A Case of Identity

"As you may see, my dear Watson, I have rounded up our line of suspects and they all appear to be black."

"Phenomenal observation, Holmes. You're holding a cage full of black mice. How is that helping us?"

Holmes put the cage down on the nearby table. "Surely not a phenomenal observation as their phenotype is all the same, yet it's their genotype we're interested in here."

"You have lost me, my friend."

"Phenotype means physical trait observable from the outside, Watson. You and I share neither hair colour nor stature, our phenotypes are quite different. Yet these mice all look the same to you, don't they?" He pointed into the cage which contained an agitated mass of tenebrous fur. I nodded in consent.

Holmes turned to the table and set up several microtubes. "However, many differences in the genomic information don't present themselves in changed outer appearance, for example slightly different lengths of the same protein." He retrieved one of the mice from the cage, holding it by its tail. "So, my dear Watson, to settle this case and find out if the genetically engineered mouse, which was stolen from our client, seemingly by Mr Windibank and hidden in this cage of ordinary black mice, we have to genotype them for the altered genomic information will spring out."

"And how do we get the DNA, the genomic information?" I asked.

"By this," he smiled, cut a miniature piece from the mouse's tail and let it fall into one of the microtubes. He marked the mouse and the microtube accordingly and proceeded in the same fashion with the rest of the flock.

Fascinated I watched until he'd finished. "And now? This can't be all."

"Well, we could go the fast way, add water and boil the tails to get DNA but I prefer a more thorough approach called DNA extraction. We'll add buffer to keep the pH stable and an enzyme called Proteinase K to digest the tissue and let it shake overnight at a cosy 60°C."

"Overnight!" I exclaimed. "And then we'll have the mice's DNA?"

"After several additional steps of purification to get rid of the superfluent tissue, we can extract the DNA by adding alcohol."

"Charming. So we have to wait until tomorrow for the results?"

"Yes, you can yell at science as much as you like, but you can't rush it."

Early next morning, Holmes dragged me to St Bartholomew's again, giddy with excitement. While I suppressed yawns, for Holmes had played

violin all night, he himself danced around the laboratory until finally he showed me ten empty microtubes.

"This is it?" I frowned.

He nodded apologetically. "The DNA pellet is hard to see." He poured another liquid into the tubes. "This will solve the pellet and we can proceed to the next step: the PCR. Polymerase chain reaction," he added sighing, noticing my befuddled look. "Very helpful tool to amplify small bits of DNA into numerous copies. Since we're only interested in one short part of the DNA, the one that has been genetically modified in our client's mouse but not in the ordinary ones, this is the way to go. Our client, Mr Sutherland, has given me the primers we'll need."

"Primers?"

"Primers are short sequences of nucleic acids, the same material DNA is made of. They serve as starting points for the copying mechanism. For PCR, you need two primers that are specifically designed to detected unique parts of the DNA so that just what lies between those primers is duplicated. Of course that's a several step process too."

While explaining, Holmes had scribbled down notes and mixed together more translucent liquids. He produced ten smaller tubes and meticulously pipetted so little into each tube, I couldn't see it from my place. "This is Mastermix, dear Watson, it contains everything to produce those multiple copies of the DNA we're looking for, for example Polymerase, another enzyme, acting as the copier and dNTPs, single units of DNA, like different coloured inks." He placed the little tubes in another machine, pushed some buttons and with a low hum the machine came alive.

"That's a thermal cycler, it now runs the several steps I've mentioned in repeating cycles. DNA consists of two complementary strands, but Polymerase can just copy DNA with a single strand. This first step is called Denaturation, splitting a double into two singles. The second step, Annealing, is where the primers find their position on the DNA and the third step, Elongation, is the actual duplication step. Through repetition of these steps thirty to forty times you'll get massive amounts of the DNA fragment which length is determined by our primers compared to all the other fragments that are in this mixture."

After another endless wait the machine stopped, Holmes took out the tubes and self-absorbed prepared another liquid, this time something colourful, and again mixed it with the samples. Then, to my astonishment, he pipetted the samples onto what looked like a thick sheet of gelatin in a tub of water.

"What's that?"

“It’s a gel, on which we can separate the different lengths of the DNA fragments we just produced with the PCR by size. Smaller fragments run faster through the gel than larger ones, so we will see different bands. I’ve stained the DNA so we can later see it under the UV light. Oh, and before you have to ask, the negatively charged DNA runs through the gel because we apply an electric field to it.” He fiddled with what looked like starter cables.

After another wait, Holmes dragged me in the dark room and switched on the UV light. “We have a winner,” he joyously exclaimed after a few seconds and pointed to one of the lanes. Indeed, all lanes were empty safe for lane five with one thin band.

“So this is …,”

“ … the mouse which was stolen from Mr Sutherland, yes. This is our proof, Watson. Let’s call Lestrade.”