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Lymphomas in sub-Saharan Africa - what can we learn and how can we help in improving diagnosis, managing patients and fostering translational research?

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Lymphomas in sub-Saharan Africa – what can we learn and how can we help in improving diagnosis, managing patients and fostering translational research?

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Abstract

Approximately 30,000 cases of non-Hodgkin lymphoma (NHL) occur in the equatorial belt of Africa each year. Apart from the fact that Burkitt lymphoma (BL) is very common among children and adolescents in Africa and that an epidemic of human immunodeficiency virus (HIV) infection is currently ongoing in this part of the world, very little is known about lymphomas in Africa. This review provides information regarding the current infrastructure for diagnostics in sub-Saharan Africa. The results on the diagnostic accuracy and on the distribution of different lymphoma subsets in sub-Saharan Africa were based on a review undertaken by a team of lymphoma experts on159 fine needle aspirationsamples and 467 histological samples during their visit to the African centres is presented. Among children (<18 years of age), BL accounted for 82% of all NHL, and among adults, diffuse large B-cell lymphoma accounted for 55% of all NHLs. Among adults, various lymphomas other than BL, including T-cell lymphomas, were encountered. The review also discusses the current strategies of the International Network of Cancer Treatment and Research on improving the diagnostic standards and management of lymphoma patients and in acquiring reliable clinical and pathology data in sub-Saharan Africa for fostering high-quality translational research.

Keywords

lymphomas; malignant haematology; epidemiology

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Introduction

Although accurate estimates are difficult, given the paucity of information, it is likely that approximately 30,000 cases of non-Hodgkin lymphoma (NHL) occur in the equatorial belt of Africa each year and these tumours are among the top ten causes of cancer in this geographical region. The fraction associated with acquired immunodeficiency syndrome (AIDS) is not available, but may be as high as 50%, although it appears to vary markedly throughout the region and is different among children and adults. The incidence of NHL since the AIDS epidemic has increased by 2- to 3-fold in some countries, and as much as 13-fold in others (Kaaya *et al*, 2006; Otieno *et al*, 2001; Orem *et al*, 2004). A realistic estimate of the incidence of Burkitt Lymphoma (BL) is 30-70 per million children in equatorial African countries. Apart from BL, the incidence for other lymphoma subtypes in equatorial Africa remains unknown.

Meaningful research on lymphomas in African cannot be conducted without accurate diagnosis. Unfortunately, striking differences still exist with respect to the reliability of lymphoma diagnosis between developed and developing countries, due partly to the limited number of haematopathologists and partly to the lack of special reagents, such as monoclonal antibodies and molecular probes that are indispensable to an accurate diagnosis based on the World Health Organization (WHO) classification of haematopoietic and lymphoid tissues (Swerdlow *et al*, 2008).

Unfortunately, the resource limitations (human and material) in Africa severely restrict the ability to make an accurate diagnosis early in the disease course and provide adequate treatment and follow-up for patients, thus fostering a belief that therapy is pointless. However, with improved education and relatively simple augmentation of resources, quite simple chemotherapy regimens that require less supportive care are, in fact, feasible in at least major centres in Africa. Under the guidance of the International Network for Cancer Treatment and Research (INCTR) (http://www.inctr.org/), many of the African centres are currently treating lymphomas successfully with protocols specifically designed for the prevailing infrastructure in these centres.

The INCTR team has been working in collaboration with many African partners to achieve:

1. Improvements in the speed and accuracy of diagnosis in Africa.
2. Organization of advanced training of pathologists and technicians from the African centres.
3. Introduction of standard therapy, adapted to the African setting.
4. Developing translational research studies partnering African and European institutions.

To achieve these objectives, the authors, comprising a group of pathologists (along with oncologists), have visited several African centres in teams to assess the infrastructure, audit the diagnoses offered by the African investigators, introduce capacity building measures, provide logistic support for telemedicine and assist with treatment protocols specifically

designed for the African setting. As a follow up, the group has conducted annual educational meetings in Europe attended by African investigators, European partners and other delegates from across the world. Meetings have been held in Italy (Siena – May 2010) and France (Paris – May 2011).

This review will focus on a) the current infrastructure for diagnostics in sub-Saharan Africa, b) the current diagnostic accuracy in the African setting, c) the distribution of different lymphoma subsets in institutions visited in sub-Saharan Africa, d) how diagnostic standards can be improved, e) management strategies for lymphoma patients that are likely to yield the best outcomes in sub-Saharan Africa, f) preliminary results of the INCTR-assisted treatment protocols in sub-Saharan Africa, g) acquisition of reliable clinical and pathology data in sub-Saharan Africa, and h) strategies to foster high-quality translational research.

Infrastructure for diagnostics in sub-Saharan Africa

During our visits, the infrastructure was found to be highly variable both in terms of equipment and available personnel. The annual biopsy numbers varied between 1500 to around 16000 among the cellular pathology departments visited. In addition, some of the departments had a substantial cytology load, in one instance recording a volume of about 5000 cases annually. Fine needle aspiration cytology (FNAC) was widely utilized. The availability of equipment was heterogeneous. Histokinettes for tissue/biopsy processing, rotatory microtome for preparing histology slides, and autostainers were available in some centres (especially those that had collaborations with institutions in developed countries). Some of these also had facilities for cytospin preparations (especially for cerebrospinal fluid) and for preparation of cell-blocks. In some centres, biopsies/tissue samples were processed manually. Formalin was used as the tissue fixative in all the places visited. Haematoxylin & Eosin (H&E) for histology preparation and H&E, Papanicolaou and May Grünwald Giemsa stains for cytology samples were routinely used. In addition, some centres were proficient in other histochemical stains that included Periodic acid Schiff, Grocott's Gordon & Sweet (reticulin), Perl's Prussian blue (iron), Masson Trichrome, Ziehl Neelsen (acid fast bacilli), Masson Fontana, Alcian blue, Mucicarmine, Congo red, Periodic acid methenamine silver and Phosphotungstic acid haematoxylin. Turnaround time (TAT) for histology depended on infrastructural capacity. Among centres with well-established infrastructure, the TAT was less than a week, whereas it was substantially longer in centres with suboptimal capacity.

The quality of histology and cytology preparations was variable. In most centres, the standard was lower than what would be considered optimal in developed countries. However, there were exceptions, where the quality of the H&E stained tissue sections were comparable to the best centres in the world. The suboptimal quality of the material could be attributed to poor fixation (over/under fixation or improperly prepared fixatives), suboptimal tissue processing or suboptimal quality of tissue sectioning. Most of the centres had adequate microscopes. The number of pathologists for the work-load was adequate, although this should not be taken to apply at a national level - the number of available pathologists in Tanzania was reported to be extremely low, and the country lacked pathology trainees. Pathologists in training within the universities of Nairobi (Kenya) and Nigeria were highly

motivated. Recent pathology textbooks and journals were not available in most centres. Despite these drawbacks, pathologists were reasonably well informed.

Although a proportion of the pathologists in sub-Saharan Africa had exposure to centres abroad, the laboratory technical staff had not been trained at centres of excellence. Apart from one privately-funded hospital, all other centres lacked facilities for immunohistochemistry. All local pathologists acknowledged that the lack of immunohistochemistry severely hampered their ability to diagnose and classify lymphoid proliferations accurately.

Diagnostic accuracy in the African setting

A panel of lymphoma experts (varied between 3-5 among different centres) reviewed 159 FNA samples and 467 histological samples during the team's visit to the African centres. The review process was as follows:

Independent review by the experts

Each expert was provided with a microscope and writing material to record their diagnostic impression in each case.

Consensus evaluation

Responses from each of the experts was analysed for consensus in each case. Consensus among two of three, three of four or four of five experts was accepted and recorded as the consensus diagnosis. Where a consensus diagnosis was achieved, cases were not further discussed, unless there was educational value to the host pathologists and pathologists in training. The Remainder of cases were considered discrepant.

Consensus discussion

All discrepant cases and those cases with consensus where the hosts wished to have further discussion were examined during a “consensus discussion.” Cases in which there was disagreement were re-evaluated by the experts. Following the discussion, consensus was achieved by the experts in a proportion of the discrepant cases.

Advice on follow-up investigations for histological samples

Cases with consensus required additional immunohistochemical investigations to conform to the criteria essential for classification based on the current WHO recommendations. Additional testing required was recorded in each case. In cases where a consensus diagnosis was not reached despite discussion, a panel of tests and an algorithm for diagnosis was documented on site.

Follow-up investigations for histological samples

Immunohistochemistry, *in-situ* hybridization (ISH) and fluorescent *in-situ* hybridization (FISH) investigations were undertaken in European institutions under the guidance of Professor L. Leoncini, and a final diagnosis was determined according to the decision-

making algorithm prepared. A few cases required further discussion among the experts during the meeting in Sienna (May 2010).

Among the 159 FNA samples where a diagnosis of Burkitt lymphoma (BL) had been suggested by the pathologists in sub-Saharan Africa, 76% were confirmed to be BL and in an additional 12%, a diagnosis of lymphoma other than BL was suggested. In the remaining cases (12%), the opinion of the panel included – non-diagnostic material; malignancy, but not lymphoma; no evidence of lymphoma or another malignancy.

Among the 467 histological samples that were submitted by the participant centres as diagnostic or highly suggestive of lymphoma, a consensus diagnosis was possible in 393 samples, which included 364 (78%) cases that were confirmed as lymphoma irrespective of the subtype. It was difficult to assess the accuracy of making diagnoses of specific lymphoma subtypes, as lymphoma subtyping had not been attempted in many cases partly due to the fact that basic immunohistochemistry was not available in most centres. A previous study suggested an accuracy of around 75% for the diagnosis of BL in Uganda (Ogwang *et al*, 2011).

Distribution of different lymphoma subsets in sub-Saharan Africa

Cases reviewed

A total of 467 histological specimens were reviewed. This included 132 cases from Lacor Hospital, Uganda; 86 cases from Kenyatta National Hospital, Kenya; 79 cases from Aga Khan University Hospital, Kenya; 92 cases from Muhimbili National Hospital, Tanzania; 44 cases from Obafemi Awolowo University, Nigeria; and 34 cases from University College, Ibadan, Nigeria. Of these cases, 258 (57%) were males, 192 (43%) were females and the gender was not recorded in 17 cases. Age had not been recorded in10 patients. Among the rest, 189 (41.4%) patients were children and adolescents (<18 years age). However, Lacor Hospital was a paediatric hospital and when this institutions' cases were excluded, there was 85/335 (26%) children and adolescents (<18 years age). Among them, the mean and median age was 35.9 years and 36 years respectively.

Among the 188 children and adolescent cases (<18 years age), 114 (62%) were males, 70 (38%) were females and the gender was not recorded in 4 cases. Among the 267 adult cases, 140 (53.5%) were males, 121 (46.5%) were females and the gender was not recorded in 6 cases.

Diagnosis

Among the 467 cases, the panel found 393 cases to be assessable. This included 312 cases of non-Hodgkin lymphoma (NHL) and 52 cases of Hodgkin lymphoma (HL). 11 cases were classified as benign lymph node lesions and 18 cases were confirmed as malignancies other than lymphomas. Among the 188 children and adolescent cases (<18 years age), 160 were assessable, of which 132 were diagnosed as NHL and 19 as HL. Among the 267 adult cases, 172 of 223 assessable cases were considered NHL and 32were diagnosed as HL.

In children and adolescents (<18 years age), BL accounted for 82% of all NHLs. However, other lymphomas such as diffuse large B-cell lymphoma (DLBCL) (7.5%), precursor lymphoid neoplasms and anaplastic large cell lymphoma, ALK positive were also recorded. Among the 19 HL encountered in children and adolescents (<18 years age), all but one case were classical HL (cHL) (Tables 1 & 2).

In adults, DLBCL was the most common NHL, accounting for 55% of all NHLs. However, a wide variety of other lymphomas, such as chronic lymphocytic leukaemia / small lymphocytic lymphoma (9%), BL (9%), plasmablastic lymphoma (4%), follicular lymphoma (FL) (3%) and plasmacytoma (3%) were also noted. Among the 5 cases of FL, 4 were of grade 1-2 and one case had features of grade 3b. Among DLBCL, 2 represented Richter transformation and, one was a cutaneous DLBCL. A component of FL was recognized in 9 DLBCLs. The nodular sclerosis subtype of cHL accounted for more than one-half of NHL cases (Tables 3 & 4).

Clinicopathological correlations

BL was predominantly a disease in children and adolescents. Even after exclusion of cases from the paediatric hospital, among the remaining 42 BL cases, more than 75% occurred in the first two decades of life. The median age for BL was 9 years. Among the 122 cases of BL where surgical biopsies were evaluated, the male:female ratio was 1:1. The more common sites of initial presentation of BL among children were abdomen (24%), jaw (24%), ovary (10%), neck lymph nodes (8%), spleen, liver, pelvis and kidney (~5% each). The more common sites of initial presentation of BL among adults were ovary (20%), parotid and jaw (13% each). It is entirely possible that this distribution pattern was as much due to the method of diagnosis chosen (fine needle versus cutting needle or biopsy) as to the characteristic distribution pattern of the tumour.

DLBCL was predominantly a disease of adults. After exclusion of cases from the paediatric hospital, 92% of the remaining 95 cases occurred in adults. The median age for DLBCL was 50 years. The male:female ratio was 1.5:1. The more common sites of initial presentation of DLBCL in adults were lymph nodes (40%), intestines (8%), stomach (7%), abdomen (7%), sinonasal/tonsillar/nasopharyngeal (6%), oral cavity (4%) and breast (3%).

HL was predominantly a disease of childhood and early adulthood. After exclusion of cases from the paediatric hospital, nearly 75% of the remaining 50 cases occurred before the age of 30 years, with a median age of 22 years. Only 10% of cases occurred after the age of 50 years. The male:female ratio was 1.2:1. This pattern is likely to be at least in part due to the age-structure of the population.

Viral associations

In-situ hybridization for Epstein-Barr virus (EBV) using the EBER-1 probe was attempted on the paraffin blocks. However, in a large proportion of cases, RNA preservation was suboptimal for definitive documentation. Among the interpretable cases EBV-association was documented in 21/27 (78%) of BL, 4/29 (14%) of DLBCL, 6/6 (100%) of plasmablastic lymphoma and 6/7 (86%) of cHL. Other EBV-positive lymphomas seen at lower frequencies included 2 cases of peripheral T-cell lymphoma, unspecified; 1 case of follicular lymphoma,

grade 3b; one case of B-cell lymphoma unclassifiable with features intermediate between DLBCL and BL; one case of B-cell lymphoma unclassifiable with features intermediate between DLBCL and cHL; and 1 case of NK/T cell lymphoma, nasal type. Documentation of HIV association among cases on which histology specimens were reviewed was extremely poor. Among them, a positive documentation of HIV-association was noted in 7 cases of BL, 4 cases of DLBCL, 2 cases of cHL and one case of plasmablastic lymphoma, and a negative documentation of HIV association was noted in 12 cases of BL.

Improving diagnostic standards in sub-Saharan Africa

Although in the developed world, the importance of the correct diagnosis is appreciated as a critical issue, this is still an evolving concept in some of the developing countries, especially in Africa, as observed by members of our group, who have regularly visited East and West sub-Saharan African countries for the purposes of reviewing lymphomas for one or more studies. In particular, there are striking differences in the TAT for lymphoma diagnosis and in the accuracy of diagnosis - differences that have a profound impact on patient outcome. The current problems in the practice of lymphoma diagnosis include basing treatment decisions on FNAC in a large proportion of cases, poor quality histology in a proportion of cases where biopsies are performed, complete lack of immunohistochemistry and other supportive investigations, lack of continuing education, and a failure to apply the current criteria for diagnosis of various lymphoid malignancies. In Africa, a majority of the laboratories still use the Working Formulation for Clinical Usage, a lymphoma classification from the early 1980's, which is based on morphology alone and does not include most of the entities recognized in the last 20 years. Without accurate diagnosis, neither meaningful research projects nor effective patient management can be instituted. Though there are no magic answers for an issue of this magnitude, “twinning” or other forms of communication for teaching purposes between pathologists in institutions in the developed countries and developing countries seems to be the most likely approach to ensuring sustainable improvements. Twinning projects focused on lymphomas would help to improve diagnostic accuracy through capacity building, consultations and both direct and indirect technology transfer.

Therefore, a systematic approach aimed at improving the diagnostic standards through partnerships with institutions in African countries in order to bring them closer to the standards of practice among their European/North American counterparts would be a helpful strategy. This approach would allow the education of collaborating African pathologists with respect to the more recent concepts of lymphoma classification and diagnosis, while introducing essential tools to modern diagnosis such as immunohistochemistry, molecular cytogenetics and other molecular tools. Building local capacity to initiate immunohistochemistry with a limited panel of immunostains would be critical for improving diagnostic standards.

Tele-pathology, through capturing of still-images and their online submission for discussion with experts in the field, is yet another strategy that might lead to improvements in diagnosis. This has been popularized by one of the co-authors (Dr. Nina Hurwitz) whose inputs into the development of the web-based case discussion site iPath hosted through the

INCTR (http://www.ipath-network.com/inctr/) have been tremendous. The facility has had an immense appeal in sub-Saharan Africa, as the provision of a microscope with digital camera attachment to capture still images is fairly inexpensive. Most of the African centres we visited have reasonably good internet facilities that enable them to post images online. The success of this strategy is highly dependent on quality of the histology/cytology preparations and the onsite pathologist's ability to adequately capture the diagnostic and the problematic areas on the slide. The dialogue and the discussion following the submission of images are intended to improve the overall diagnostic standards. Lack of immunohistochemistry support on lymphomas requiring immunohistochemistry could restrict the experts in making a specific and definitive diagnosis on these submissions.

Strategies for management of lymphoma patients in sub-Saharan Africa

Burkitt lymphoma

The clinical syndrome of BL was first recognized in Ugandan children (Burkitt, 1958) and proved to be an important model tumour because it was the first human tumour to be associated with a virus (EBV) and with another common infection in equatorial Africa - malaria. Another important lesson learnt in the pioneering days of chemotherapy was that BL is highly sensitive to, and indeed, curable, by chemotherapy alone (Magrath, 2009). This is fortunate, given that the vast majority of tumours are inoperable at presentation, and surgery may never be curative, while radiotherapy was then and remains extremely limited in availability, although, like surgery, is of limited value in the treatment of BL. It is particularly sad, therefore, that the survival rate of this tumour in equatorial Africa – now over 90% overall in children in Europe, remains very low and not even measurable – even in major centres where this would be possible, the quality of the data collected generally prevents an estimate of the survival rate. A study conducted in Africa in the early 1970's, although not completed, suggested that a simultaneous rather than sequential combination of active drugs, consisting of cyclophosphamide, vincristine and methotrexate (COM) would be more effective at controlling systemic disease (Olweny *et al* 1980), although at the time, effective central nervous system (CNS) prophylaxis had not been developed and a number of isolated CNS relapses were observed. The efficacy of COM in conjunction with effective CNS prophylaxis has never been tested in the African setting because of resource limitations, lack of training in the conduct of clinical trials, abandonment of therapy and poor follow-up. However, a high fraction of major African centres still use a variant of COM therapy. Thus, in spite of the considerable amount of time that has passed since the original study, these results provide a reasonable starting point for the present stepwise approach to the development of BL therapy that is feasible in an African setting where trained oncologists are few or absent, supportive care is limited and a fraction of patients (varying with age) will also have AIDS. In contrast, intensive chemotherapy of the kind used in Europe is not feasible for reasons of cost, complexity of treatment, and the need for excellent supportive care and management by experienced oncologists, which is currently not available in Africa.

Diffuse Large B Cell Lymphoma and AIDS-associated NHLs

A similar situation pertains with respect to DLBCL, where if any therapy can be called standard in Africa, it is CHOP (cyclophosphamide, hydroxyrubicin, vincristine and prednisone). Many patients in Africa do not receive the full complement of drugs, possibly because hydroxyrubicin is relatively expensive. Limited information is available with respect to the treatment of DLBCL in Africa. However, recent data has suggested that in children and young adult patients up to the age of 30-35, the predominant DLBCL subtype is the germinal centre cell type (Oschlies *et al*, 2006), which has a better prognosis. Indeed in one large population-based study, over a third of children diagnosed with DLBCL had a molecular profile of BL (Klapper *et al*, 2008). No difference in outcome between histologically diagnosed DLBCL and BL when treated with BL therapy has been demonstrated in this young age group in Europe. In a recent study from Uganda, BL comprised approximately one quarter of all NHL, a high fraction of the remainder were DLBCL and the majority of patients were less than 20 years of age (Mwakigonja *et al*. 2008, Oriol *et al*, 2008). Thus, for most patients, even with DLBCL, BL therapy may well be a reasonable and feasible approach for a high fraction of patients; this could have the advantage of avoiding the use of the more expensive and potentially toxic hyroxyrubicin without compromising results.

Preliminary results of the INCTR-assisted treatment protocols in sub-Saharan Africa

For the reasons provided above, the INCTR BL strategy group opted to conduct a pilot study using a standardized version of COM as first-line therapy in a simple clinical trial for previously untreated BL. Guidelines for the safe administration of therapy and the provision of supportive care were included in the protocol, and cytotoxic drugs and antibiotics were provided free-of-charge, in addition to salaries and training for local persons to collect accurate data and follow-up of patients who failed to return for appointments after completion of therapy. Patients who developed progressive tumour at any time in their treatment (i.e., during remission induction or after achieving a complete remission) were treated with a second combination regimen, also felt likely to be feasible in the African setting, consisting of ifosfamide, mesna and cytosine arabinoside. To date, over 460 patients, nearly all children, with BL (including the few associated with HIV infection) have been treated since 2004 in centres in Kenya, Tanzania and Nigeria. High remission rates have been achieved with front line therapy, and excellent survival rates for the African context obtained in some centres, although significant variability in survival rates was observed among different centres – most probably due to failure to complete therapy, or to be treated with second-line therapy (which was shown to be active) in the presence of progressive disease in centres with a worse outcome.

Importantly, in addition to markedly improved outcome of treatment compared to patients treated prior to initiation of the study, follow up and documentation of results have all been greatly improved. Moreover, the identification of avoidable deaths and the proven efficacy of the second-line regimen suggest that this result can be improved upon in the coming years and the benefits extended to additional patients in the same or other African countries.

Acquiring reliable clinical and pathology data in sub-Saharan Africa

Laboratory studies require patient tissues and/or fluids, which must be correctly collected, stored and documented in order that correct results may be obtained and, where appropriate, correlations made with clinical findings, such that accurate evidence is collected. The INCTRs strategy for serum and tissue collection is as follows:

Serum samples

Ideally, serum samples (from 7-10 ml of blood) should be obtained on at least two occasions: one immediately prior to treatment (ideally, on the day that treatment begins, but at least within 72 h prior to the initiation of treatment) and the other immediately before cycle 2 (within 72 h prior to the initiation of the cycle. Ideally, serum should be stored in one 2 ml aliquot and the remainder in 1 ml aliquots (3 vials per patient). Each vial should be labelled with the patient study number (which should be a unique identifier) and date. The label on the tube should be firmly attached, or if more convenient, an indelible marker pen or a pen used for writing on glass should be used. Vials should then be placed in a sequentially numbered serum storage box with standard compartments of a size to hold the serum vials firmly. All boxes should be kept at −80C. Precautions should be taken against thawing, and the freezer should be connected to an emergency generator that automatically switches on in the event of a power failure.

Tissue samples

An attempt to obtain tumour/lymph node samples should be made in every case. Diagnosis rendered on FNA has limitations, and using only FNA for diagnosis can compromise at the specific diagnosis and impact on correct treatment. Errors in FNA diagnosis can be difficult to resolve even during a review undertaken later by a specialist haematopathologist. If an open or a surgical biopsy is not possible, a needle core biopsy should be attempted. An imprint of the needle core biopsy or the open biopsy can be used for immediate diagnosis, on which treatment can be based and initiated. Diagnosis rendered on imprint cytology should be followed by a more formal diagnosis on histology possibly supported by immunohistochemistry. Informed consent should be sought from the patients or their guardians regarding the performance of the procedure prior to obtaining consent for participation in the treatment protocol.

Appropriate handling of tissue samples is critical for correct diagnosis and reliable research. Once the imprints have been obtained, the remainder of the material should arrive at the pathology service within 30 min of being surgically removed. The tissue should be sectioned by a pathologist into 3 mm slices for obtaining ‘touch preparations’ or ‘imprints’. At least two touch preparations should be obtained - one imprint should be air-dried and stained with May-Grunwald-Giemsa, and the other fixed with cytofix and stained with Papanicolaou. Tissue slices should then be fixed in 10% buffered formalin at the optimal fixation time of 24 h at room temperature. If the lymph node is large enough, additional slices should be prepared for cryopreservation. Labelling of frozen samples should be performed similar to those described for serum, although larger vials and boxes will be required, and frozen samples should be stored in a different part of the freezer than serum samples.

Records on storage of frozen serum and tissue samples and on the paraffin block archives should be maintained on an electronic data base so that data is readily available and searchable, as well as analysable.

Data Management

Robust electronic database recording clinical and pathological information is critical for translational research. The INCTR has a purpose-built database that has been installed in all its participating centres. It overcomes problems of intermittent internet connection in Africa because data is entered off-line, then uploaded and synchronized with the central database in Brussels as data entry for individual patients is completed. This both increases the security of data storage (because it is now centralized on a secure server) and enables data to be examined immediately and any necessary queries generated without delay, as local site data is synchronized with the data kept on the central server immediately it has been entered at the site level. It also avoids use of e-mail for data transfer with its associated delays and hazards. The database records all the essential patient presentation data, clinical findings and results of pathological and imaging investigations. Data regarding treatment and follow-up is also appended periodically as per standard practices determined by INCTR's Clinical Trials Office.

Fostering high-quality translational research

A respectful partnership between investigators in Africa and developed countries can indeed result in high-quality translational research and the establishment of research infrastructure in Africa working partnership. Within our group, Professor Leoncini and his associates have been able to establish this. Studies from this group have led to: 1) Identification of differences between endemic BL and sporadic BL in terms of the differences in the immunoglobulin gene mutation-rates and the possible role of antigen selection (Bellan *et al*, 2005); 2) Identification of the role of *RBL2* gene in endemic BL and the target genes of RBL2/p130 (De Falco *et al*, 2007); 3) Identification of alternate pathogenetic mechanisms involving microRNA in *MYC* translocation-negative BL (Leucci *et al*, 2008); and 4) several studies on gene and microRNA expression in BL (Leucci *et al*, 2010; Onnis *et al*, 2010; Piccaluga *et al*, 2011). It should be noted that microRNA studies and studies requiring immunohistochemistry, ISH or FISH can be performed on formalin-fixed tissue samples.

Though the opportunities for high-quality research through collaboration with centres in Africa is huge, investments into improving diagnostic accuracy and infrastructure, and optimal patient management while maintaining a good clinical and pathology database, are of paramount importance.

Conclusion

The discovery of BL in Africa has led us to extraordinary achievements in oncology, medicine, epidemiology and science. Half a century after this discovery, treating African patients with BL remains a challenge. The study of African lymphomas is likely to lead us to new discoveries. However, these ventures require robust partnerships between Africa and

the rest of the world. Improving the infrastructure for accurate diagnosis and optimal patient management are key to meaningful translational research and scientific advances.

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Table 1

Lymphoma subtype among 132 paediatric and adolescent (<18 years) NHL cases

Precursor lymphoid neoplasms	*4 (3%)*
B lymphoblastic leukaemia/lymphoma	1 (0.7%)
T lymphoblastic leukaemia/lymphoma	1 (0.7%)
Blastic leukaemia/lymphoma, not otherwise specified	2 (1.5%)
Mature B-cell neoplasms	*123 (92.5%)*
Diffuse large B-cell lymphoma (DLBCL)	10 (7.5%)
Burkitt lymphoma (BL)	109 (82%)
B-cell lymphoma, unclassifiable with features intermediate between DLBCL & BL	4 (3%)
Mature T-cell & NK cell neoplasms	*3 (2.2%)*
Peripheral T-cell lymphoma, not otherwise specified	1 (0.7%)
Anaplastic large cell lymphoma, ALK positive	2 (1.5%)
NHL, not further classifiable due to technical reasons	*2*

DLBCL, diffuse large B-cell lymphoma ; BL, Burkitt lymphoma

Table 2

Histological subtypes among 19 paediatric and adolescent (<18 years) HL cases

Nodular lymphocyte predominant Hodgkin lymphoma	1 (5%)
Nodular sclerosis classical Hodgkin lymphoma	7 (37%)
Mixed cellularity classical Hodgkin lymphoma	7 (37%)
Lymphocyte-depleted classical Hodgkin lymphoma	4 (21%)

Table 3

Lymphoma subtype among 172 adult NHL cases

Precursor lymphoid neoplasms	*8 (4.7%)*
B lymphoblastic leukaemia/lymphoma	1 (0.6%)
T lymphoblastic leukaemia/lymphoma	4 (2.4%)
Blastic leukaemia/lymphoma, not otherwise specified	3 (1.8%)
Mature B-cell neoplasms	*157 (91.3%)*
Chronic lymphocytic leukaemia / small lymphocytic lymphoma	16 (9.4%)
Lymphoplasmacytic lymphoma	1 (0.6%)
Plasmacytoma	5 (3%)
Extranodal marginal zone lymphoma of MALT type	2 (1.2%)
Nodal marginal zone lymphoma	4 (2.4%)
Mantle cell lymphoma	1 (0.6%)
Follicular lymphoma	5 (3%)
Primary cutaneous follicular lymphoma	1 (0.6%)
Diffuse large B-cell lymphoma (DLBCL)	95 (55%)
Plasmablastic lymphoma	6 (3.6%)
Burkitt lymphoma (BL)	15 (9%)
B-cell lymphoma, unclassifiable with features intermediate between DLBCL & BL	5 (3%)
B-cell lymphoma, unclassifiable with features intermediate between DLBCL & classical Hodgkin lymphoma	1 (0.6%)
Mature T-cell & NK cell neoplasms	*5 (3%)*
Extranodal NK/T cell lymphoma, nasal type	1 (0.6%)
Peripheral T-cell lymphoma, not otherwise specified	3 (1.8%)
Primary cutaneous anaplastic large cell lymphoma	1 (0.6%)
NHL, not further classifiable due to technical reasons	*2*

DLBCL, diffuse large B-cell lymphoma ; BL, Burkitt lymphoma ; MALT, mucosa-associated lymphoid tissue

Table 4

Histological subtypes among 32 cases of HL in adults

Nodular lymphocyte predominant Hodgkin lymphoma	3 (9.4%)
Nodular sclerosis classical Hodgkin lymphoma	17 (53%)
Lymphocyte rich classical Hodgkin lymphoma	1 (3%)
Mixed cellularity classical Hodgkin lymphoma	10 (31.2%)
Lymphocyte depleted classical Hodgkin lymphoma	1 (3%)