cryobio

Additional Agreement to Use Donor CB 493

I, (_____ (Recipient) and _____ (Partner, if applicable)), specifically request and accept frozen semen from Cryobio donor CB 493. I understand that this Additional Agreement is an additional part of the Sperm Use Agreement specific to donor CB 93. CB 493 had expanded genetic carrier screening to determine their carrier status for 502 recessive genetic conditions. Please note that Cryobio thoroughly evaluates each donor's results and assesses potential risks of any identified results before allowing donors to remain in our donor program.

I have reviewed genetic test results on this sperm donor, and I understand that donor CB 493 has been found to be a carrier of the following recessive genetic conditions:

Gitelman syndrome;

Nephrotic syndrome (*NPHS2*-related)/Steroid-resistant nephrotic syndrome;

Neuronal ceroid-lipofuscinosis (*CLN3*-related); and

Ornithine aminotransferase deficiency.

Purpose of genetic carrier screening: Carriers of genetic conditions have changes, called pathogenic variants or mutations, in a specific (or multiple) gene(s). Most of the genetic conditions that the Cryobio donors are tested for are inherited in an autosomal recessive pattern (see Figure 1). Typically, we all have two copies of every gene---one from the egg source and one from the sperm source. Autosomal recessive conditions require a mutation in both copies of the same gene in order for it to cause the condition. Therefore, individuals who carry just one mutation in a gene that causes recessive genetic conditions are 'carriers' of that specific condition. Carriers of most of the genetic conditions Cryobio donors are tested for do not typically show symptoms of the condition, i.e., they are asymptomatic, although there are rare exceptions. Most individuals are carriers for at least one if not multiple recessive genetic conditions.

Carrier status is helpful to know because if both the egg source and the sperm source are carriers for pathogenic variants or mutations in the same gene, then there is a 1 in 4 chance of the resulting child having that specific condition; a 2 in 4 chance of the resulting child being a carrier for that specific condition; and a 1 in 4 chance of the resulting child being neither a carrier or having that specific condition. Some of the conditions Cryobio donors are tested for have genotype-phenotype correlation, meaning that specific genetic pathogenic variations (the genotype) in a specific gene can be predictive of the type/specific features of a condition that may present in the individual (the phenotype), but not all do. Additionally, some of the genes can be linked to dominant conditions, meaning having a mutation in just one gene may increase the risk of a specific condition. If a specific change in a gene is linked to a dominant condition, it will be noted in this consent form.

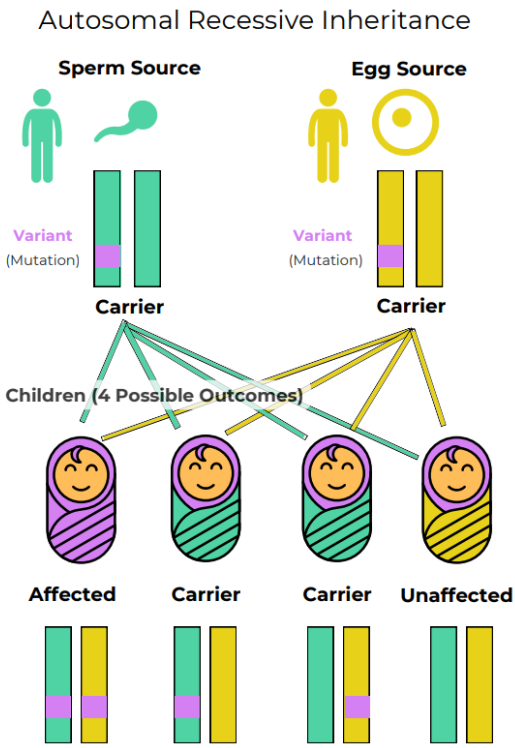

Figure 1. Graphic representing autosomal recessive inheritance of two carrier gametes.

Gitelman syndrome (*SLC12A3* gene): Gitelman syndrome is an autosomal recessive, pan-ethnic condition caused by pathogenic variants in the gene *SLC12A3*. In this condition, the kidney does not retain necessary particles/ions, causing an imbalance in the body. Symptoms usually begin in late childhood or adolescence, and include muscle spasms or cramps, tingling sensations, joint pain, and fatigue. Most patients have mild symptoms, but severe ion imbalances could lead to seizures or heart arrhythmias. With treatment, including dietary management, patients have a normal life expectancy. It is not currently possible to predict the severity of symptoms based on the variants inherited. Studies have suggested carriers of Gitelman syndrome may have a lower blood pressure compared to that of the general population. However, carriers are not expected to have clinical problems or symptoms of the condition.

Of note: There has been one case report of the possibility of digenic inheritance in an individual with Gitelman syndrome. Digenic inheritance means genetic variants in two *different* genes combined cause a condition. In this case report, it is *suspected* that one *SLC12A3* and one *CLCKNB* variant were the cause of disease. However, the pathogenicity of the *CLCKNB* gene could not be confirmed. More evidence is needed to conclude digenic inheritance as the cause of Gitelman syndrome. As an extra precaution, recipients should consider testing the recipient (or egg source if different) for *CLCKNB* carrier status in addition to *SLC12A3* carrier status.

Nephrotic syndrome (*NPHS2*-related/Steroid-resistant nephrotic syndrome (*NPHS2* gene): Nephrotic syndrome (*NPHS2*-related), also known as steroid-resistant nephrotic syndrome, is a genetic condition caused by pathogenic variations or mutations in the *NPHS2* gene. It is inherited in an autosomal recessive inheritance pattern. Individuals from all ethnicities can be carriers for the condition. Onset is usually during childhood or adolescence. Symptoms include loss of protein in the urine, which results in progressive kidney failure. Death will occur without a kidney transplant, usually by adolescence; however, many patients are cured after kidney transplant. Several specific variants may be associated with an earlier or later age of onset of the condition, but not all variants are known to have a genotype-phenotype correlation.

Of note: The specific variant identified in donor CB 493 (p.R229Q) is a common pathogenic variant which is known to cause disease only when in combination with specific variants in the *NPHS2* gene, but it does not cause disease with *all* other variants of the *NPHS2* gene. Regardless, if the egg source were also identified to be a carrier of a *NPHS2* variant, we strongly encourage and recommend review of the information with a genetics professional to better understand the risks of disease to children. Carriers are not expected to show symptoms of this disease.

Neuronal ceroid-lipofuscinosis (*CLN3*-related) (*CLN3* gene): Neuronal ceroid-lipofuscinosis (NCL) is a term used to refer to a group of 13 genetic lysosomal storage disorders that primarily affect the nervous system. The 13 different subtypes are classified primarily based on their underlying genetic cause and vary in symptoms and age of onset. The genetic changes that cause NCLs disrupt the cells' ability to dispose of certain wastes. In NCLs, buildup of these specific wastes occurs most dramatically in nerve cells (called neurons) of the brain, but also in other tissues of the body, such as the eyes. This buildup of these wastes causes deterioration of the brain, neurological impairment, seizures, eye disease, and other symptoms that may vary by subtype. Neuronal ceroid-lipofuscinosis (*CLN3*-related) is the form donor CB 493 is a carrier for, and it is caused by pathogenic variants in the gene *CLN3*. NCL (*CLN3*-related) is the most common form of NCL and is also known as/referred to as Batten disease.

NCL (*CLN3*-related) is inherited in an autosomal recessive pattern, and it has been reported in patients from different ethnicities around the world. Most *CLN3*-caused neuronal ceroid-lipofuscinosis results in

a juvenile form, in which symptoms begin between 4 and 10 years of age. Clinical features include progressive visual loss which proceeds to blindness in childhood. Neurologic and psychiatric symptoms include seizures, difficulty speaking, intellectual disability, psychosis or dementia, and ataxia (loss of full control of body movements) leading to an inability to walk. Affected individuals often die between the ages of 20 and 40. Some patients have a milder form with less neurologic involvement.

Currently, there are medications to help treat symptoms of NCL (*CLN3*-related) (for example medications are available to help reduce/treat seizures, physical therapy to assist with ataxia, etc.), but there is no available treatment to cure the underlying condition itself. Severity of the condition can vary widely, and it is not currently possible to predict the severity for each individual with NCL (*CLN3*-related).

Ornithine aminotransferase deficiency (*OAT* gene): Ornithine aminotransferase (OAT) deficiency (also known as gyrate atrophy) is an autosomal recessive disorder caused by pathogenic variants in the gene *OAT*. While affected individuals have been reported worldwide, the condition has an increased prevalence in Sephardic Jewish individuals from Iraq and Syria and individuals of Finnish descent due to the presence of founder mutations.

The *OAT* gene codes for a protein that aids in protein metabolism. When the *OAT* gene is not functioning properly, it leads to a buildup of a molecule in the body called ornithine. Buildup of ornithine is thought to result in the symptoms/features of OAT deficiency. Clinical features include the onset of night blindness and myopia (near-sightedness) in the first decade of life, with progression to blindness in adulthood. Cataract development usually occurs in adolescence or early adulthood. While most individuals with OAT deficiency have no other symptoms than vision loss, others may present with muscle abnormalities/muscle weakness or other symptoms. Life expectancy and intelligence are typically normal. Vitamin supplementation and dietary restriction to limit ornithine build up may aid in slowing the progression of vision loss. There is currently no way to predict severity of the condition based on the genetic variants present in affected individuals.

Carrier status frequency: Carrier status frequency is the chance of an individual being a carrier for a genetic condition, based on general population risks or based on ethnicity, prior to any genetic screening. If an individual tests negative as a carrier for a condition or conditions, then the chance of being a carrier is significantly decreased. There is still remaining risk called residual risk. Residual risk means the risk of being a carrier even after negative genetic testing for a condition. Residual risk data on the conditions the donor tested negative for can be requested from Cryobio. The carrier frequency provided is from the test provider. As with all genetic information, these carrier frequency numbers may change over time, and may slightly vary from lab to lab depending on the data used to curate them. Therefore, the carrier frequencies from this additional agreement are based on the numbers available from the performing laboratory on the date the donor's test results were reviewed by the lab.

Gitelman syndrome (*SLC12A3* gene) carrier status frequency in different ethnicities, from SEMA4:

Worldwide	1 in 82
African	1 in 138
Ashkenazi Jewish	1 in 121
East Asian	1 in 28
Finnish	1 in 239
European (Non-Finnish)	1 in 73
Native American	1 in 131
South Asian	1 in 145

Nephrotic syndrome (*NPHS2*-related)/steroid-resistant nephrotic syndrome (*NPHS2* gene) carrier status frequency in different ethnicities, from SEMA4:

Worldwide	1 in 16
African	1 in 77
Ashkenazi Jewish	1 in 9
East Asian	1 in 528
Finnish	1 in 7
European (Non-Finnish)	1 in 13
Native American	1 in 36
South Asian	1 in 18

Neuronal ceroid-lipofuscinosis (*CLN3*-related) (*CNL3* gene) carrier status frequency in different ethnicities, from SEMA4:

Worldwide	1 in 434
African	1 in 1697
East Asian	1 in 589
Finnish	1 in 1722
European (Non-Finnish)	1 in 242
Native American	1 in 1538
South Asian	1 in 2552

Ornithine aminotransferase deficiency (*OAT* gene) carrier status frequency in different ethnicities, from SEMA4:

Worldwide	1 in 595
African	1 in 2898
Ashkenazi Jewish	1 in 614
Finnish	1 in 138
European (Non-Finnish)	1 in 749
Native American	1 in 1291
South Asian	1 in 905

Recommendation: Cryobio recommends that the recipient, or egg source if different than recipient, be tested for Gitelman syndrome (*SLC12A3* gene and consider *CKCNB* gene); nephrotic syndrome (*NPHS2*-related)/steroid-resistant nephrotic syndrome (*NPHS2* gene); neuronal ceroid-lipofuscinosis (*CLN3*-related) (*CLN3* gene); and ornithine aminotransferase deficiency (*OAT* gene) carrier status and consider genetic counseling. Please contact Cryobio with any questions or to arrange genetic counseling. Genetic counseling services can also be found through the National Society of Genetic Counselors. We also strongly recommend that you discuss the donor's genetic carrier status results with your health care provider. Finally, we recommend that any future child be notified of this donor's carrier status once they are of reproductive age, as even if they do not have a recessive disease, they could be a carrier and their carrier status could help them identify risks related to their own reproductive future.

Cryobio has advised me of the following:	Please initial to show your understanding and agreement:
The donor I have chosen has positive results from genetic testing looking at carrier status for 502 genes. These results indicate that the donor is a carrier for Gitelman syndrome; nephrotic syndrome (*NPHS2*-related)/steroid-resistant nephrotic syndrome; neuronal ceroid-lipofuscinosis (*CLN3*-related); and ornithine aminotransferase deficiency.	Initials: _____ Initials: _____

The genetic conditions tested for are inherited as recessive patterns. This means that if both the egg source and the sperm source are carriers for the same condition, there is a significantly higher chance of the resulting child having that genetic condition.	Initials: _____ Initials: _____
By the donor testing positive for carrier status for Gitelman syndrome; nephrotic syndrome (*NPHS2*-related)/steroid-resistant nephrotic syndrome; neuronal ceroid-lipofuscinosis (*CLN3*-related); and ornithine aminotransferase deficiency the risk to a resulting child would now be higher than that of the general population.	Initials: _____ Initials: _____
When an individual tests negative for carrier status, it does not completely eliminate their chance of being a carrier for that condition, however their remaining risk is greatly reduced. This remaining risk is called residual risk, and the residual risk can vary significantly from person to person. For more detailed information regarding the sensitivity of testing and remaining risk after negative screening, please contact Cryobio.	Initials: _____ Initials: _____
As genetic testing evolves and more data becomes available, I understand that there is the possibility of updated genetic information that may be uncovered for this donor for a variety of reasons. It is my responsibility to check back with Cryobio to see if any new genetic information is available for this donor.	Initials: _____ Initials: _____
Genetic testing for me (or the egg source, if different) can also be done to better understand and further reduce the risk to offspring.	Initials: _____ Initials: _____
Genetic testing is *strongly recommended* for me (or the egg source, if different) to see if I am a carrier for Gitelman syndrome (*SLC12A3* gene and consider *CLCKNB* gene); nephrotic syndrome (*NPHS2*-related)/steroid-resistant nephrotic syndrome (*NPHS2* gene); neuronal ceroid-lipofuscinosis (*CLN3*-related) (*CLN3* gene); and ornithine aminotransferase deficiency (*OAT* gene).	Initials: _____ Initials: _____
Expanded genetic carrier screening is continuing to evolve, and at the time this donor entered the program this was the screening available. This donor had genetic testing with SEMA4 in 2022. My health care provider may recommend an expanded carrier screen that includes/included more than the 502 genes screened for in this donor. It is my responsibility to share this information with my health care provider and review the risks and benefits of being screened for more (or fewer) genetic conditions.	Initials: _____ Initials: _____
The genetic testing done on the donor does ***not*** screen for all known genetic conditions.	Initials: _____ Initials: _____
While genetic testing can lower the likelihood for certain genetic conditions, no amount of genetic testing can guarantee that a child will be free of all genetic conditions.	Initials: _____ Initials: _____

Genetic counseling is available to me if I have additional questions regarding these test results and potential risks.	Initials: _____ Initials: _____
Both the donor's carrier status and whether the donor is acceptable for my use should be discussed with my health care provider.	Initials: _____ Initials: _____

I have read the above material and assume the risk of using donor sperm from a donor who has been found to be a carrier of genetic conditions. I am making the choice to use donor sperm from donor CB 493 willingly and agree to release any legal claims, including negligence, that may arise from or are related to insemination or assisted reproduction using donor sperm from donor CB 493.

I have read and had the chance to ask questions, and I understand and agree to the terms of this Additional Agreement to use donor CB 493.

_____ _____

Recipient Date Email

_____ _____

Partner, if applicable Date Email

William C. Baird, PhD, HCLD 04-06-2022

Cryobio Date