	2.270 ± 0.2569 N=30
Difference between means	0.4667 ± 0.5399
95% confidence interval	-0.6143 to 1.548

P value	0.3910
P value summary	ns
Are means signif. different? (P < 0.05)	No
One- or two-tailed P value?	Two-tailed
t, df	t=0.8643 df=58