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|  | * Results:  After trying two different procedures and several sample runs with only our DNA and Mr. Thiel�s DNA, we have come up with no results. We are inclined to believe that the reason for no results lies within the school�s PCR machine. When we started collecting data, we decided to get all our samples and freeze them until ready to PCR. Well, once we got the samples and tried running our first sample run through the PCR machine and gel electrophoresis and then stained the gel, we looked to see our results and saw a blank gel. It was a little upsetting. So, we figured maybe we did something wrong in staining the gel. We went online again, printed out a staining procedure for ethidium bromide, and did another test run with new samples of Mr. Thiel�s cheek cells and our cheek cells. Well, this didn�t work either and we didn�t see any bands. Maybe it wasn�t dark enough� so we went into a storage room and shut off the lights to try to see the bands in the gel under UV light without any other ambient light. Still no bands visible. Then we tried a second procedure which used a toothpick swipes instead of a saline solution to suspend cheek cells. Too bad this didn�t work either. But with this procedure, there was Marker DNA that did not go through the PCR process of the procedure, which we were able to see in the gel. We were able to see these bands distinctly. This told us that our staining procedure was fine, but that there was something wrong with either the PCR procedure (annealing and denaturing times) or that we did something wrong before that. So to find out the exact problem, we called Maria Abilock, who provided us with the procedures and reagents needed, for some help. Through talking with her, we decided on a time when the two of us, plus Mr. Thiel, could come down to PE Biosystems and try our experiments in one of their labs. When we went down there, we got two samples for both procedures from all four of us, and left one set of samples there and one set of samples with us back to school. At school, we ran the samples in the PCR and in the gel while Maria also ran the samples, but in the PCR machines at PE Biosystems, and then ran the gel. At school, we again had no results, but Maria did come up with bands in her gel. Here is a picture of her gel:  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | | * 1 | * 2 | * 3 | * 4 | * 5 | * 6 | * 7 | * 8 | * 9 | * 10 | * 11 | * 12 | * 13 | * 14 | * 15 | * 16 | * 17 | * 18 |  |  |  * Since she got results and we did not, and both of us did the exact same things before the PCR process, it is a problem with the PCR machine at school that is the cause of us not getting any results.  The bands in the gel represent the following: Lane 1 100 bp marker 2 Maria�s +Control (550 bp, 850 bp plus lots of non-specific amplification) 3 Maria�s �Control (no DNA) 4 Maria�s Saline (homozygous 550 bp) 5 Maria�s toothpick (homozygous 550 bp) 6 Maryann�s +Control 7 Maryann�s �Control 8 Maryann�s Saline (550 bp, 850 bp) 9 Maryann�s Toothpick (550 bp, 850 bp) 10 Thiel�s +Control 11 Thiel�s �Control 12 Thiel�s Saline (homozygous 550 bp) 13 Brian�s +Control 14 Brian�s �Control 15 Brian�s Saline (550bp, 850 bp) 16 Brian�s Toothpick (appears to be homozygous 550 bp) 17-20 Mitochondrial DNA that Maria was testing and didn't want to pour a separate gel for. 17-20 Mitochondrial DNA that Maria was working with * Conclusion: Since the many procedures we have used have produced no DNA bands in our agarose gels our results are inconclusive. Once we find a procedure that will work with the equipment we are using we hope to see results similar to those Cold Springs Harbor Laboratories has been getting. But with the results we did get from working with Maria Abilock, we conclude that the saline solution procedure works better than the Toothpick swipe procedure. This can be seen in Brian�s lanes of DNA, where in lane 15 is Brian�s saline solution sample and it is visible that both bands are there, and that he has and Alu fragment on one chromosome, but not on the other. In lane 16 is Brian�s toothpick swipe sample where only one band can be seen. Other than that, though, we have no conclusions since our results were nonexistent. * Recommendations: * Find a procedure where the materials are easily accessible if something goes wrong. * Find a procedure for the PCR machine that works and will provide results. * Write down everything you do and the results from each step as in a journal. Analyze in the journal the results so the write-up is easier. * Find someone or some business that will help out; for example, a place where you can do the procedure with newer equipment than the stuff at school. Also, at a different site, there might be less contamination of data. * Always wear goggles, aprons, and gloves whenever working in the lab with this project and similar ones. The main reasons for this is for safety and for prevention of contamination. * Start at least 4 months before science fair or the due date in case something with your project goes wrong. * Leave at least a month to do the write up. | |
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