Whole Exome Sequencing of a Family with Local Anesthetic Resistance

Steven Clendenen, M.D.1, Steven Porter, M.D.1, Christopher Robards, M.D.1, Nathan Clendenen, M.D.2
1 Department of Anesthesiology, 2 Yale Department of Anesthesiology
Mayo Clinic Florida, Jacksonville, FL

Introduction

Local anesthetics (LA) work by blocking sodium conductance through voltage gated sodium channels and failure to produce blockade is uncommon. The cause of such resistance is unknown, although genetic variation has been proposed as a potential mechanism. A patient presented for surgery involving her upper extremity with a history of LA failure. A nerve block of the brachial plexus was performed with ultrasound guidance with injection of 30 ml of 1.5% mepivacaine with a complete failure of the block. (Figure 1)

Aims

We hypothesized that LA resistance is due to a variant form of voltage gated sodium channel and we test this hypothesis by performing whole exome sequencing for a family with 3 of 4 family members with LA resistance. (Figure 2)

Methods

• Whole Exome Sequencing
The patient and their immediate family provided informed consent for DNA sequencing and they were screened with a questionnaire to identify family members with a history of LA resistance. A whole blood sample was collected and sent for exome sequencing using the Illumina HiSeq 2000 platform and analyzed using the Genome Analysis Toolkit.

• Genetic data analysis
Exome sequencing results for four individuals were referenced to the 1000 Genomes Project and the NHLBI ESP to identify variants associated with local anesthetic resistance, present in less than 1% of the general population, and located in functional regions of the genome.

• Identifying Nav 1.5 in Peripheral Nerves
To determine whether Nav 1.5 is present in peripheral nerves we performed immunohistochemistry with highly specific antibodies on healthy human peripheral nerve tissue. The staining revealed Nav 1.5 staining throughout the cell body and along the peripheral nerve fiber (Figure 3A) similar to Nav 1.7 (Figure 3B).

Results

Whole exome sequencing of the family members was performed to identify genetic variants shared by the three individuals with LA resistance but not present in the unaffected family member. Genetic variants were identified, only one of which was identified in a voltage gated sodium channel susceptible to LA inhibition (Nav 1.5). (Figure 1)

Conclusions

Resistance to local anesthetics in humans has been documented in the literature and we identified a genetic variant in the gene encoding for Nav 1.5 that is associated with LA resistance. We demonstrate that Nav 1.5 is present in human peripheral nerves to support the plausibility that an abnormal form of the Nav 1.5 protein could be responsible for the observed local anesthetic resistance.

Figure 1
Ultrasounographic image of the patient’s brachial plexus in contact with local anesthetic, which is visible as the hypoechoic fluid-tissue interface surrounding the needle.

Figure 2
Ultrasoundographic image of the patient’s brachial plexus in contact with local anesthetic, which is visible as the hypoechoic fluid-tissue interface surrounding the needle.

Figure 3A
Human peripheral nerve stained with Nav 1.5 antibody.

Figure 3B
Human peripheral nerve stained with Nav 1.7 antibody.