Expert Shreds Dolphin Meat In Canned Tuna DNA Research

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Recently, the Mexican tuna industry was dumbstruck by the news that research by a local university found dolphin meat inside cans of tuna sold domestically. *Atuna* spoke with the researchers who clarified that what their study revealed was the presence of dolphin DNA and not meat in the products. However, an expert on the scientific method that the University used in the research has found several anomalies in the methodology which cast serious doubt on the validity of the results.

Enrique de la Vega, a molecular biologist with over 15 years of experience with PCR testing, told *Atuna* that a series of essential steps were skipped by the student and her supervisors in charge of the research, which could explain the reported findings.

“Without these steps, it's impossible to reach the conclusion that the dolphin DNA was present in the cans tested without a shadow of a doubt,” said the scientist.

The research was carried out by the National Autonomous University of Mexico’s (UNAM’s) Food Engineering student, Karla Hernández, for her Bachelor's thesis. She was supervised by Dr. José Francisco Montiel Sosa, a Professor at the educational institute. In an interview with Atuna last month, they revealed that the research could not conclude that dolphin meat was present in can tuna but just dolphin DNA.

The most crucial mistake found by de la Vega was the fact that there was no human DNA control. This is an imperative step in PCR “since the most common source of contamination in DNA testing comes from the scientists in charge of the extraction and subsequent amplification.” Thus, it is highly probable that the testing also processed human DNA, which without the proper control could result in false positives.

On top of this, the student performed different extraction methods on the shark, pork, bovine, strawberry, and tomatoes than on the tuna samples. For the former, she used chloroform and for the latter a DNA extraction kit. Hence, it is possible that the false positive only occurs in the tuna sample and not in all the other samples.

“With these very important differences in the method of DNA extraction between samples, it is not possible to make a valid comparison to justify the lack of contamination during processing,” said de la Vega.

TEMPERATURE

The molecular biologist continued by highlighting that there were also important issues to consider such as the annealing temperature used in the PCR testing - the temperature at which the test is performed. According to him, this is the most important factor in guaranteeing the results of this analysis.

He explained that every set of primer (DNA sequence) has a very limited range of optimal annealing temperature at which “it sticks to the complementary genetic sequence of the DNA one is evaluating.” Changes of just one degree Celsius can have vast effects on the results.

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The annealing temperature used for the PCR with tuna primers was almost three degrees Celsius lower than what was recommended by the kit. Because of this, the first tests with the tuna primer were not very specific and gave positive results for shark and dolphin. However, the temperature was not adjusted but rather the student classified the primers as not specific, which ended up misinterpreting the results of the test.

Furthermore, de la Vega found more temperature slips, in particular, in the test that concluded that there was dolphin DNA in the canned tuna samples. Again, a temperature of two degrees below the recommended level was used which gave "a high probability of generating non-specific hybridizations and therefore false positives."

MORE IRREGULARITIES

The abnormalities in the study do not end here. The PCR expert also found some irregularities concerning the way the testing was carried out that increased potential false positives.

He mentioned how the student decided to use PCR with 40 cycles of amplification, which is the process through which this method tests DNA matches. This number is a massive amount of series, and well above the normal 30 cycles for this type of analysis. "The more cycles you do the higher the chance of a false positive."

"To me, the faint bands found in the analyses in which dolphin DNA was present after 40 amplification cycles and with a fairly low annealing temperature suggests more a non-specific amplification than confirmation of dolphin meat in tuna cans," said de la Vega.

Finally, he concluded that an imperative step that must be taken when results such as these are obtained was completely overlooked. And that is to test the positive bands and demonstrate that they are in fact dolphin DNA. "Unfortunately, in this case, this crucial step was omitted and without which, it is impossible to conclude the presence of dolphin meat in the tuna cans."

Overall, de la Vega believes that while he understands that this was a Bachelor thesis, the supervision for proper PCR test was lacking. Essential scientific steps were not included in the methodology used for the research, making it impossible to even claim that dolphin DNA was found in tuna cans, much less dolphin meat.

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