Dna and rna in blood cells

Leukocytes or white blood cells protect the body in various ways against disease. Five different types of white blood cells exist in the blood and are made from a multipotent cell in the bone marrow known as the hematopoietic stem cell. The five different white blood cells are often classified according to one major distinguishing factor, the presence of granules; white blood cells are often characterised as granulocytes or agranulocytes.

Granulocytes are white blood cells that are characterised by the presence of different staining granules in their cytoplasm when viewed under light microscopy. There are three types of granulocytes: neutrophils, basophils, and eosinophils.

Agranulocytes are white blood cells that are characterised by the absence of granules in their cytoplasm. The cells include lymphocytes and monocytes.

Introduction of nucleic acids

Definition of nucleic acids

Nucleic acids are macromolecules which are polynucleotide. A nucleotide consists of a nitrogenous base, a pentose sugar and a phosphate unit. The two most common nucleic acids are Deoxyribonucleic acid (DNA) and Ribonucleic acid (RNA).

DNA is a long polymer with a double helix structure consisting of a deoxyribose sugar and phosphate backbone and four different bases: adenine, guanine, cytosine and thymine. It is found in the nucleus and plays the role of a medium for long-term storage and transmission of genetic

information. The genetic information is used in the development and functioning of all known organisms.

RNA is a polymer with a single stranded structure consisting of a ribose sugar and phosphate backbone and four different bases: adenine, guanine, cytosine and uracil in placed of thymine. There are different forms of RNA each have different functions: messenger RNA, ribosomal RNA and transfer RNA. The main job of RNA is to transfer the genetic code need for the creation of proteins from the nucleus to the ribosome. This process prevents the DNA from having to leave the nucleus, so it stays safe.

Back ground of nucleic acids

Nucleic acids were first discovered by Johannes Friedrich Miescher by isolating various phosphate-rich chemicals, which he called nuclein (now nucleic acids), from the nuclei of white blood cells in 1869 at Felix Hoppe-Seyler’s laboratory at the University of Tübingen, Germany, Dahm R (Jan 2008).

Theory

Nearly all the DNA is contained within the nucleus of the cell, with the exception of mitochondrial DNA (mtDNA), while RNA can be found around the cytoplasm of the cells. Methyl green-pyronin was used as a stain as methyl green has a preferential affinity for DNA which stains green. Similarly pyronin has preferential affinity for RNA which stains rose-red. The higher the intensities of the colours would indicate a higher concentration of the nucleic acids.

Aims of the experiments

There are three main aims of the experiment. Firstly, it is to observe the locations of the DNA and RNA in the cell by using methyl green-pyronin on the white blood cell. Secondly, to classify the white blood cells into two groups, granulocytes and agranulocytes. The last of the main aims is to observe the effects of DNase and RNase on DNA and RNA respectively. Since the experiment involves the use of a little blood, the experiment aims to prepare blood smears and safe science being practiced as body fluids (blood) are being handled.

Hypothesis of the experiments

For the slide that was stained only with MGP, the nucleus of the cell is expected to be stained rose-red and green while the cytoplasm will be stained rose-red. Granulocytes and agranulocytes are expected to be observed on the slide.

The slide that was treated with RNase and stained with MGP, on the other hand, is expected to have a nucleus stained green only. Granulocytes and agranulocytes are also expected to be observed on the slide.

Results of the experiment

The results of the experiment are as shown in Table 1

As the results shown, the MGP slide, (the control slide) was stained with both green and rose-red also the green stained was found to be concentrated at the nucleus of the cell while the rose-red stain was found around the cytoplasm. This confirms that there are DNA and RNA present on the slide.

The MGP + RNase slide however was stained green only. This would indicate the presence of DNA, which was stained green by the methyl green, and the absence of RNA which was supposed to be stained red by the pyronin.

Although under microscopic observation, both granulocytes and agranulocytes were observed. However, further classifications of the granulocytes and agranulocytes into their specific types (eosinophils, basophils or neutrophils) were not made as they were undeterminable from this practical.

Discussions of the results obtained

Explanations of the obtained results

The slide that was only stained with MGP contained both DNA and RNA, which can be concluded from the fact that the cell was stained green in the nucleus and rose-red in the cytoplasm. As no RNase was introduced to the slide, the RNA remained on the slide instead of being degraded.

On the other hand, the slide that was treated with RNase and stained with MGP only contained DNA, as inferred from only the green stain in the nucleus of the white blood cell. This is due to the fact that RNase, an enzyme, catalyses the degradation of RNA. Therefore, no RNA was present on this slide.

Agranulocytes and granulocytes were both observed on both slides. Since blood contained white blood cell, both agranulocytes and granulocytes would be observed. However, more granulocytes was observed in both slides, which can be explained by comparing the highest concentration between the granulocytes and agranulocytes. Among the granulocytes, neutrophils have

a higher concentration than compared with the other granulocytes (Eosinophils and basophils) which make up for a range of 54%-62% of the white blood cells. Compared with the agranulocytes which has the higher concentration among other agranulocytes (Lymphocytes with the range of 25%-33% of the white blood cells), it can be concluded that more granulocytes would be observed than agranulocytes.

Comparison between actual and theoretical results

As observed at the results, most of the hypothesised results were proven correct as the results support the hypothesis. However, there are differences between the actual results and the theoretical results.

In theory, the green stain, DNA can be found in the cytoplasm of both slides. Although not as concentrated as those found in the nucleus, green stains were still expected to be observed on the cytoplasm of the cell as mtDNA would stain green.

Also, the red stain, RNA was expected to be found in the nucleus of the cell on the MGP slide. The red stain on the nucleus was expected to be less concentrated than those observed on the cytoplasm. In the nucleus, RNA does exist and was expected to be stained red.

Possible sources of errors

Firstly, to dry the blood smear, a hair dryer was used to speed up the heating process. Although by using a hair dryer, the process would indeed be sped up, but it may also overheat the blood smear. This would cause cracks to form as the blood smear was overheated and damaging the cell. In this experiment, it is important to retain the morphology of the cell as much as

possible. Instead of using increasing values of ethanol concentration to dehydrate the water, 95% ethanol was used. This would result in incomplete dehydration of the water in the smears and hence would result in problems such as artifacts such as air bubbles being observed in the smears, affecting the overall result. As the results shown, mtDNA was not observed in the actual experiment. (If mtDNA was observed, certain areas in the cytoplasm of the cell would be stained green) This might be due to the fact that xylene was not used. Xylene would provide a clearer background for the cells to be observed. However, it was not used as it is carcinogenic.

Suggestions to overcome errors

Instead of using a hair dryer to dry the blood smears, the smears could be left in the air to air dry them. This would prevent the blood smears from overheating and cracking and yet it allows the blood smears to dry naturally which helps to retain the cell morphology. Next, the blood smears could be dehydrated by placing them in increasing ethanol solutions. For example, 50% ethanol; 75% ethanol; 90% ethanol; 95% ethanol; 100% ethanol. This would ensure that there would not be any artifacts originating from water (for example air bubbles) to affect the results. Also, xylene could be used to aid in a clearer observation of the slides.

Conclusion

On the whole, the experiment was successfully carried out as most of the actual results tally with the hypothesis. Both granulocytes and agranulocytes were also observed. It was also observed that RNase did degrade RNA and the staining were observed. Although, not all of the expected results was observed, there are several methods that can be done or steps to be

changed to obtain the hypothesized result. Safe science was also practiced when handling body fluids.