INDIAN PLANTS AS A POTENTIAL TOOL FOR PREVENTING MULTIFUNCTIONAL SPINAL MUSCULAR ATROPHY

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Abstract

Spinal muscular atrophy is a hereditary degenerative condition of lower motor neurons characterized by increasing muscle weakening and atrophy. It results from lower amounts of the "survival of motor neuron" (SMN) protein. Because of mutations in the SMN1 gene on chromosome 5q13, on each copy of chromosome 5, the SMN gene, known as SMN1 and SMN2, forms an inverted duplication. SMN2 and SMN1 differ from each other by five nucleotide alterations that do not alter the amino acids. Exon 7 of SMN2 is excluded from most transcripts by a crucial single nucleotide alteration in an exonic splice enhancer. Because of this, the (SMN2) gene's duplication creates a less functional SMN protein. The majority of people who have spinal muscular atrophy have homozygous deletions of the SMN1 gene, but they still have at least one copy of the SMN2 gene. The SMN2 gene copy number, which varies naturally throughout the population, the concentration of SMN protein, and the severity of the illness are roughly correlated. Investigations on the function of the SMN protein are still ongoing. There also seem to be genes that modify, which may play additional functions in motor neuron function. In addition to lower motor neurons, problems at the neuromuscular junction have been seen in animal models of spinal muscular atrophy. Animal models and aberrant muscle growth in patients who are most severely affected to increase the expression of SMN2 or have an impact on other modifying genes, pharmacological substances are being developed as potential therapeutics. In addition to employing stem cells to replace deteriorated motor neurons, researchers are attempting to accomplish this goal through gene therapy, antisense oligonucleotides, and other methods. Recommendations in a consensus statement for the multidisciplinary supportive care of people with spinal muscular atrophy have helped many patients survive longer and live better lives during the past ten years. In this review we focus on some treatment measure for SMA and suggested some natural extract which may be potential inhibitors for SMA illness.

Keywords: Spinal Muscular Atrophy, Neuromuscular, Mutation, Genetics, Motor Neuron

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1. INTRODUCTION

The term spinal muscular atrophy (SMA) refers to a collection of hereditary illnesses that are all defined by the degeneration of anterior horn cells and the subsequent muscle atrophy and weakening. Wedding and Hoffmann were the first to write about the illness in the 1890s. The survival motor neuron (SMN) gene was discovered as the disease-causing gene in 1995, and this allowed for the localization of the genetic error to 5q11.2-q13.3. [1]

Motor neuron death and gradual muscle atrophy happen from the loss or mutation of the SMN1 gene, which lowers the level of the SMN protein. Despite recent advancements in our knowledge of the molecular pathways underlying the pathophysiology of the illness, there is still no known cure for SMA. We discuss the clinical manifestations, molecular pathogenesis, diagnostic approach, and development of therapeutic regimes for better comprehension and management of SMA in this review.

Following cystic fibrosis, SMA is the second most prevalent fatal autosomal recessive condition, with an estimated incidence of 1 in 6,000 to 1 in 10,000 live births and a carrier frequency of 1/40 to 1/60. [1]

Discovery

Guido Werdnig of Austria and Johann Hoffman of Germany were the two researchers who first described SMA. Both men had observed several new-borns who, within the first few months of life, started to exhibit signs of muscle weakness. They additionally noticed that this illness appeared to run in families.

As they investigated this enigmatic disease, particularly in the anterior horn, a region of the spinal cord, they observed that the motor neuron cells in these infants appeared to degenerate. This area of the spinal cord, which is close to the front of the cord and is connected to the skeletal muscles, contrasts with other spinal cord segments that are connected to touch and other sensations. They conducted investigations and found spinal muscular atrophy.[3]

SMN GENES, RNA, PROTEIN, AND FUNCTION

Despite the wide clinical variety of the disease, homozygous deletions of the SMN1 gene or, in rare cases, additional mutations, are the causes of all types of 5q SMA (OMIM 600354).

A person with SMA who carries more SMN2 gene copies (OMIM 601627) exhibits a milder phenotype. The main factor affecting how severe a disease is this copy gene. Additionally, independent modifiers like neurocalcin delta (NCALD; OMIM 606722) and plastin 3 as well as uncommon SMN2 variants can significantly affect the severity of the disease (PLS3; OMIM 300131). The following sections cover the structure of the SMN gene, cis- and trans-regulatory domains that control SMN splicing, and the function of the SMN protein. [2]

Population variations and the development of the SMN gene region

Only humans have two SMN paralogs, SMN1 and SMN2, while all other animals only have one SMN gene. Despite reports of SMN duplication in monkeys, long-read next-generation sequencing research has shown that they only have one copy of the gene. This suggests that during the evolution of primates into humans, SMN and the nearby genes on chromosome 5q13.2 were duplicated. The SMN duplication caused one copy to be able to distinguish into the current SMN2 copy.

Between various racial groups, there are significant structural differences. There are eight times more people with a 2 SMN1/0 SMN2 haplotype in the black African population than there are in the Caucasian population. According to the theory from outside of Africa, the duplication most likely happened in the African population. Then, when one copy of the African population diverged, SMN2, which may have served as the foundation for the rest of the world, was created. Therefore, 1 SMN1/1 SMN2 is the most common haplotype among Caucasian and Asian people [5]. This variation in the number of SMN1 copies per haplotype could also explain why these populations have a higher SMA carrier frequency.

Classification and description

Based on the age of onset and level of motor function acquired, SMA is clinically divided into four phenotypes. [3]

SMA TYPE 1

The most severe and prevalent form of SMA is type 1 (Werdnig-Hoffmann disease), which affects roughly 50% of patients. Infants with SMA type I typically begin to exhibit clinical symptoms before 6 months of age, never learn to sit alone, and, without treatment, typically do not live through their first two years.

Clinically speaking, all children with SMA type I exhibit a mix of severe hypotonia and weakness, sparing the facial muscles, and are generally accompanied by a typical respiratory pattern. The weakness is often symmetrical, more proximal than distal, and lower limbs are typically weaker than higher limbs. Although sensitivity is maintained, deep tendon responses are missing or lessened.

SMA TYPE 2

The start of SMA type II often occurs between the ages of 7 and 18 months. While some patients are cannot stand up unassisted and others are able to walk on their own.

There are no deep tendon reflexes, and upper-extremity tiny tremors are frequent. The more severe type II patients are more likely to develop joint contractures and kyphoscoliosis in the first few years of life. There are no deep tendon reflexes, and upper-extremity tiny tremors are frequent.

SMA TYPE3

Clinically diverse patients with SMA type III (Kugelberg-Welander disease) are included. They normally accomplish all significant motor milestones.

They do, however, begin to lose proximal muscle strength support during infancy. While some children may require wheelchair help, others may be able to walk and lead fulfilling adult lives despite having a slight physical weakness. Patients who have difficulty walking frequently develop scoliosis and other health issues including obesity and osteoporosis are linked to poor mobility.

SMA TYPE 4

For patients with adult-onset (> 18 months) and mild course, SMA type IV has been included in this categorization. Patients in this category include those who can walk as adults and are healthy in terms of their respiratory and nutritional systems.

Since all SMA forms fall under a single spectrum and have a common origin, the characteristics of the intervention and the goals chosen ultimately define which patients will participate in clinical trials, not the historical classification.

SMA Type	Copies *SMN2*	Percent of Cases	Onset	Motor Milestones	Clinical Features	Natural History Prior to Disease-Modifying Therapy
0	1	Rare, <1%	Prenatal, at birth	Non-sitter, no head control	Generalized weakness, hypotonia, respiratory failure, poor feeding, contractures	Death within weeks of birth
1	1–2	45%	0–6 mo	Non-sitter	Proximal predominant weakness, respiratory insufficiency, poor feeding, tongue fasciculations	Death by age 2
2	3	20%	6–18 mo	Sits independently, never stands or ambulates	Proximal predominant weakness, tongue fasciculations, minipolymyoclonus, scoliosis	Most alive at 25 years
3	3–4	30%	A: 18 mo –3yr B: 3–30 yr	Ambulates independently	Proximal, lower extremity predominant weakness, abnormal gait	Normal lifespan
4	4 or more	<5%	> 30 yr	Ambulates independently	Maintain ability to ambulate	Normal lifespan

2. Etiology and molecular genetics

On chromosome 5q13, there are two virtually identical SMN genes known as the centromeric (SMN2) and telomeric (SMN1) genes, which control spinal muscular atrophy.

Exon 7 is alternatively spliced, which results in SMN2 genes producing less full-length transcripts (SMN-fl) and proteins as well as larger amounts of mRNA without exon 7 (SMN-del7), which leads to a shorter and more unstable protein. More than 95% of patients have a homozygous SMN1 disruption because of SMN1 gene deletion or SMN1 to SMN2 conversion.

The SMN protein is expressed and localised in the cytoplasm and nucleus of all cells. It is made by the SMN genes and is mostly located in motor neurons in the spinal cord. The SMN protein is mainly found within the nucleus' "gems," which are dot-like structures related to coiled (Cajal) bodies (Gemini of coiled bodies) [26]. Although

though the exact cellular function of the SMN protein implicated in the pathogenesis of SMA is yet understood, cells from people with spinal muscular atrophy have less gems than cells from controls and carriers.

The following is supported by experimental data from several sources: The SMN protein, which is a constituent of a high molecular weight complex involving at least eight other proteins, is necessary for the Smith class core proteins with the hypothesis rich snRNPs to effectively assemble (U snRNP). Spliceosomes, which are biological components that largely consist of U snRNPs, are responsible for pre-mRNA splicing.[7]

Epidemiology

Spinal muscular atrophy is estimated to affect 7.8 to 10 live births per 100,000, or one in every 6000 to 10,000, with spinal muscular atrophy type I expected to affect 4.1 live births per 100,000. According to a 2005 Cuban study, people with African ancestry are more likely than the general population to develop type I spinal muscular atrophy (3.53 per 100,000 live births) (0.89–0.93 per 100,000 live births). The estimated carrier frequency for SMN1 gene changes was 1:38-1:50, although lower frequencies have also been noted. An epidemiologic investigation to ascertain the prevalence in various ethnic groups in North America was completed in 2009. Hispanics had the lowest carrier frequency (1 in 10) among all racial/ethnic groups.[7] (1 in 37, or 2.7%) are Caucasians. Also of intermediate frequency were Ashkenazi Jews (1 in 46 or 2.2%) and African Americans (1 in 56 or 1.8%). In spite of the high carrier frequency, spinal muscle atrophy is less common than expected.

3. Clinical characteristics

The condition Wedding-Hoffman, also known as type I spinal muscular atrophy, frequently shows symptoms between birth and 6 months of age. Infants exhibit progressive proximal weakness that mostly affects their legs rather than their limbs. They have weak head control, hypotonia, which enables them to hang upside down and "slide through," and are flexia, which makes them "non-sitters," or people who cannot sit down. Additionally, the diaphragm is mostly unaffected by intercostal muscle weakness, which results in a bell-shaped chest and a paradoxical breathing style commonly known as "belly breathing." babies with type I spinal muscular atrophy. Tongue fasciculation and swallowing issues, which raise the risk of respiration, are the conventional indications of atrophy. (Kolb, 2016) Form I spinal muscular atrophy

Type II spinal muscular atrophy

Individuals who have intermediate spinal muscular atrophy, also known as type II spinal muscular atrophy, occasionally can sit without assistance but never walk. They exhibit hypotonia, areflexia, and progressive proximal weakness, which mostly impacts the legs as opposed to the arms. People also endure intercostal muscular weakening as scoliosis develops over time, which results in severe restrictive lung disease. In some situations, they develop mandibular ankyloses as well as joint contractures. Hand tremors or polyminimyoclonus are seen. Although the body mass index of the high-functioning, no ambulatory individuals may be low (at the third percentile or less, compared with normal children), they have a higher relative fat mass index and are more susceptible to being overweight.

Type III spinal muscular atrophy

Those with Kugelberg-Welander illness, sometimes referred to as type III spinal muscular atrophy, can eventually walk ("walkers"). They rarely or never get scoliosis or respiratory muscle weakness, but they do experience increasing proximal weakening that eventually affects the legs more than the arms and may result in the need for a wheelchair. They might have polyminimyoclonus or hand tremors. Their life expectancy is not significantly different from that of the general population.[10]

Outlier

Extreme phenotypic outliers at either end of the spectrum exist in some patients. Spinal muscular atrophy "type 0" refers to neonatal patients who exhibit severe hypotonia and weakness, likely from the prenatal onset, and who have a history of reduced fetal movements. Typically, no motor milestones are ever reached. Other findings include joint contractures, facial diplegia, are flexia, and atrial septal abnormalities. The need for non-invasive ventilation and endotracheal intubation at delivery is caused by respiratory failure, a significant cause of morbidity and mortality in spinal muscular atrophy type 0.

As a result, fewer people are expected to live past the age of six months. Additionally, it was discovered that individuals with SMN1 gene deletions and spinal muscular atrophy had a congenital condition called arthrogryposis multiplex, which is characterised by joint

contractures affecting the last two body regions. There have been reports of congenital axonal neuropathy affecting the motor and sensory nerves, facial weakness, joint contractures, ophthalmoplegia, and respiratory failure in newborn siblings with deletions in the region of the chromosome linked to spinal muscular atrophy. Additionally covered was type IV spinal muscular atrophy, a less severe variation of adult-onset SMA. Type 0 and type IV phenotypes of spinal muscular atrophy are most common in individuals with SMN1 homozygous exon 7 deletions.[11]

4. Genetics

The SNM gene

According to research using linkage analysis, all three kinds of spinal muscular atrophy can be located on chromosome 5q11.1–13.3. Lefebvre et al. found this area to be the location of the SMN (survival of motor neuron) gene in 1995. This gene was absent or disrupted in 98.6% of the patients in their group. A significant inverted duplication of a 500 kb fragment is part of the region's complex structural makeup. The SMN2 gene, a duplication of SMN1 that varies from SMN1 by only five nucleotides, is placed in the centromeric portion of the region, whereas the SMN1 gene, which is earlier in evolutionary history and is present in tins duplication, is found in the telomeric portion of the area. The primary difference between SMN1 and SMN2 is an exonic splicing enhancer in exon 7 of SMN2, which possesses a critical C-to-T transition. Since it is translationally silent, this change has no impact on the amino acid sequence. Since it has an impact on the alternative splicing of the gene, exon 7 is frequently spliced out of or excluded from SMN2 messenger RNA transcripts. This altered messenger RNA is converted into a shortened form of the SMN protein, most of which is degraded. Exon 7 is not always removed from SMN2 pre-messenger RNA, resulting in a negligible quantity of full-length transcript and consequently functional protein being produced by SMN2, but this yield is only 10% greater than that of SMN1. Patients with spinal muscular atrophy have two copies of the SMN1 genesis that are both deleted or disrupted, which causes the remaining copies of SMN2 to only produce a very small amount of SMN protein. There are 95–98% Telomeric SMN1 gene deletions in spinal muscular atrophy patients. The rest is present or has undergone gene conversions or small intragenic mutations between SMN1 and SMN2. The second scenario involves an exon 7 disruption caused by a frame shift or point mutation in SMN1, which effectively changes SMN1 into SMN2 [4]. The instability of this region of chromosome 5 and the presence of repeats with low copy numbers as well as the inverted SMN1 and SM2 genes cause de novo mutations to happen often. 15–25% of people with normal chromosome 5 compositions lack copies of SMN2, which varies among normal individuals. In individuals with spinal muscular atrophy, SMN2 copy quantity and phenotypic severity were discovered to be closely associated. It has been discovered that the SMN2 gene possesses a positive modifier.

In the DNA of three unrelated individuals, a single nucleotide mutation in exon 7 (c.859G>C) results in the production of a new exonic splicing enhancer, boosting the inclusion of exon 7 and, consequently, the amount of full-length protein. The phenotype in these patients was less severe and did not correspond with their SMN2 copy counts, confirming the idea that not all copies of SMN2 are created equal and the favourable modifying effect of this sequence alteration. In SMN1-deleted females with the same number of SMN2 copies as their affected siblings, plastin 3 expression was likewise found to be a sex-specific protective modulator of spinal muscular atrophy. Yet a later study discovered that the gene's expression was highest in post pubertal females with spinal postpubertalphy type III, intermediate in spinal muscular atrophy type II, and lowest in spinal muscular atrophy type I, highlighting a correlation with disease severity. A sex-, age-, or puberty-specific modifier is also possible for plasmid.[13]

5. Diagnosis

Clinical traits are very suggestive of a SMA diagnosis in the severe variety of a floppy infant or weak young child. Mental clarity and focus are always beneficial. It is common for the weakness to be symmetrical, more proximal than distal, and stronger in the legs than the arms. Clinical classification shows an association between the severity of weakness and the age at which it first manifested with postponed motor milestones. Deep tendon reflexes are more or less active, depending on the age at disease onset and the length of the disease. Sensitivity is unaffected.

The initial level of diagnostic testing for a patient suspected of having SMA must look for homozygous deletion of the SMN1 gene. The absence of SMN1 exon 7 supports the diagnosis of SMA (with or without exon 8 deletion). The

test has a sensitivity range of up to 95% and an approximate 100% specificity range.

The kid should be assessed and given additional diagnostic testing, taking other disorders into consideration, if the electrophysiological test excludes a motor neuron disease.[14]

The function of the SMN Protein

With a molecular weight of 38 kilo daltons, SMN is a 294 amino acid long, widely expressed protein (KD). SMN is present in both the cytoplasm and nucleus. It is concentrated in the nuclear structures Cajal bodies and Gem bodies as well as the nucleoplasm of the nucleus. It is also present in significant quantities in the cones that generate motor neurons [35]. Aside from having a substantial effect on the splicing apparatus, SMN has also been connected to ribonucleoprotein biogenesis, which includes the synthesis, metabolism, and transport of several ribonucleo-proteins. It is a member of the SMN complex, a bigger protein complex that also contains profilins, spliceosome UsnRNPs, Gemini, and Sm proteins. This complex is essential for the production of snRNPs [36–45]. Even cell cultures cannot survive without SMN, which is not surprising considering the wide range of activities that SMN has been implicated in. The complete lack of SMN genes is embryonically fatal in almost all metazoan life forms investigated.

Molecular mechanism: SMA's splicing problem

The complex called the spliceosome mediates the process of splicing, and a number of factors can affect it. Classification, Diagnose, Background, and Molecular Mechanism: Spinal muscular atrophy...SMN1 as well as SMN2 splicing is precisely controlled by various cis- and trans-acting factors. Exon 7 of the SMN2 gene experiences a C-T transition at position 6, which affects the function of an exonic splice enhancer (ESE; recognised by SF2/ASF to promote exon 7 inclusion) and/or develops an exonic splice suppressor (ESS; recognised by hnRNP A1/A2), which results in exon 7 being skipped.[15]

6. Therapeutic techniques

Despite the fact that the SMN2 gene locus is a target for SMA drugs, SMA cannot be cured.

The general treatment approaches for SMA aim to compensate completely or partially for the absence of the SMN1 gene by increasing the levels of functional SMN protein using three different techniques—inducing the expression of SMN2, altering SMN2 transcript splicing, and stabilising full-length SMN mRNA and/or protein. Further being investigated for the treatment of SMA are gene and stem cell treatments. These and other strategies are covered below.

SMN-dependent therapies: As was previously mentioned, there is an inverse relationship between the SMN2 gene copy number and the severity of the disease [1,3,6], which leads to the hypothesis that one of the keys to developing a SMA drug treatment may be to directly target the SMN2 gene in SMA patients through various pathways. A different approach to generating SMN protein is gene replacement therapy.

Activation of SMN2 promoter: By chromatin condensation, histone deacetylases (HDACs) inhibit the transcription of many genes, including SMN2, which activates the gene's promoter. As a result, HDAC inhibitors can increase the transcription of the SMN2 gene, leading to the synthesis of more full-length SMN transcripts.

Correction of splicing Another treatment strategy being looked into for SMA is inhibiting exon 7 skipping to increase the amount of full-length transcript from the SMN2 gene. repair of the splice. HDAC inhibitors like VPA, TSA, and sodium butyrate appear to have a dual effect on the production of SMN mRNA; in addition to influencing splicing, they also appear to open the chromatin structure and hasten transcription.

Full-length SMN transcript stabilization: The vital enzyme Dcp for the degradation mechanism was discovered and is used in this extremely novel technique by Singh et al. Targeted by substand-located quinolones, which bind to the enzyme and change its conformation such that it is no longer catalytically active. This is how blocking the process leads to an increase in full-length SMN mRNA in cell culture.

Gene therapy: Adeno-associated virus (scAAV) 8 and 9 vectors encoding the SMN1 cDNA have been employed during the past three years by a number of research groups to treat mice models of SMA, and this is one of the most exciting advances in SMA therapy. Apart from an overall improvement in disease phenotype, theseveralts have caused the most notable prolongation of mice lifespan yet known. However, in order for this therapy to be effective—as it is with other treatment modalities—early pre-symptomatic intervention is necessary.

7. Future Directions

Combinatorial approaches for treating SMA will probably be necessary to target not only the CNS but also other tissues that are impacted by the absence of SMN, unless and until gene therapy and ASO therapies are approved for clinical safety as a therapeutic option. Monotherapy will probably struggle to match the outstanding results achieved so far in the field of SMA using gene therapy and ASOASOombinatorial approaches for treating SMA will likely be necessary to target not only the CNS but also other tissues that are impacted by the absence

There are numerous approaches to target SMA, as was previously discussed. By activating the SMN2 gene (PRL), stabilising the SMN2 transcript (p38 pathway activators like celecoxib), stabilising the SMN protein (proteasome inhibitor bortezomib), activating the SMN2 gene, stabilising the SMN protein (proteasome inhibitor bortezomib), stabilising the SMN protein (neuro protective compounds, Rho kinase inhibitor) Genetics of the disease

The complex genomic region on chromosome 5q13.1 contains the harbor's gene, which is the root cause of SMA disease. This region's genetic makeup can be seen in a piece of the harbor's senses that underwent an inverted duplication. Spinal Muscular Atrophy: Classification, Diagnostics, Background, Molecular Mechanism. There are copies of (SMN, neuronal apoptosis inhibitor protein "NAIP," SERF, and GTFH2) in both the telomeric and centromeric regions. 95 percent of SMA patients had homozygous deletions of the SMN1 gene, which are most likely the cause and were discovered in a 1995 analysis. Each and every case of SMA has one or more copies of the nearly identical SMN2 gene.

Five nucleotide changes in exons 7 and 8 distinguish this set of genes. Position 6 in exon 7 contains differences in the critical nucleotide A C to T transition, which makes SMN2 only partially functional. Exon 7 alterations are generally not included in transcripts. After translation of this mRNA, an unstable, truncated, non-oligomer zing isoform of the SMN protein is produced.

Nonetheless, 5–10% of fully functional SMN transcripts are still produced by the SMN2 gene. All SMA patients have one or more copies of the SMN2 gene, which, due to its limited functionality, acts as a protective disease modifier. The SMN2 gene has variable copy numbers in the general population. As a result, the quantity of SMN2 genes, which, depending on copy number, can produce between 10 and 50% of SMN protein, is inversely connected with the severity of the illness.

In spite of the fact that low levels of SMN protein are required for embeven though Type II patients have three copies of SMN2 compared to Type I patients' two copies, long-term motor neuron survival in the spinal cord is not achievable at low levels of SMN protein. Type III and type IV both have three to four copies of the SMN2 gene. Those with 5 or more copies of the SMN2 gene are completely asymptomatic and shielded from the disease's expression, despite the fact that they do not have a functional SMN1 gene.[17]

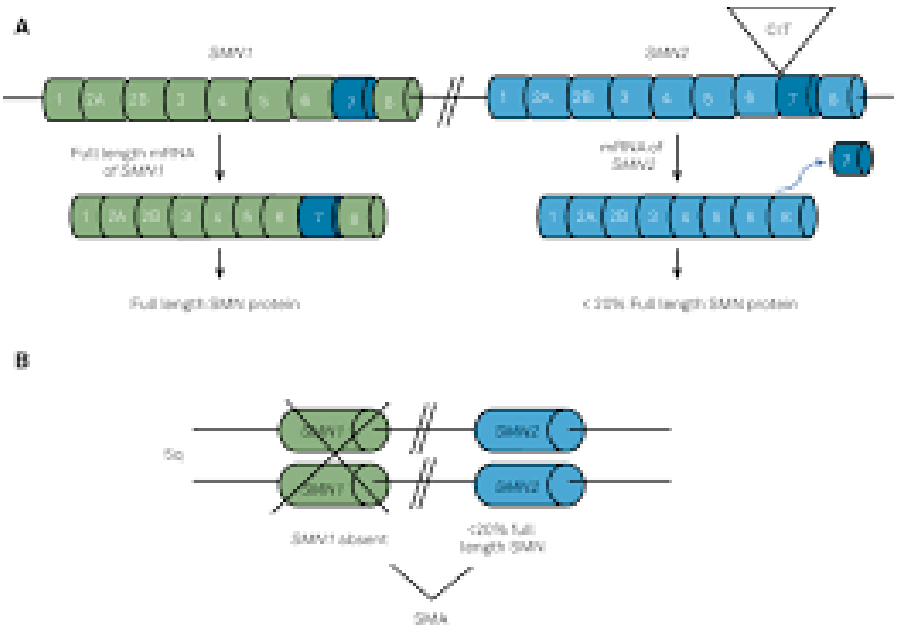

Fig1: Genetics of Spinal Muscular Atrophy

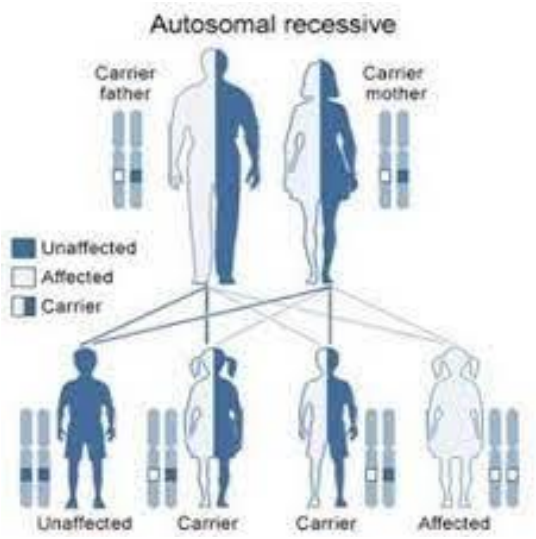

Fig 2 : Autosomal recessive condition of the disease

8. GENETIC TESTING AND PHENOTYPE-GENOTYPE CORRELATION

96% of SMA patients have homozygous SMN1 absence, while 4% have point mutations. This is a remarkable mutation spectrum. Due to the intricate genomic structure, gene conversion and de novo rearrangements are quite frequent. In addition to the inverse correlation between illness severity and SMN2 copy number, other SMN2 gene variations or independent modifiers, including PLS3 or NCALD, may also affect disease severity.

Genetic Testing

The most reliable genetic test for SMA is a multiplex ligation-dependent probe amplification of SMN1 and SMN2 [10]. Using this technique, it is possible to determine the precise number of SMN2 copies, healthy heterozygous carriers, and SMA patients with a homozygous SMN1 deletion. It is also possible to determine SMA patients with one SMN1 copy who may also be compound heterozygous for a second, subtle SMN1 variant. It cannot differentiate between persons with one SMN1 gene on each chromosome 5 and individuals with two copies of the SMN1 gene on each chromosome 5 (cis version). Furthermore, it is incapable of identifying minor mutations in SMN1 (which affects 6% of SMA cases). A person with two copies of SMN1 can nonetheless be a SMA carrier as a result (5% false-negative rate).

The human genome contains two SMN genes, which complicates the search for SMN1 variations. Subtle SMN1 variations can be found in two ways: (a) Exaggeration and Cloning of SMN cDNA products, followed by the PCR-based detection of those who carry the SMN1 gene, or (b) amplification and cloning of SMN cDNA products, followed by the PCR-based detection of those who carry the SMN1 gene.

The Mutation Spectrum of SMN1

Regardless of the severity of their conditions, all 5q SMA patients exhibit biallelic SMN1 mutations. An exon 7 or exons 7 and 8 homozygous deletion in SMN1 or SMN1 gene conversion into SMN2 is the genetic aetiology of SMA.

While the majority of SMA type I patients have a true SMN1 deletion, gene conversion of SMN1 leads to an increase in SMN2 copy number in SMA types II and III. Incomplete gene conversion produces hybrid SMN1/SMN2 genes with exon 7 from the SMN2 origin and exon 8 from the SMN1 origin. A study of SMN1 deletion screening in SMA patients found 96% homozygous deletions.

In about 4% of SMA patients with SMN1 deletion, subtle mutations on the second copy of chromosome 5 are also present. Rarely, and exclusively in consanguineous families, two modest SMN1 variations have been reported. The SMN1 gene now contains 108 unique pathogenic SMN1 mutations. The most prevalent frameshift mutations are p.Arg133fs148 in the Spanish population, caused by a 4-bp deletion (c.399 402delAGAG), and p.Gly261fs269 in the Spanish, French, and US populations, caused by an 11-bp duplication. Tyr272Cys and p.Thr274Ile are the most common subtle mutations in the German and Polish populations, respectively. (Kolb, 2016)

De Novo Mutations

A 500-kb repeat unit that is duplicated, inverted, and contains other genes besides SMN1 and SMN2 is the primary cause of the SMA region on 5q13's extreme instability.

The area is highly polymorphic, as shown by physical and genomic maps, long-read next-generation sequencing, and other data. The various units vary in both number (ranging from zero to four per chromosome) and orientation.

A high rate of de novo mutations occurs in the area as a result of unbalanced recombination and gene conversion. Consequently, only 2% of SMA instances are due to de novo mutations, which are more typically brought on by uneven recombination than gene conversion events.

Phenotype–Genotype Correlation

The SMN2 copy number, with more copies leading in a milder phenotype, has the greatest impact on the severity of the SMA phenotype. Despite how strong the link is, it is not unqualified.

Thus, 50% of SMA type IIIa patients have three copies, 61% of SMA type IIIb patients have four copies, and 75% of SMA type IV patients have four copies of the SMN2 gene. By comparison, 73% of SMA type I patients have two copies of the gene, 78% of SMA type II patients have three copies, and 78% of SMA type II patients have three copies of the gene. Three copies of the SMN2 gene are present in 20% of type I patients, 78% of type II patients, and 51% of type III patients, however they have a lesser predictive value than two or four copies.

While p.Thr274Ile causes a milder phenotype, certain missense mutations, such p.Tyr272Cys, generate a severe phenotype. The amount of SMN2 copies linked to the modest mutation, however, also affects how severe the phenotype will be. A missense mutation in SMN2 called c.859G>C (p.Gly287Arg), for example, increases full-length SMN2 transcripts, which benefits the SMA phenotype. Variants in SMN2 can also impact severity.

Carrier Detection and Frequency

The global SMA carrier frequency, or 3,795/193,586 is 1:51, according to molecular genetic data. All investigations eliminated SMA carriers with two copies of SMN1 on one chromosome (2:0) or with a point mutation or minor deletion within SMN1 (1:1D). The proportion of control people in the German population with two SMN1 genes on each chromosome was 4.8%, while the proportion of SMA parents with a mild SMN1 mutation was 1.7%. A quantitative examination of SMN1 screening will therefore reveal these individuals as false negatives. Europeans seem to have the highest frequency (1:41), whereas Sub-Saharan Africans appear to experience the lowest frequency (1:145).

The homozygous SMN1 deletions were found in 30 out of 213,276 new-borns in a pilot new-born screening study in Germany, yielding a SMA incidence of 1:7,109 and a carrier frequency of 1:42. This carrier frequency closely resembles the earlier predicted frequency for fewer Europeans (1:41). Out of 1,530 newborns who underwent the first newborn screening in the US, 38 were found to be SMA carriers (1:40), which is higher than the published frequencies for other American ethnic groups, which range from 1:48 to 1:97 but fairly similar to the European frequency. In contrast to the other studies, fewer people in the United States were checked for newborns overall.
Molecular mechanism of splicing defects of SMA protein

The complex called the spliceosome mediates the process of splicing, and a number of factors can affect it. Classification, Diagnose, Background, and Molecular Mechanism: The particular splicing of Spinal Muscular Atrophy is controlled by many cis- and trans-acting factors. Exon 7 of the SMN2 gene experiences a C-T transition at position 6, which affects the function of an exonic splice enhancer (ESE; recognised by SF2/ASF to promote exon 7 inclusion) and/or develops an exonic splice suppressor (ESS; recognised by hnRNP A1/A2), which results in exon 7 being skipped.

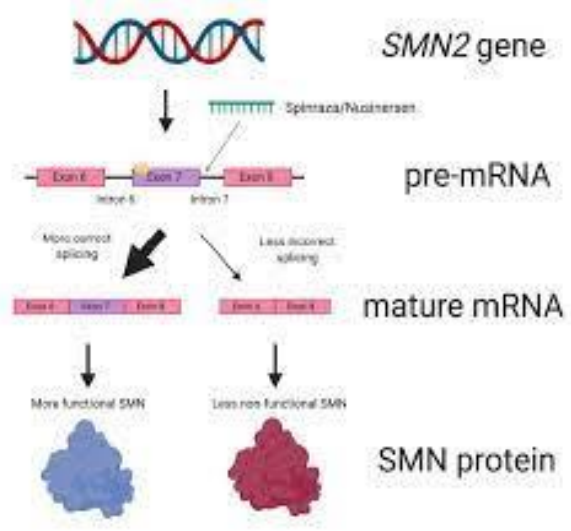

Fig 3: SMA Splicing

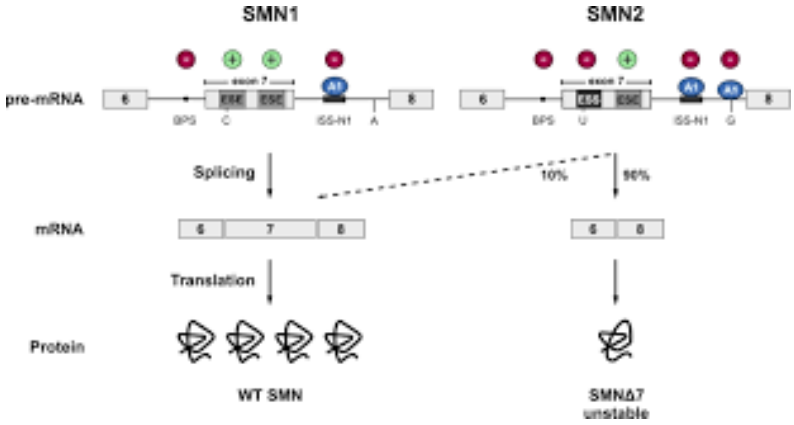

Fig 4: Minor Intron Splicing

Identifying the Knowledge Gap in SMA Education and Support

With the above-mentioned changes in therapies and phenotypes, as well as earlier diagnosis through newborn screening, there is an urgent need for support, training, and mentoring for physical therapists with less experience with SMA in urban and rural settings lacking well-established support systems or neuromuscular programmes. Physical therapists and clinical assessors engaging in SMA clinical trials acquire training and expertise in evaluation and standard of care as part of the trial process; however, such chances are scarce for community-based practitioners who are not involved in research. There are consequently little possibilities for SMA assessment and care-specific professional development. For practitioners without a formal education track, it can be challenging to stay current with the physical therapy assessment and treatment of an uncommon condition.

It is crucial to give possibilities for SMA research, treatment, and care closer to home as well as to teach and educate working physicians on optimal practises in research and clinical settings. For doctors who are new to SMA research and/or clinical care, a knowledge-to-practice gap has been found. This led to the adoption of numerous procedures to close the gap, providing physical therapy researchers and clinicians who treat people with SMA with tools and focused evidence-based information. These procedures—needs assessment for the SMA Clinical Trial Preparedness Program, creation of the SMA Best Practices Clinical Evaluator Toolkit resource, and (3) recently established education initiatives—are the subject of this study.[20]

9. Therapeutic strategies

The SMN2 gene area is a target for SMA care, although there is no current treatment for SMA.

The three primary tactics used in the general therapy options for SMA are full-length SMN mRNA and/or protein stabilisation, SMN2 expression, and SMN2 transcript splicing. Increased amounts of functional SMN protein are the goal of these methods, which are intended to either completely or partially make up for the lack

of the SMN1 gene. Treatments using stem cells and genes are also being developed for SMA.

These and other strategies are covered in the next paragraph.

- **SMN-dependent therapies**: As was already indicated, there is a negative correlation between the SMN2 gene copy number and the severity of the condition, which raises the possibility that one of the keys to producing an effective SMA therapy is to directly target the SMN2 gene in patients. A different way to make SMN protein is by gene replacement therapy.
- **Activation of SMN2 promoter**: Histone deacetylases (HDACs) prevent the transcription of SMN2 by condensing chromatin. Since more full-length SMN transcripts and proteins are produced as a result of increased SMN2 gene transcription, HDAC inhibitors may benefit patients. In cell culture, mouse models, and clinical studies, various HDAC inhibitors have been investigated as potential SMA therapies. They all demonstrated promise in cell culture and mouse models, and patients tolerated sodium butyrate, valproic acid (VPA), and phenylbutyrate all three well. Recent studies using a variety of HDAC inhibitors, such as LBH588, Trichostatin A (TSA), and Suberoylanilide Hydraemic Acid (SAHA), have demonstrated SMN2 gene activation in both culture and in a number of animal models of neurodegeneration. In addition to these substances, we have demonstrated that the lactation hormone prolactin (PRL), which may penetrate the blood-brain barrier and, through binding to its receptor, activates the JAK2/STAT5 pathway, upregulates the transcription of the SMN2 gene. It's interesting to note that compared to wild-type mice and cell culture, the level of SMN induction with prolactin in the genetically modified 7 SMA mouse model is substantially higher. This is because the SMN2 gene is the only source of SMN protein. The difference between the promoter regions of the SMN1 and SMN2 genes, the latter of which only possesses STAT5a transcription binding motifs, has led us to the conclusion that this is the cause of the problem. This may be favourable because SMN2 is the only SMN protein source for all SMA patients. As a result of PRL's successful testing and demonstration of safety in humans for the treatment of lactation-insufficient mothers, it may displace other substances that have not yet undergone clinical safety testing and join the small group of drugs that may have immediate potential for SMA therapeutic potential.
- **Correction of splicing**: In order to boost the amount of full-length transcript from the SMN2 gene, exon 7 skipping suppression is another treatment approach being researched for SMA. HDAC inhibitors such sodium butyrate, TSA, and VPA appear to have a dual effect on the synthesis of SMN mRNA; they not only appear to affect splicing but also appear to open the chromatin structure and speed up transcription. The antibiotic aclarubicin has been shown to increase full-length SMN transcript by altering the splicing process in vitro. The most effective treatments to stop SMN2 exon 7 skipping and thus correct splicing are antisense oligos (ASOs). According to research, an ASO complementary to the pre-mRNA sequences of SMN2 exon 7 inhibits positive splicing factors, resulting in a rise in the production of full-length SMN transcript and protein. Antioxidants are not effective as SMA therapies because they cannot cross the blood-brain barrier. But Hua and associates (2011) reported that systemic

ASO therapy greatly enhances motor function and lengthens survival in SMA mice by raising SMN levels mostly in peripheral tissues, particularly the liver. Unexpectedly, they discovered that the SMN levels in the Brain tissues had barely grown. A number of issues (clinical safety, the amount of ASO, cost, immune response, etc.) need to be answered before ASOs are used in clinical settings to treat SMA.

- **Full-length SMN transcript stabilization:** An important part of the RNA degradation machinery, the decapping enzyme DcpS, was targeted by C5-substituted quinolones in this relatively recent technique by Singh et al. This interaction caused the enzyme to become catalytically inactive. In this way, full-length SMN mRNA degradation is inhibited, leading to an increase in SMN protein levels in cell culture. A separate approach has shown that the SMN mRNA's 3' UTR contains a specific AU-rich element-rich region that marks the mRNA for destruction. The RNA binding protein HuR accumulates in the cytoplasm upon activation of the p38 pathway and stabilises the stabilising factor by binding to the ARE in the 3'UTR region of the SMN mRNA, as shown by research from our group. It's important to note that transcript stabilisation doesn't appear to stop the

translation of the SMN protein. By using p38 activating substances that may cross the blood-brain barrier, this team developed a novel method for stabilising SMN mRNA, opening the door to the development of new SMA therapeutics.

- **Full-**length **SMN protein stabilization:** Aminoglycosides are a subclass of antibiotics that can mask the premature stop codon mutations in some genes, enabling read-through translation. Translation termination is changed as a result of the changing ribosomal reading site conformation. Many aminoglyco-sides, including tobramycin and amikacin, have been used successfully to increase SMN protein levels in patient fibroblasts. However, they have not yet proven that they are reliable and secure when used in living organisms. Another potential therapy method targets the ubiquitin-proteasome system, which controls intracellular protein turnover. Proteins are tagged with polyubiquitin thanks to the operations of the enzymes E1 (Ub activating enzyme), E2 (Ub conjugating enzyme), and E3 (Ubpolyubiquitinb ligase). The polyubiquitin modification marks the protein for the proteasome complex's destruction. Many proteins, including SMN, are part of the ubiquitin proteasome. It has been shown that the FDA-approved proteasome inhibitor bortezomib increases SMN in vitro and in vivo by inhibiting the proteolytic degradation of the protein. Bortezomib must be used with drugs that can treat SMA because it cannot cross the BBB on its own.

1. **Gene Therapy** Gene therapy, which has several potential applications, is one of the most hopeful therapeutic advancements for SMA. In the past three years, various research teams have employed potential elementary adeno-associated virus (scAAV) 8 and 9 vectors carrying the SMN1 cDNA to treat mice models of SMA. These therapies have resulted in the greatest lifespan extension of mice ever observed as well as a general improvement in disease phenotype. The effectiveness of this therapy depends on early pre-symptomatic intervention, as it does with other treatment techniques. There are various challenges to be resolved before this approach to treating SMA may be successfully used in the clinic.
2. **SMN-independent strategies:** There have been some recent developments in SMN-independent SMA therapy methods. These comprise:

- Stem cell therapy: As a potential cure for diseases affecting motor neurons, including SMA, stem cell therapy has generated a lot of interest. It may be used to replace missing motor neurons or, maybe more realistically, to support the current neuron population. From mouse embryonic stem cells as well as primary neural stem cells, brain stem cells. When the medication is injected into the spinal cord, improvements in SMA animal models have been seen. Low disease features and high survival rate. It's unclear if this is finished. replacing host motor neurons with other cells or shielding host motor neurons from various neuroactive chemicals released by the donor cells. These challenges include the production of a large number of stem cells and their efficient implantation into patients, where they might multiply and cover the entire nervous system. Moreover, lentivirus vectors are used to deliver the mixture of components required to create iPS cells in a lab setting; however, these are not suitable for use in patients due to the possibility of insertional mutagenesis, which may result in oncogenesis. A meaningful connection between motor neurons and the host CNS must now be seen as exceedingly implausible, even if motor neurons could grow in situ.
- **Modifying neuromuscular junctions through actin dynamics**: Pharmacological Rho-kinase inhibitor, a downstream effector of RhoA-GTP and a contributor to actin dynamics, significantly prolongs longevity and ameliorates disease phenotype in a mouse model of moderate SMA. This improvement in the disease phenotype is not associated with an increase in SMN and is primarily caused by better, larger, and more established neuromuscular connections (NMJ). This demonstrates that there are fresh, SMN-free approaches for developing SMA treatments.[20]

10.BIOMARKERS

Finding clinically significant biomarkers is urgently needed because there are disease-modifying treatments for spinal muscular atrophy (SMA). Neurological disorders can be measured and evaluated over time using biomarkers. The development of a disease can be tracked by changes in biomarkers, which can also show biological, physiological, or pharmacologic processes that occurred before clinical identification.

Several prospective molecular and physiological markers that evaluate biological media (such as blood and cerebrospinal fluid [CSF]) or nervous

system activity have been identified as potential SMA biomarkers. SMA is a hereditary motor neuron disease marked by motor neuron degeneration and weakening. Examples of such biomarkers include the copy number of SMN2, the levels of SMN mRNA and protein, neurofilament proteins (NFs), plasma protein analytes, creatine kinase (CK), and creatinine (Crn), as well as numerous electrophysiological and imaging tests.

Recent therapeutic advances in SMA have yielded promising results, but there is still a great need to identify and comprehend the role of biomarkers in disease onset and progression.

- **Molecular Biomarkers**

1. Survival motor neuron 2 (SMN2) copy number

SMN2 is an SMN1 paralog with two exonic base changes that result in a significantly reduced ability to splice out intron 7 and thus produce full-length, functional SMN mRNA and protein from the SMN2 gene.

Multiple copies of SMN2 are common in humans. In human and mouse models of SMA, the number of SMN2 copies, also known as SMN2 copy number, is proportional to the amount of full-length SMN protein produced.

In an infantile natural history study, SMA infants with fewer copies of SMN2 had lower motor function scores (MFS) than those with more copies.

2. Survival motor neuron (SMN) mRNA and protein levels

SMN mRNA and protein levels reflect not only the extent of SMN2 gene expression but also the transcription and translation that occurs as a result. Increased mRNA and protein levels, like increased SMN2 copy number, are associated with milder types of SMA.

As a result, SMA therapies have primarily focused on the development of drugs that increase the expression of the SMN protein, particularly in nervous tissue. SMN is ubiquitously expressed and thus detectable in all cell types24, even though previous studies have primarily measured SMN levels in the blood and, less frequently, CSF.

Because of developmental and cell-type specific regulation, SMN expression varies between tissue types.

This, combined with the variety of approaches for measuring SMN expression, has resulted in a wide range of reported expression levels, making comparisons across studies particularly difficult.

3. Neurofilament proteins

Neurofilament (NF) is a cytoskeletal protein that regulates axonal caliber and maintains axonal structural integrity. Following injury, neurons release it, and elevated NF levels can be detected in both blood and CSF. 83,84 With a half-life approaching 8 months, NF may provide insight into axonal changes occurring many weeks before measurement.

The majority of NFs are phosphorylated proteins with a high molecular weight known as phosphorylated neurofilament heavy chains (Pchains, which are resistant to protein degradation. NF can also be found in light (NF-L) and medium-chain conformations. NF proteins have previously been studied in the context of axonal injury, degeneration, and disease as markers of active axonal loss.

4. Creatine kinase (CK) and creatinine (Crn)

The creatine kinase (CK) system is required for the maintenance of energy homeostasis and skeletal muscle function. 105,106 CK is an enzyme that catalyzes the reversible transfer of phosphate to creatine, resulting in phosphor-creatine, which serves as a rapidly mobilizable energy reserve primarily in skeletal muscle.

Creatinine (Crn) is a metabolic waste product of the CK system and a marker of muscle mass that has previously been shown to correlate with disease severity in other denervating motor neuron diseases such as spinal and bulbar muscular atrophy (SBMA).

5. Spinal muscular atrophy multi-analyte panel (SMA-MAP) protein analytes

A subset of the protein analytes was studied in an SMN7 mouse model, and a few were found to be responsive to both the SMA phenotype and postnatal SMN restoration.

Five analytes (dipeptidyl peptidase-IV, fetuin-A, osteopontin, vitronectin, and vitronectin) were significantly altered in SMA mice when compared to wildtype controls, and levels returned to normal after morpholino ASO treatment and subsequent SMN restoration.[22] (22. L. Bürglen et al.)

Review of literature

A category of diseases known as SMA are defined by the degeneration of spinal motor neurons.

We give an update on the most prevalent kind of SMA, proximal or 5q SMA, and talk about the methods used now for diagnosis and therapy. EM-imaging and muscle biopsy
Prior to the development of molecular testing for homozygous deletion, denervation features were utilised to make a diagnosis. Nevertheless, the mutation of the SMN1 gene now allows for precise and targeted diagnosis. People with SMN1 deficiency also have variable numbers of copies of SMN2, a second related gene, which leads to decreased amounts of the survival motor neuron (SMN) protein, which is insufficient for typical motor neuron activity.

The homozygous deletion of the SMN1 gene was found to be the cause of SMA in 1995. Humans have two virtually identical inverted SMN genes on chromosome 5q13. The coding sequence of the two copies of the SMN gene, SMN1 in the telomeric region and SMN2 in the centromere, differs by just one nucleotide. 90% of SMN2 transcripts lack exon7.5-8 as a result of this C>T mutation in the SMN2 exon7 coding sequence, which has an impact on splicing.

Because of this, the SMN2 gene generates an isoform that is shorter, unstable, and easily destroyed as opposed to SMN1, which generates full-length SMN protein.

The involvement of SMN in RNA splicing suggests that splicing errors might cause a disease-relevant transcript or transcripts to be disrupted (isoform specific to motor neuron function). Although SMN is expressed in all bodily tissues, this putative specialisation may help to explain why motor neurons seem to be the sole cells impacted by SMA. This hypothesis is backed up by the observation that in SMA mice models, phenotypic severity closely correlates with biochemical tests of SMN's capacity to assemble Sm proteins onto short nuclear RNA.

Clinically, SMA can be identified by the atrophy and weakening of the muscles brought on by motor neuron loss and malfunction. The majority of the time, weakness is proximally dominant and symmetric. The disorder can range in severity from little proximal limb weakness in adults to severe widespread weakness and respiratory failure in newborns. More often than upper limbs, lower limbs are affected, and bulbar and respiratory weakness is more frequent in situations of severe limb weakness.

The onset and progression of weakness differ from that of many other motor neuron disorders in that, with the exception of the most severe cases (type 0), there is typically a presymptomatic phase followed by rapidly progressing functional loss and a later relatively static phase with slow progression.

Using gene therapy to replace the SMN1 gene, the first highly effective treatments for SMA in murine models were described in 2010–2011. Similar encouraging outcomes have been observed with antisense oligonucleotide therapy that can change SMN2 splicing to incorporate exon7 and boost production of full-length SMN protein 120,124-126.

The pathogenic mechanism of SMA, whereby reduced levels of SMN protein lead to the selective death of motor neurons, is still unknown, despite great advancements in our understanding of the biological effects of SMN decrease. Nonetheless, preclinical research has led to the creation of a number of SMN-restoring treatments that have demonstrated striking efficacy in animal models of the disease. Several of these candidates are now being investigated in early-stage human trials.

The necessity of early SMN restoration and the diminishing benefits of late rescue in mice models have also been frequently shown in preclinical research. 120-123 Due to the lack of animal models with milder forms of SMA, it is unclear if extremely late restoration of SMN protein has any discernible impact on motor neuron function and whether the timing of therapeutic interventions will be the same in varying degrees of SMA.

TARGET PROTEIN

• SMN PROTEIN

THE TUDOR DOMAIN OF THE HUMAN SMN PROTEIN

Homo sapiens is the organism, Escherichia coli BL21(DE3) is the expression system, and there have been no mutations. 2001-11-02 Date of Deposit 2001-05-02: published

- Deposition Author(s): Selenko, P., Sprangers, R., Stier, G., Buehler, D., Fischer, U., Sattler, M. • Criteria for selection include: structures with acceptable covalent geometry, favourable non-bond energy, fewest restraint violations, and structures with the lowest energy.
- Atom count: 441; total structure weight: 9.8 kDa 56 modeled residues, 88 deposited residues, and 1 unique protein chain were found. (3. C. L. Lorson)

Natural Compounds

FLAVONOIDS

Flavonoids are phenolic compounds found naturally in fruits, vegetables, grains, bark, roots, stems, flowers, tea, and wine. These natural products are well known for their health-promoting properties, and efforts are being made to isolate the flavonoids.

Flavonoids are anti-cancer, antioxidant, anti-inflammatory, and antiviral. They are also neuro protective and cardio protective. The type of flavonoid, mode of action, and bioavailability all influence these biological activities.

They are classified according to their chemical structure, degree of unsaturation, and carbon ring oxidation. Anthoxanthins (flavanone and flavone), flavanones, flavanones, flavans, chalcones, anthocyanins, and bioflavonoids are the various subgroups of flavonoids.

GM09677 cells were chosen as the more responsive line to study the effects of 20mM quercetin treatment (the most effective concentration on SMN transcript level). After 24 hours, 48 hours, and 72 hours of quercetin treatment, SMN protein levels were determined using a Western blot. In contrast to real-time RT-PCR studies, no differences in SMN protein levels were detected at any time point in comparison to untreated control samples.

Curcuma

Turmeric is a genus of plant in the Zingiberaceae family that includes seeds of turmeric and Siamese tulips. Its natural habitat is Southeast Asia, southern China, the Indian subcontinent, New Guinea, and northern Australia. [3] Some species are said to have been naturalized in other temperate regions of the world, including various islands in tropical Africa, Central America, Florida, the Pacific, Indian and Atlantic Oceans. Most turmeric grows in loose, sandy soil in shade.

Curcumin therapy for neurological disorders such as spinal muscular atrophy, Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis, and multiple sclerosis has only recently attracted the attention of researchers and the general public. That's what I mean. (14. S. Lefebvre et al.)

Withania somnifera

Withania somnifera, also known as Ashwagandha, has been used in Ayurvedic medicine since ancient times. It is classified as a 'Rasayana' herb for its adaptogenic and rejuvenating properties. Due to the plant's diverse properties, both root and leaf extracts have been used to treat a variety of conditions, including cancer, anxiety, inflammation, and neurological disorders.

The potential neuroprotective role of WS in scopolamine-induced amnesia and reported that WS protects brain-derived cells against amnesia The researchers discovered that WS prevented scopolamine-induced down regulation of BDNF and GFAP (glial fibrillary acidic protein) expression in a dose-dependent manner. Furthermore, WS leaf extract has been shown to effectively protect brain-derived cells from amnesia and glutamate stress by up regulating activity-regulated cytoskeletal-associated protein.[23] (23. U. R. Monani)

Tools

- **PUBCHEM**

PubChem is a database of chemical molecules and their biological activities. The National Centre for Biotechnology Information (NCBI), a division of the National Library of Medicine,

. Each result includes links to other NCBI databases, such as PubMed, as well as details on synonyms, chemical properties, chemical structure (including SMILES and InChI strings), bioactivity, and structurally related compounds.

By placing the field name in square brackets before the search term, you can perform a text search on the database fields. A numerical range is represented by two numbers separated by a colon.

- **PYMOL**

PyMOL is a proprietary but open-source molecular visualisation system that was created by Warren Lyford Delano. It was first made available for purchase through DeLano Scientific LLC, a private software company devoted to developing practical tools that are widely available to scientific and educational communities.

One of the few free model visualisation tools for structural biology is PyMOL. The word "Py" in the software's name indicates that it was created using the Python programming language.

In order to solve Poisson-Boltzmann equations, PyMOL makes use of the OpenGL Extension Wrangler Library (GLEW) and Free GLUT. PyMOL had native Aqua binaries for macOS through Schrödinger and used Tk for its GUI

widgets, but with the release of version 2.0, a PyQt user interface was introduced across all platforms.

The Python License applied to the earliest releases of PyMol. August 1st, 2006 saw the introduction of a controlled-access download system by Delano Scientific for its precompiled PyMOL builds (including betas). Currently, only registered users who have purchased these executables are able to access them; students and teachers can still access educational builds for no cost. Even so, the majority of the most recent source code and older precompiled builds are still freely accessible.

• AUTODOCK VINA

A brand-new tool for molecular docking and virtual screening has been unveiled: Auto Dock Vina. In its local optimization process, Vina uses an advanced gradient optimization technique. The optimization algorithm effectively receives a "sense of direction" from the calculation of the gradient after only one evaluation. By making use of multiple CPUs or CPU cores, Vina can accelerate execution even more.

METHODOLOGY

1. Select the target protein.
2. Preparing the target protein by downloading it from PDB, opening it in pymol, adding the necessary hydrogen, and removing water and ligand.
3. Docking is accomplished with the Autodockvina tool.
4. We can obtain the score of our target protein with the plants selected and flavonoids using this docking tool.

Using this docking tool, we can obtain the score of our target protein with the selected plants and flavonoids.

RESULT

1:

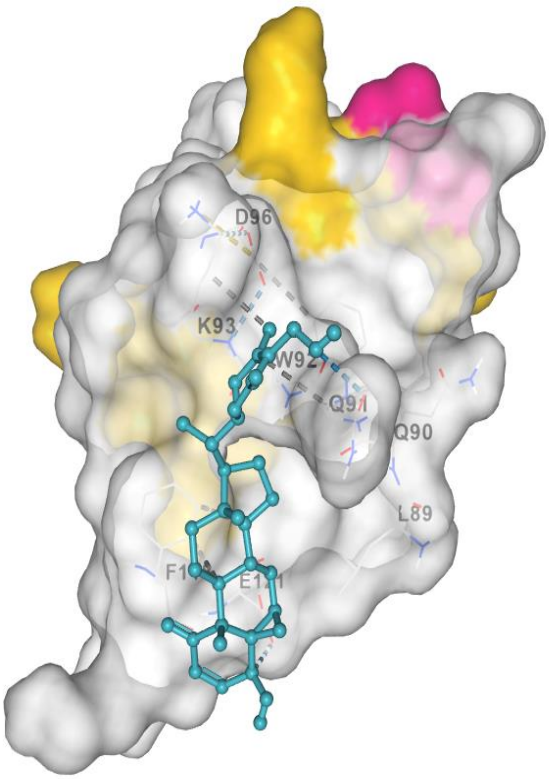

Fig 5- SMN-ashwagandha dock complex

Vina score	Cavity size	Center			Size		
		x	y	z	x	y	z
-6.2	55	-7	-20	0	25	25	25
-6.1	47	-6	-2	-1	25	25	25
-6	14	4	-18	-3	25	25	25
-5.9	18	-7	-15	7	25	25	25
-5.7	63	2	-11	8	25	25	25

2:

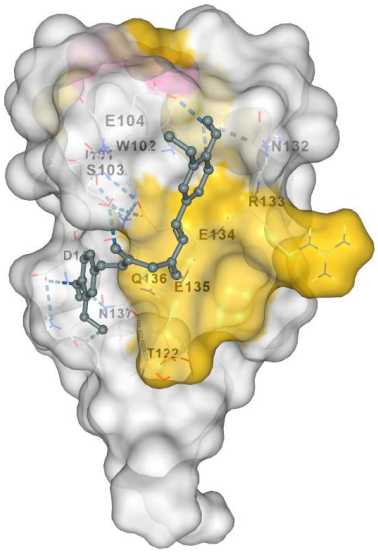

Fig 6- SMN-curcumin dock complex

Binding Modes Download a

Vina score	Cavity size	Center			Size		
		x	y	z	x	y	z
-5.9	63	2	-11	8	26	26	26
-5.9	47	-6	-2	-1	26	26	26
-5.4	14	4	-18	-3	26	26	26
-5	18	-7	-15	7	26	26	26
-4.9	55	-7	-20	0	26	26	26

3:

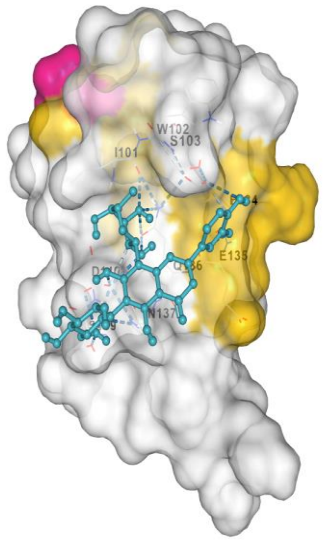

Fig 7- SMN-Flavonoids dock complex

Binding Modes Download all

Vina score	Cavity size	Center			Size		
		x	y	z	x	y	z
-5.9	63	2	-11	8	24	24	24
-5.8	18	-7	-15	7	24	24	24
-5.5	55	-7	-20	0	24	24	24
-5.5	14	4	-18	-3	24	24	24
-5.3	47	-6	-2	-1	24	24	24

DISCUSSION

Vina Score: An empirical scoring function computes the affinity, or fitness, of protein-ligand binding by aggregating the contributions of several individual terms.

Ashwagandha Vina's score is -6.2
Curcumin vina score is -5.9
Flavonoids vina score is -5.9

CONCLUSION

Although neurodegeneration is the main pathology in SMA, there is mounting evidence from clinical reports and animal studies that other tissues play a role in the overall phenotype, particularly in the most severe forms of the disease. Patients may also experience metabolic deficiencies, liver, pancreatic, and intestinal dysfunction, as well as autonomic nervous system involvement and congenital heart defects. In SMA type and healthy cells, there are variations in the expression of antioxidant genes. Our research on oxidative stress and neuronal development after curcumin therapy prompted this idea. Curcumin, an antioxidant supplement, may be used as a treatment option for SMA pathogenesis. Despite not increasing SMN protein levels, flavonoids significantly influenced SMN2 and mRNA expression levels in fibroblast cells. For this reason, more research is being done. The effects of quercetin on neurons, which are more susceptible to degeneration than other tissues in SMA, as well as the effects of quercetin on SMN expression that are specific to particular cells or tissues, must be studied.

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