Case Study – Bowel Screening Cellular Pathology

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Introduction

A 56-year-old asymptomatic female participating in the UK national bowel cancer screening programme tested positive via faecal immunochemical testing (NHS, 2021). Subsequent diagnostic procedures, including colonoscopy and biopsy, confirmed adenocarcinoma necessitating a right hemicolectomy, followed by histological examination that revealed no further invasion and no need for further treatments.

Clinical Details

This patient is a 56-year-old female who did not present with any noticeable signs or symptoms. The was no family history of colorectal cancer. The patient was identified through the UK national bowel cancer screening programme (McKigney & Coyne, 2020) where a faecal immunochemical test detected traces of blood in the sample, indicating further testing was necessary (D'Souza et al., 2019). A colonoscopy was performed, and it was noted that suspicious tissue was present. It was originally thought to be possible pre-cancerous cells or a polyp, and an endoscopic mucosal resection was attempted to try and remove the tissue ((Hwang et al., 2015). This was unsuccessful as the tissue would not lift from the colon wall- indicating it could be invasive. A biopsy was taken and sent for histological testing which revealed that the mass was adenocarcinoma. CT and MRI scans were undertaken to attempt to grade the progression of the disease. This ultimately led to the decision to perform a right hemicolectomy to remove the tissue. This was sent to the histology laboratory to pathologically grade the disease and establish if there was lymphatic or vascular invasion.

Tests

A right hemicolectomy was performed, removing the terminal ileum, caecum, ascending colon, proximal transverse colon, appendix, and a portion of omentum (Mandava & Mitchell, 2023) to eliminate the tumour and surrounding affected tissue. Histological analysis aids in evaluating the tumour's characteristics and prognosis (Chen et al., 2021).

The specimen was dissected and processed to dehydrate the tissue and impregnate it with a medium to support the tissue structures (Aziz & Zeman-Pocrnich, 2021). It was then thinly sliced (4 microns thick) using a microtone and the sections were placed into a flotation water bath to smooth out any wrinkles in the wax. The sections were then picked up using a slide before being stained. The slides were stained with haematoxylin and eosin which caused the basophilic structures, such as nuclei, to turn blue, and the acidophilic structures to turn red, such as cytoplasm and cell membranes. This staining method helps identify tissue structures under a light microscope, assisting the assessment of disease progression (Al-Sabaawy et al., 2021), including lymphatic, vascular, perineural, and local invasion depth (Loughrey et al., 2023).

Following recommendations from the National Institute for Health and Care Excellence (Loughrey et al., 2023), an immunohistochemistry test was performed to test for DNA mismatch repair (MMR) which indicates Lynch syndrome- a gene mutation that can cause colorectal cancer. It is hereditary and can therefore be used to screen family members of patients positive for Lynch syndrome.

Results

The results of the slide analysis following a haematoxylin and eosin stain are shown below in Table 1. The results of the immunohistochemical testing for DNA mismatch repair are shown below in Table 2.

Table 1: A table to show the observations noted for defined characteristics when viewing the stained sample slide under a light microscope.

Characteristic	Observations	Clinical Report Code
Differentiation	Well/Moderate	G1/2
Local invasion depth	Beyond muscularis propria	pT3
Involvement of all margins	Margins free	R0
Number of lymph nodes involved	1	N1a
Apical lymph node involvement	No	N/A
Vascular invasion depth	Extramural	V0
Lymphatic invasion depth	Extramural	L0
Perineural invasion depth	No invasion detected	Pn0

Table 2: A table to show whether a protein was identified as present or absent following immunohistochemistry testing.

The results shown in Table 1 lead to a description of ‘colorectal adenocarcinoma G1/2 T3 N1a R0 V0 L0 Pn0’. These observations are sufficient in determining the progress of the disease and establishing if further treatment is needed. The code R0 indicates that the circumferential and longitudinal margins are free from invasion, indicating that the cancer has not spread further than the local area identified. Furthermore, the codes ‘L0’, V0’, and ‘Pn0’ indicate that the disease has not significantly spread into the lymphatic system, vascular system, or the surrounding nerves. This is supported by the lack of involvement of the apical lymph node.

Table 2 shows that all proteins associated with the DNA mismatch repair mutation are present indicating that the disease is unlikely to be caused by Lynch syndrome, negating the need for further BRAF testing.

Diagnosis and Discussion

This patient has already received a diagnosis of colorectal adenocarcinoma following a biopsy. It is the third most common malignancy and occurs in the glandular tissue of the bowel, often starting as a polyp or legion before forming a cancerous tumour (Lotfollahzadeh et al., 2023). The histology testing in this case aims to assess the progression and prognosis of the the disease and determine if the right hemicolectomy was successful in being curative as intended. To do this, each observed characteristic must be assessed.

Protein	Outcome
MSH2	Present
MSH6	Present
MLH1	Present
PMS2	Present
BRAF	Not tested

Firstly, the differentiation of the tumour cells should be defined. In this case, the tumour cells are defined as well to moderately differentiated, giving it a clinical grade of 1 / 2. This grade has a relatively good prognosis compared to poorly differentiated cells. Well to moderately differentiated cells are more like healthy cells and do not show as much change as poorly differentiated cells. Poor differentiation is linked to faster proliferation of cells and therefore, faster growth and more aggressive disease with a poorer outcome (Qi et al., 2022).

The depth of local invasion should be assessed also. The further the tumour has invaded into the local area, the poorer the prognosis. This case shows that the tumour has invaded beyond the muscularis propria and into the subserosa, giving it a clinical code of pT3. Although there is significant depth to the invasion, the outcome of pT3 tumours is notably better than that of pT4 tumours that have breached the serosa and possibly extended into neighbouring organs (Baguena et al., 2019).

It is noted that the circumferential and longitudinal resection margins were free from disease involvement. Balbaa et al. (2020) indicate that the involvement of these margins is associated with a higher likelihood of distant metastasis as well as disease recurrence following a right hemicolectomy, suggesting that this case has a lower likelihood of these issues occurring.

Lymphatic and vascular invasion are other notable factors that are known to increase the chances of distant metastasis as well as suggest if the right hemicolectomy was unsuccessful at being curative and if further treatments are needed, such as chemotherapy. Studies have shown that lymphovascular invasion in colorectal cancer cases is linked to a substantially lower 5-year survival rate and an increased risk of death (Jiang et al., 2019). The apical lymph nodes, found between the origin of the left colonic artery and the inferior mesenteric artery, are strong indicators of the involvement of the lymphatic system. Studies have shown that the rate of distant metastasis is much higher in patients who have apical lymph node involvement compared to those who do not (Ishii et al., 2022). This suggests that this patient is

unlikely to have distant metastasis as they do not have apical lymph node involvement. Furthermore, the patient has a better prognosis due to the observations showing that, although one lymph node is affected, the lymphatic and vascular systems have not been invaded.

As per the National Institute for Health and Care Excellence recommendations (Loughrey et al., 2023), immunohistochemistry testing was performed to assess if the patient has Lynch syndrome. Lynch syndrome is an autosomal dominant hereditary condition that increases the risk of developing certain cancers, such as colorectal cancer, and lowers the average age of development. It is caused by a variant in one or more of the four mismatch repair genes – MSH2, MSH6, MLH1, and PMS2. If the testing reveals a mutation or absence of one of the gene-coded proteins, BRAF testing is performed (Tanakaya, 2019). This patient was shown to have all four proteins present, indicating that they are not suffering from Lynch syndrome and further BRAF testing is not necessary. If the patient was positive for Lynch syndrome, their family members would be offered testing and, if positive, they would be offered regular cancer screening due to their increased risk of development.

The prognosis of this patient is good due as the tests reveal they are unlikely to have distant metastasis and lymphovascular involvement. Furthermore, the resection margins are free from involvement indicating that all the affected tissue was removed by the right hemicolectomy. This shows that the procedure was curative, and the patient does not require further treatment, however, it is recommended that follow-up blood tests, CT and MRI scans, and a colonoscopy be performed in 5 years-time to confirm the disease has not returned.

Research has shown that traditional right hemicolectomy procedures can be adjusted in the future to include complete mesocolic excision (CME) - possibly improving long-term outcomes for patients. CME increases the number of lymph nodes removed from the areas during surgery and has been shown to increase absolute survival rates for colorectal cancers from 81% to 89% whilst also lowering the rate of disease

recurrence. It has been shown that CME is not any less safe than a normal right hemicolectomy, however, it is a significantly more technical operation which could mean it may be less readily available as a routine procedure. Due to this, it has not become standard practice, however, with further clinical technological advancements, it could be a possibility (Anania et al., 2021).

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Appendices

Figure 1: A photograph showing the right hemicolectomy specimen during dissection.

Figure 2: A photograph showing the dissected sections of the specimen.

Figure 3: A photograph showing the dissected sections placed within cassettes ready to be processed.

Figure 4: A photograph of assisting the identification of lymph nodes with a trained member of staff.

Figure 5: A photograph taken during the embedding process.

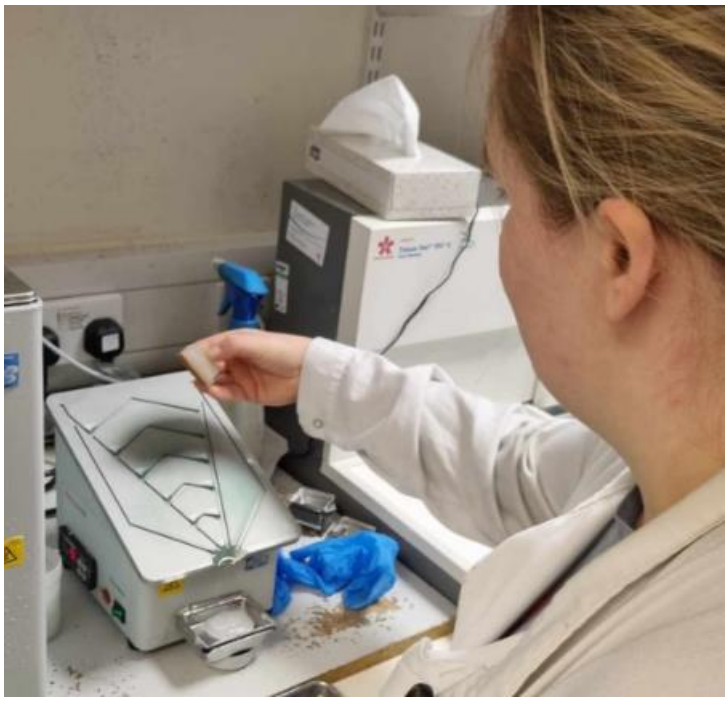

Figure 6: A photograph showing the paratrimmer being used to removed excess wax.

Figure 7: A photograph showing a trained member of staff using the microtone.

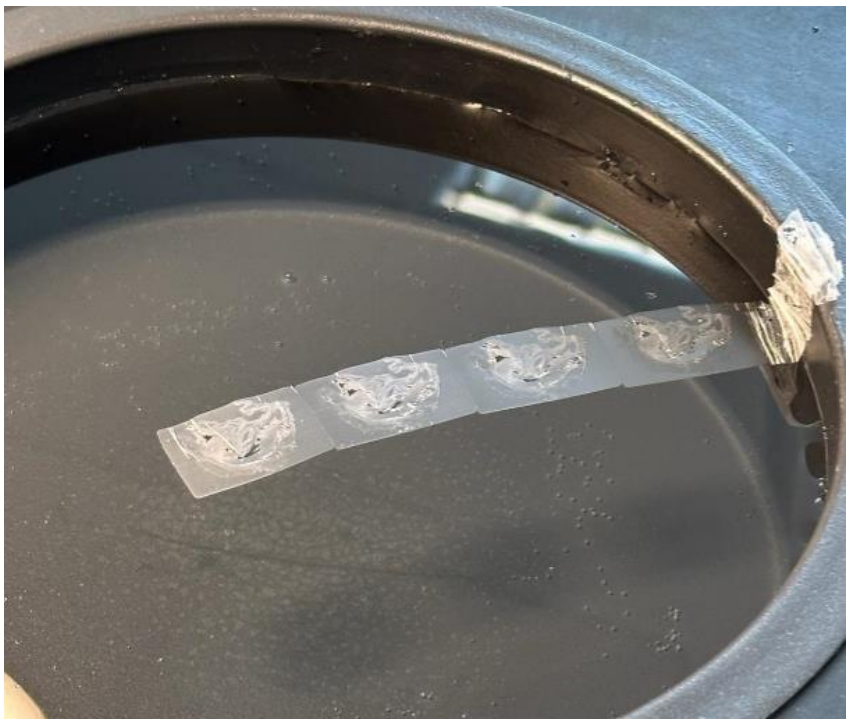

Figure 8: A photograph showing the sections of sample floating in the water bath.

Figure 9: A photograph showing a section being picked up on a slide.

Figure 10: A photograph taken during the staining process.

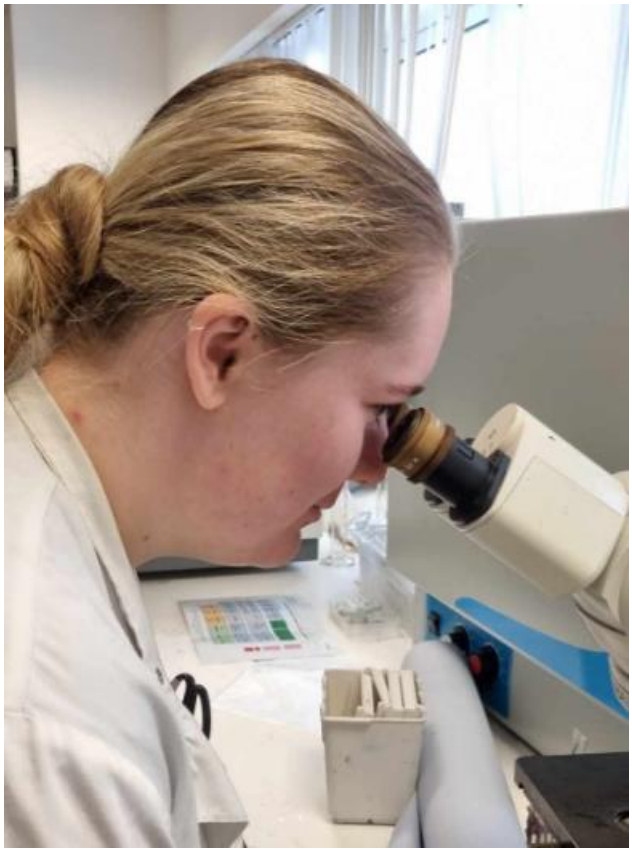

Figure 11: A photograph taken whilst using the light microscope to ensure the staining process worked as expected.

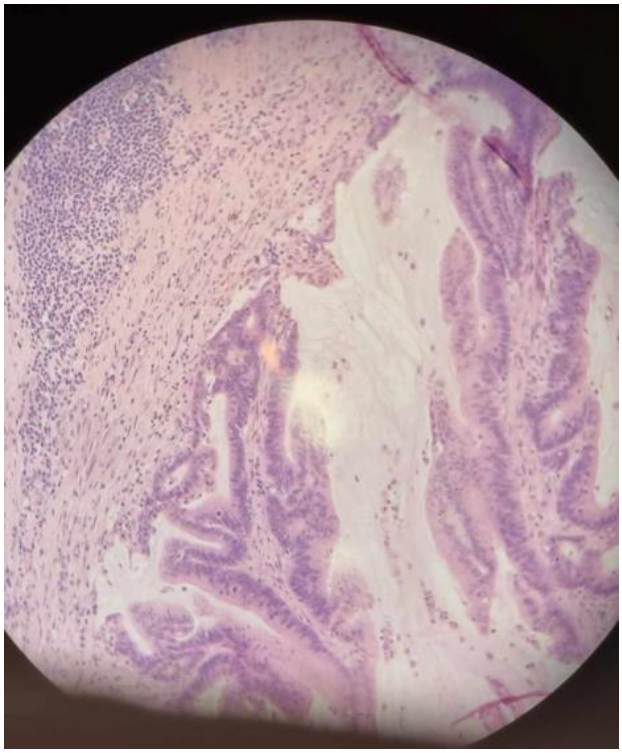

Figure 12: A photograph taken down the light microscope showing the tumour tissue.

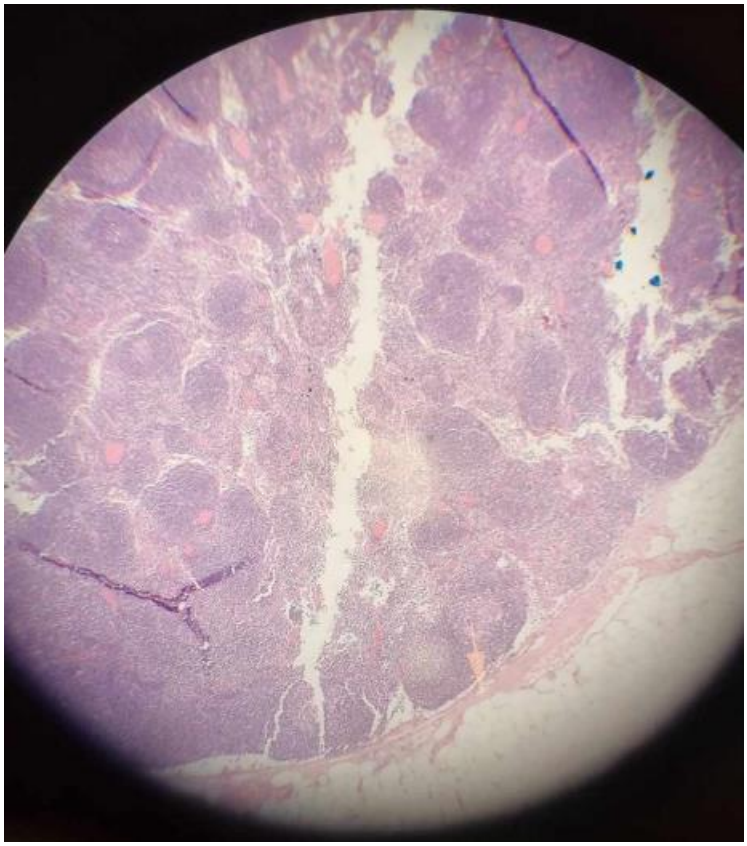

Figure 13: A photograph taken down the light microscope showing the non-affected apical lymph node.

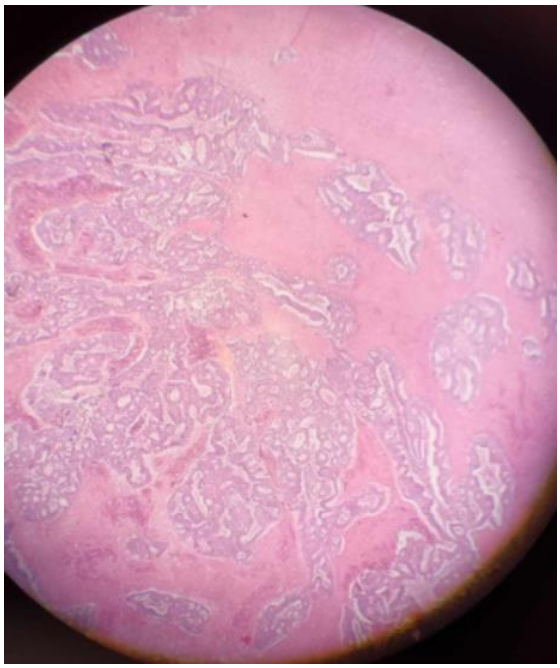

Figure 14: A photograph taken down the light microscope showing the invaded lymph node.