Figure 1. Biomarker confirmation frequency of biomarker use for diagnostic confirmation (cd20, Ki67, c-myc, cd10, bcr, cd30, cd15, Mum1, pax5, cd45, desmin, myogenin, cd3, WT-1, cd99).

Figure 2. Impact of the diagnosis changes between H&E and IHC concordance between conventional pathology and IHC.

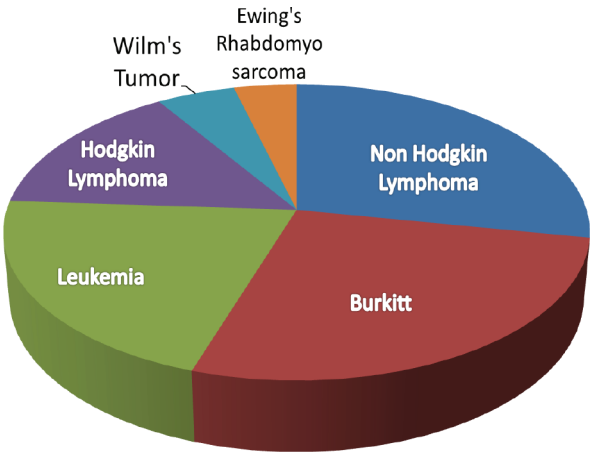

Figure 3. Paediatric cancers diagnosed at BMC annually the most common paediatric cancer convectional pathology diagnosis at BMC (*N* = 105).

Table 1. Annually cancer diagnosis between H& E stain and IHC comparison of paediatric cancers impacts for diagnosis at BMC annually (*N* = 105).

Comparison (H&E versus IHC)	Cancer/tumour diagnosed	Frequency (*n*(%))
Total concordant Dx, 55 (51%)	Leukaemia	16 (29%)
	BL	12 (22%)
	HL	10 (18%)
	Carcinoma	7 (12%)
	Sarcoma	6 (11%)
	Fibrosis	2 (04%)
	Inflammation	2 (04%)
Total specific (additional Dx required), 18 (17.6%)	ALCL	7 (39%)
	DLBCL	5 (27%)
	LL	3 (17%)
	FL	3 (17%)
Total changed Dx, 32 (31.4%)	BL	8 (25%)
	Sarcoma	6 (19%)
	Necrosis	6 (19%)
	Leukaemia	5 (16%)
	Inflammation	4 (12%)
	Carcinoma	2 (06%)
	Fibrosis	1 (03%)

Discussion

IHC is critically important for the accurate diagnosis of paediatric cancer, with over 30% of all cases identified as having treatment-changing diagnoses among our cohort. The changes in diagnosis improved the outcome for the treatment of choice at BMC, the management regime was completely altered from the confirmatory diagnosis of IHC from MNH. A case example from confirmed acute leukaemia cases from flow cytometry results from MNH resulted in a complete remission and prevented the relapse of the disease to the paediatric confirmed diagnosis of specific acute leukaemia, the confirmed lymphoma cases of Hodgkin and non-Hodgkin lymphoma, and inflammatory reactions considered malignancy at BMC are managed accordingly. IHC training would be important to include in capacity development for paediatric cancer programmes in low-resource settings. Most common discordant NHL (especially ALCL, DLBCL), the main reason can be the presence of many sub-types of NHL; hence, specificity on the exact type of NHL without biomarkers becomes difficult meaning difficulties in distinguishing types of NHL on convectional histology (H&E staining).

A higher discordance resulting from the complete changes in diagnosis between the two diagnostic techniques from the BMC and MNH facilities is mainly attributed to missing IHC diagnostic reagents that could provide a significant range for specific and accurate diagnosis. Advanced tools for flow cytometry provide the best room for confirmation of leukaemia cases for accurate diagnosis scores and minimal residual disease. These are the reasons for the discordance in diagnosis between the two techniques and facilities. A study done by Jaffe *et al* [9] shows difficulties of NHL classification on histological appearance (H&E staining) alone not to be a reliable indicator and encouraged the use of advanced diagnosis markers (IHC, immunophenotyping).

A study done by Tran *et al* [10] shows there is high specificity in the diagnosis of Hirschprung's disease using marker calretinin IHC compared to (H&E staining) on the rectal suction biopsies on frozen embedded tissue. Pileri *et al* [11] showed the identification and classification of neoplasm using different IHC markers CD68, Lysozyme, CD21, CD35 and S100 protein in the classification of these neoplasm high sensitivity and specificity rate was observed.

The importance of accurate paediatric cancer diagnosis is crucial at the beginning of the treatment to avoid further complications on the paediatric cancer advancement, poor prognosis and finally death due to mismanagement attributed to inaccurate diagnosis.

A study done by Proctor *et al* [12], an important expert shows discordance rate variation between 3.6% and 34.1%; the study demonstrated the importance of review in accurate diagnosis and timely lymphoid management.

We found that a limited common biomarker panel can successfully be used to confirm diagnosis among the paediatric cancer population. WHO recommended, on the common panel that 78 different paediatric cancer genes be identified by biomarkers, all 15 biomarkers present in this study were identified from the WHO paediatric cancer identification panel.

A study done by Hayes *et al* [13] from a report on AHOPCA Pathology, still reveals the struggle made in the anatomical pathology of LMICs. The training was offered a 5-day pathology training work to Pathologist and Histo-technologists from various LMICs of the Caribbean region. Assessment was made in the review and evaluation of the quality of IHC slides produced after the training course. A comparison was made between the training slides and the original sides from the institute, 5 days of training slides appeared effective. However, it is part of quality improvement and validation in the development of IHC in LMICs. Ottmann *et al* [14] on the detection of a mitotic figure in thin melanoma. IHC does not replace Hematoxylin and Eosin stain. A study found that comparing several mitotic figures has a slight variation which is not significant. IHC for mitotic figures could not replace H&E's careful evaluation of slides. IHC detection is only an additional tool. A study was done by Nielsen *et al* [15] on Proliferation indices of phosphohistone H3 and Ki67 strong prognostic markers I/II melanoma. The finding from the study was phosphohistone H3/MART and Ki67/MART were strong and effective to Hematoxylin and Eosin stain, this presentation is similar to the study done at BMC on the impacts of IHC compared to H&E on paediatric cancer patients, these two IHC stains seem to be a robust alternative to conventional mitotic detection by H&E stain in melanoma. Study done by Mezheyeuski *et al* [16] the results were compared with those of conventional IHC, and related to corresponding RNA-sequencing expression values. We found a strong correlation between the visual and digital quantification of lymphocytes for CD45RO (correlation coefficient: r = 0.52), FOXP3 (r = 0.87), CD4 (r = 0.79), CD20 (r = 0.81) and CD8 (r = 0.90) cells. The conclusion was, that the fluorescence multiplexed IHC method, based on only one tissue sec-tion, provided reliable quantification and localisation of immune cells in cancer tissue.

The application of this technique to clinical biopsies can provide a basic characterisation of immune infiltrates to guide clinical decisions. The 30% (30 children out of 100) discordance diagnosis difference between the two techniques (H&E and IHC) is a discouraging result because having lower efficacy on a true finding could bring a proper and specific treatment regime. The discordant is not acceptable by the World Health Organisation in LMICs where diagnostics tools are limited, places a significant focus on improving access to reliable diagnostic methods and training health care professionals through capacity building.

Conclusion

IHC is critically important to improve diagnostic accuracy for paediatric cancer in lower middle-income Countries based on the 31.4% of the changed diagnosis between IHC and convectional histology (H & E) staining among paediatric cancer patients.

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Conflicts of interest

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Ethical approval

Ethical clearance certification number No. 016/2023. from the joint Catholic University of Health and Allied Sciences-Bugando Medical Centre: Research and Ethics Committee (REC).

Author contributions

JB contributed to data collection for BMC pathology results and Muhimbili IHC results, writing of manuscript JB and KS. OO contributed to the design and implementation of the research, JB, diagnosis for BMC pathology results, OO, JB, diagnosis for Muhimbili IHC results, ET, design and implementation of the research, ET, diagnosis for Muhimbili IHC results, design and implementation of the research, KS, the analysis of the results and to the writing of the manuscript.

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