NON GENETIC THERAPEUTIC APPROACH TO THE MUSCULAR DYSTROPHIES

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Editor's Comment Tag : *The first author of this article is a son of two members (Dr Md Akbar Husain Bhuiyan & Dr Shahanara Chowdhury) of CMCTA who is a junior medical student in a UK University. This article has been welcomed in this issue to provide a glimpse of the scholastic level and endeavor of a medical student abroad that should be an eye opener for all of us, teachers and sutdents of CMC alike.*

Abstract

Duchenne muscular dystrophy is an incurable genetic disease occurring once in every 3500 male births. Its onset is early and most affected children are diagnosed by the age of 5. It has a poor prognosis and patients rarely live past their early 20s. Treatment with corticosteroids and creatine has proved to be effective in delaying disease progression as supported by findings from several clinical trials, five of which are critically appraised in this review. The effectiveness of these treatments based on the findings of these trials is also discussed.

Key words : muscular dystrophies; therapy

Introduction

A structural defect in the dystrophin glycoprotein complex of muscles results in a condition called muscular dystrophy[1]. Various types of muscular dystrophy are known; of which the most common and severe is Duchenne muscular dystrophy (DMD)[2]. It occurs as a consequence of mutation of the dystrophin gene, the la rgest gene of the human body[1], located at Xp21[3]. The gene codes for dystrophin, a protein which binds actin to a transmembrane protein β dystroglycan[4,5]. Dystrophin protects myofibrils from contractile stress induced tear and necrosis by improving stability of the sarcolemma[5,6].

Xp21

Fig 1 : Chromosome X with the Xp21 region highlighted. (adpted from Muntoni et al)[1]

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Patients suffering from DMD lack dystrophin in their skeletal and cardiac muscles and this is often diagnosed by performing muscle biopsies. Elevated levels of serum creatine kinase can also be detected in many patients[7]. Diagnosis is typically made between the ages of 2 to 5 yrs[8] when parents report that their children are falling frequently, having difficulty rising from the floor or are unable to run. Gower's manoeuvre while rising from the floor, accompanied by pseudohypertrophy of the calves and a waddling gait are the classic clinical signs[9]. Most boys lose ambulation by the age of 10[10]. Pulmonary and cardiac functions also deteriorate in their early teens and most patients die due to respiratory failure or cardiomyopathy by their late teens or early twenties[11]. The disease affects one in 3500 males born[8] and although approximately 1/3 of the cases are a result of new mutations, most cases are inherited from a carrier mother[2].

Until now, DMD remains incurable, although several forms of treatments have been tried to slow disease progression. Their main targets have been dystrophin production, improving muscle repair, the inflammatory response as a result of muscle tear and the unstable sarcolemma. Some of the drug therapies include treatment with corticosteroids and creatine.

Corticosteroids:

Corticosteroids are considered as the most effective form of medication for DMD[12] and are thought to work by interfering with the inflammatory response. Prednisone and deflazacort are two types of corticosteroids among which prednisone is more frequently used. Corticosteroid treated patients have been known to report several side effects, leading to a lot of controversy over the use of corticosteroids as a treatment for DMD[13].

Creatine:

Recently the use of creatine for improving muscle strength has been a hotbed for research. It is known to improve performances where athletes require powerful muscle contractions[14], although its mode of

Table I : A summary of the nine types of muscular dystrophies

Type	Age at Onset	Symptoms, Rate of Progression, and Life Expectancy
Becker	Adolescence to early adulthood	Symptoms identical to Duchenne but less severe; progresses more slowly than Duchenne; survival into middle age.
Congenital	Birth	Symptoms include general muscle weakness and possible joint deformities; disease progresses slowly; shortened life span.
Duchenne	2 to 6 years	Symptoms include general muscle weakness and wasting; affects pelvis, upper arms, and upper legs; eventually involves all voluntary muscles; survival beyond 20s is rare.
Distal	40 to 60 years	Symptoms include weakness and wasting of muscles of the hands, forearms, and lower legs; progression is slow; rarely leads to total incapacity.
Emery-Dreifuss	Childhood to early teens	Symptoms include weakness and wasting of shoulder, upper arm, and shin muscles; joint deformities are common progression is slow; sudden death may occur from cardiac problems.
Facioscapulo humeral	Childhood to early adults	Symptoms include facial muscle weakness and weakness with some wasting of shoulders and upper arms; progression is slow, with periods of rapid deterioration; life span may be many decades after onset.
Limb-Girdle	Late childhood to middle ages	Symptoms include weakness and wasting, affecting shoulder girdle and pelvic girdle first; progression is slow; death is usually due to cardiopulmonary complications.
Myotonic	20 to 40 years	Symptoms include weakness of all muscle groups accompanied by delayed relaxation of muscles after contraction; affects face, feet, hands, and neck first; progression is slow, sometimes spanning 50 to 60 years.
Oculo pharyngeal	40 to 70 years	Symptoms affect muscles of eyelids and throat causing weakening of throat muscles, which, in time, causes inability to swallow and emaciation from lack of food; progression is slow.

action is not well understood. This has given rise to the idea that it may be helpful in slowing down muscle strength deterioration in DMD patients and a few RCTs have been carried out to test this hypothesis.

Aims and objectives

A set of objectives mentioned below were drawn up for the successful completion of this module.

* To gain an understanding of the progression of DMD

* To briefly assess the current non genetic therapies available for DMD patients.

* To compare the pros and cons of corticosteroid treatment demonstrated by long and short term studies

* Evaluate the effectiveness of creatine in treating DMD

Methodology

Initially, text books were looked into in order to provide a platform for an understanding of muscle physiology and pathophysiology of the disease, followed by searching the web for articles. DMD proved to be a very popular muscular dystrophy demonstrated by the abundance of research conducted in this field. Next, various databases were searched for clinical trials of different treatments of DMD. The University of Liverpool's Metalib search engine was used to collectively search eleven databases which included the Cochrane library, Medline (ovid), Scopus and Science Direct. The Electronic journal collections of the University of

Liverpool were also searched for articles are extremely useful but are generally hard to locate on databases. The webpage of this section says "Many of these titles might not be covered by databases like Web of Science". The hits were followed and the abstracts of the articles with relevant titles were read to assess the articles. Once thought to be useful, most articles were successfully downloaded in full, while some articles were unavailable.

Electronic journal collections section of the University of Liverpool

Articles relating to various non genetic treatments were found, of which corticosteroids and creatine were the most abundant. Five papers, of which three were corticosteroid trials and two were creatine trials were chosen for this review. Generally, randomised control trials (RCTs) were preferred but due to the absence of any long term RCTs with corticosteroid, only one was included in this study. All the articles dealing with creatine appeared to be short term and two RCTs were chosen. In order to maintain consistency, all the trials that were chosen dealt only with Human males.

Critical appraisal of the corticosteroid trials:

Study 1: Corticosteroid treatment and functional improvement in Duchenne muscular dystrophy[17]

This long term retrospective study observed the progress of 49 boys suffering from DMD. The boys were divided into two treated groups, one receiving prednisone (N=18) and the other deflazacort (N=12) and a non treated group (N=19). All the subjects of the study had been observed for over 7 years.

The study had a clear objective; to compare the benefits and side effects of steroid use among DMD patients. The approach to achieve the objective was substantial. Following steps were taken to enhance consistency:

* Using patients who have been on steroid therapy for > 2 yrs before losing ambulation

* Having an effective inclusion/ exclusion criteria

* Using only one physical therapist

* Similar mean age, stage of disease and pulmonary function in the patients of the different groups

The study recognises that the patients' efforts may not have been maximum at all times, producing a bias. The side effects of steroid therapy are recorded and the authors mention gaining informed consent, making the trial ethically viable.

The patients however were only assessed every 6 months and the date of the visit when a patient first failed to do a task was considered the first date of loss of that ability. This inevitably would give rise to inaccuracy in the date of loss of a certain function. X-rays for spinal curvature assessment were performed once a year, and this gap in time was too wide too. Pulmonary function was only assessed thrice, at the ages of 7, 10 and 15 yrs making it a not very reliable end point. Administered dose of prednisone was inconsistent since boys with excessive weight gain were put on a lower dose or discontinued. Nothing is mentioned about how this affected the progression of these boys compared to others in the prednisone group. It seems these boys were still included in the trial which could have made prednisone appear to be less effective especially in comparison to deflazacort. The patients were not randomly allocated. It is recognised that many patients chose a certain treatment option due to its cost but the socio-economic difference and its accompanying factors like diet and living conditions were not recognised.

Statistical significance was regarded to values of $P < 0.05$. This study established that boys in the steroid groups were functionally better on all motor tests ($P<0.05$). Prevalence of scoliosis surgery was also less for the steroid groups ($p<0.05$). The pulmonary function of the non treated group was observed to decline significantly after the patients reached 10 years of age. The study concluded that corticosteroid treatment is an effective way of slowing the progress of DMD and should be continued until the side effects become unbearable.

Study 2: Long-term benefits of deflazacort treatment for boys with Duchenne muscular dystrophy in their second decade[18]

This study was also retrospective in nature and compared the progress of a deflazacort treated group (N=40) to a non treated group (N= 34) of patients with DMD.

The method followed by the study to achieve its designated aim was appreciable. A large sample was used and the following steps taken to increase consistency:

* Accepting only patients that were ambulatory at age 7

* All treated patients were on deflazacort for at least 2 years and started between the age of 6 and 8

* Had a well demarcated inclusion / exclusion criteria

* All patients had similar height, weight and pulmonary function before treatment

* The same multidisciplinary team followed all patients

The results were well presented, analysed and adjusted for confounders. The patients' families were informed about deflazacort and given the option of choosing treatment with it, strengthening the study's standing on ethical grounds. The authors also recognised that though patients with deflazacort were on dietary restrictions, inaccurate recording of weight gain as a side effect may have been done due to variable compliance. Other side effects of deflazacort are also mentioned and statistically analysed.

Some of the drawbacks of the study were long intervals of 4 to 6 months between follow up appointments and non random allocation of patients. Though all the boys started on the same dose/kg per day of deflazacort, the dosage was changed through the course of treatment owing to side effects. The change was not the same for all patients and this could have contributed to skewed findings. Differences within the two measurement techniques used to find out left ventricular ejection fraction produced inconsistency. Also, this was only done once every 12 to 24 months and hence, detecting a decrease in left ventricular systolic function could have been delayed by up to almost 2 years, considerably increasing the difference between the actual and apparent onset of cardiac dysfunction.

Setting the value of $P < 0.05$ as statistically significant, the study considered various endpoints to evaluate the benefits of deflazacort treatment. Motor function was greatly improved amongst the treated group and the improvement in pulmonary function reached statistical significance for the treated patients from age 10 onwards. With age, the difference in the pulmonary function of the two groups increased. Cardiac function greatly deteriorated for the non treated group compared to the deflazacort group ($P<0.002$). 90% of the non treated boys had a spinal curvature of >20% and consequently had to undergo spinal surgery, compared to 10% of the treated group. The last endpoint revealed that 12 of the 34 non treated patients died in their second decade of life compared to 2 of their 40 treated counterparts.

Study 3: Intermittent prednisone therapy in Duchenne muscular dystrophy[19]

This is the only RCT of steroid treatment included in this study. This trial followed the course of 16 patients over 1 year 2 months, during which they underwent treatment with creatine and placebo.

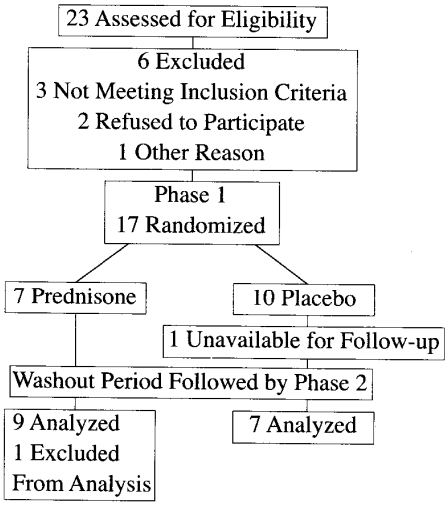

Fig 1 : A summary of how the patients were randomised and followed through in the trial by Earnesto A et al [19]

The trial was double blinded to reduce bias; received ethical approval and patients who gave informed consent were included. The study used a cross over pattern of treatment with creatine and placebo, thus increasing the reliability of the results. Patient selection was based on a well defined inclusion/ exclusion criteria and all patients included were ambulatory at the start of the study both of which helped in increasing consistency amongst the sample. The trial also allowed for a two month washout phase in order to get rid of the corticosteroid remaining in the systems of the patients receiving prednisone treatment first. This increased the accuracy of this trial's findings. The objective of the study was very clearly outlined and easily comprehendible.

The study had only a few drawbacks one of which was the administration of the steroids only on the first 10 days of every month while measurements were taken on day 1, 10 and 30 of every month. For the steroid groups the results obtained on day 30 and day 1 of the next month would be very similar since they would be very close by and be quite different from the result obtained on day 10. Since 2/3 of the

recordings were made when the patients had been without steroid for about 20 days, it is likely that the effects of steroid therapy were not truly represented. Assessment of the Quality of life (QoL) of each patient was done based on the patients' perception of their disease. This gave rise to an inconsistent set of results since expectations from the treatment varied from individual to individual which most likely would affect their evaluation of the disease before and after treatment.

This study treated $P \leq 0.05$ as statistically significant and showed that the force of various muscle groups were improved (P=0.02) during the prednisone treated phase. The study found that the loss of muscle function was slower during the prednisone treated phase based on the fact that time needed to run 9 metres (P=0.005) and climb four stairs (P=0.02) increased significantly less during the prednisone treatment phase.

Table II : A summary of the trials that used corticosteroids to treat DMD

Variables/ study number	**Study I:**		**Study 2:**		**Study 3:**	
Steroid used	Prednisone and deflazacort		Deflazacort		Prednisone	
Sample size	49		74		17	
Nature of study	Retrospective		Retrospective		Double blind, cross over RCT	
Patients followed up till the end	-		-		16	
Length of observation	7 yrs		-		6 months prednisone 2 months washout 6 months prednisone	
Age range of sample	12-15		10-18		5-8	
No. of patients receiving steroid treatment	Prednisone- 18 Deflazacort- 12		40		17	
No. of patients receiving other/ no treatment	No drug treatment-19		No drug treatment 34		Placebo 17	
Presence of inclusion criteria	-		Yes (onset of weakness before 5yr age, proximal muscle weakness, diagnosis of DMD)		Yes (diagnosis of DMD, ambulatory)	
Presence of an exclusion criteria	Yes (changing types of steroid, comorbidity, delayed steroid use)		Yes (boys with significant cognitive delays)		Yes (use of steroids 2 months prior to the trial)	
Duration of steroid treatment	> 2 yr before loss of ambulation		> 2 Yrs.		-	
Outcome	Steroid groups	Non steroid group	Steroid group	Non steroid group	Steroid group	Non steroid group
Improved motor function	√		√		√	
Improved pulmonary function	√		√		-	-
Higher prevalence of surgery for scoliosis		√	-	-	-	-
Development of spinal curvature (>20%)	-	-	10%	90%	-	-
Prevalence of LVS dysfunction (ejection fraction <45%)	-	-	10%	58%	-	-
Death in 2nd decade	-	-	35%	5%	-	-
Noted side effects of steroid treatment	Cataracts, behavioral changes, hypertension, weight gain, vertebral fractures		Cataracts, weight gain		Irritability, cushingoid appearance, hyperactivity	

Critical appraisal of the creatine trials:

Study 1: Creatine monohydrate enhances strength and body composition in Duchenne muscular dystrophy[20]

This study was a RCT, the gold standard for clinical trials. The study used a cross over mechanism to obtain results of 30 patients to find out whether creatine increases muscle strength and fat- free mass (FFM) in DMD patients.

The study had a well outlined objective and was double blinded to reduce bias. Patients were randomised into either a creatine or a placebo group and then crossed over to the other group, with a wash out phase of at least 5 weeks.

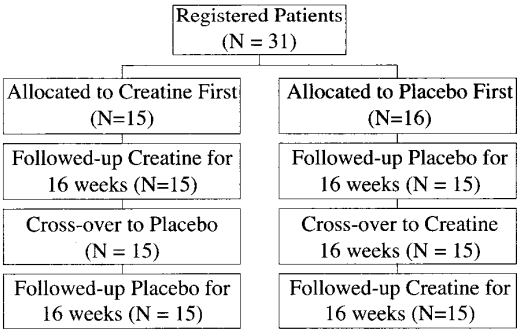

Fig 2: A summary of the method of randomisation followed by Tarnopolsky MA et al[20]

The crossing over would help reduce anomalies of the result due to variations within the sample. It was made sure that none of the subjects had taken any creatine supplement for at least 3 months prior to treatment. Strength testing was done in the morning to reduce the chances of getting biased results due to build up of tiredness as the day progressed. Informed consent was obtained from the guardians of the patients and ethical approval was taken from ethics committees.

One of the flaws of this study was that 50% of its patients were already undergoing steroid therapy for DMD for atleast 6 months before starting this trial. However, crossing over should have reduced irregularities in data. The process of blinding is not described and the trial was conducted over two venues by two different investigators. Though they spent time together to reduce observer bias, the chances of such bias still occurring are quite high. The patients were asked to complete an activity scale increasing the probability of recall bias and inconsistency due to individual variation.

$P \leq 0.05$ was regarded as statistically significant and this study observed that creatine use produced significant increased grip strength in the dominant hand ($P<0.05$). A reduction in the concentration of bone degradation product N-telopeptides was also noted ($P<0.05$) for the creatine group. Creatine therapy appeared to also be successful in increasing patients' FFM ($P<0.05$). There was a trend of increased muscle weakness for the placebo phase ($P=0.056$). However, creatine failed to improve pulmonary function and functional tasks. Side effects of creatine were absent and neither did it produce adverse effects on steroid treated patients.

Study 2: CINRG randomized controlled trial of Creatine and Glutamine in Duchenne muscular dystrophy[21]

This RCT looked to compare the positive and negative effects of creatine and glutamine on DMD patients. A total of 50 patients were divided randomly into one group treated with creatine (N=15), a second group treated with glutamine (N=19) and a third placebo treated group (N=16). The patients were followed up for 6 months during which 5 patients dropped out.

This gold standard clinical trial study design made its aims very clear early on. The study was double blinded using the double dummy technique which helps reduce observer bias. None of the 50 boys had undergone steroid therapy before and this helped in accurately evaluating the effect of creatine as a solo treatment. Patients that had recently taken creatine or other supplements were excluded. Patients with ventricular arrythmias and symptomatic cardiomyopathy were also excluded from the trial. All patients that were included were ambulatory, enhancing consistency of the disease stage of the sample. Informed consent was obtained before the trial began.

The age of the patients used ranged from 4 to 10 years and this could give rise to erroneous results since DMD progresses with age, though it was treated as a confounding variable in the analysis. The patients were recruited and followed up at 10 different sites and this greatly increases inconsistency of the findings due to inter tester variation.

The study failed to show any statistically significant improvements of manual muscle testing (MMT)

results amongst the creatine group compared to the placebo group though trends of improvements were observed. Qualitative muscle testing (QMT) results however were greatly improved for the creatine group (P= 0.07), demonstrated by slower strength deterioration. No side effects of creatine were observed.

Table III : A summary of the trials that used creatine to treat DMD

Variables/ study number	**Study I**		**Study II**	
Sample size	30		50	
Nature of study	Double blinded, cross over RCT		Double blinded RCT	
Patients followed up till the end	30		45	
Length of observation	4 months creatine 6 week washout 4 months creatine		6 months	
Age range of sample	Mean age: 10 +/- 3		4-10	
No. of patients receiving treatment with creatine	30 (for 4 months)		15	
No. of patients receiving other/ no treatment	30 (for 4 months)		Glutamine-19 Placebo-16	
Outcome	Creatine	Placebo	Creatine	Placebo
Improved results of manual muscle testing	-	-	Unchanged	Unchanged
Improved results of quantitative muscle testing	√		√	
Increased Fat free muscle mass	√		-	-
Reduction of bone breakdown marker	√		-	-

Discussion

The low prevalence and lack of an established form of treatment for DMD has made effectively researching this disease difficult. Despite this, treatment with corticosteroids and creatine have shown a lot of promise. Some such researches were the ones mentioned above.

Both, the long term corticosteroid trials conducted by Balaban et al[17] and Biggar et al[18], and the short term RCT conducted by Beenakker et al[19] showed similar trends as evident from table II. Findings from all three studies show that corticosteroid treatment helps improve motor function while improved pulmonary function was also observed in the long term studies. Biggar et al[18] in their study found that long term steroid use among DMD patients significantly slowed down the onset of left ventricular systolic dysfunction. This is particularly important since respiratory failure and cardiomyopathy as a result of DMD are the leading causes of death among DMD patients[8]. A reduction in the development of severe scoliosis requiring surgery was observed among the steroid treated patients of both long term studies. In their study Biggar et al[18] also observed that death in the second decade was much higher for non steroid treated patients compared to the steroid treated group. All steroid trials also noted side effects of varying severity and it was observed that deflazacort had fewer and less severe side effects associated to it compared to prednisone.

The data in table III depicts the usefulness of creatine in the treatment of DMD. Both Tarnopolsky et al[20] and Escolar et al[21] demonstrated in their results that creatine had an association with increasing hand grip strength. Significantly increased FFM and reduction in the plasma marker for bone degeneration were also demonstrated by Tarnopolsky et al[20]. However, in the same study it was also observed that creatine had no role in slowing down the rate of deterioration of pulmonary function.

Conclusion

DMD patients have shown benefits from treatment with both corticosteroids and creatine. However, to maximise their benefits, further studies are required. Researches aimed at producing a better picture of the pathophysiology of DMD should be undertaken. Since the beneficiary effects of corticosteroids are already established, researches should be done to reduce the severity of its side effects. This could be done by finding the ideal dose and establishing the ideal age at which to start and stop treatment. Treatment with creatine may be improved by accomplishing a better understanding of its mechanism. DMD patients have also been known to benefit from other drugs such as oxandrolone and as demonstrated in a recent study, from perindopril. Further research on DMD should also look into these drugs and evaluate their efficiency when used alongside corticosteroids to slow disease progression and improve QoL of the patients.

Limitations

Inability to access some articles were the major limitations encountered while working on this review.

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