An international journal published by the

Nigerian Society for Experimental Biology

Antibody and complement levels in patients with Burkitt's lymphoma

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Abstract

The concentration of immunoglobulins (IgA, IgG, and IgM) and complement (C_3 and C_4) were measured in fifty-seven (57) Nigerian children with Burkitt's lymphoma (BL), and in twenty-eight (28) apparently healthy control subjects, using the single radial immunodiffusion (SRID) technique. The sera analyzed were obtained from the four clinical stages of BL. There were high titres of IgA (stage I: n=22; 667.27 ± 509.16; stage II:n=11; 616.82 ± 408.55; stage III: n=20; 695.65 ± 699.93 and stage IV: n=4, 485.75 ± 289.90); IgG (stage I: n=22, 260.59 ± 232.56; stage II:n=11 195.18 ± 102.86;stage III:n=20, 309.65 ± 276.43; stage IV:n=4, 244.85 ± 400.107). IgM (stage I:n=22, 253.55 ± 104.99, stage:II n=11, 217.82 ± 77.8; stage III:n=20, 419.20 ± 74.21, stage IV:n=4, 233.35 ± 108.84). These values are significantly higher ($P<0.05$) than the values for the control subjects (IgA, 330.11±260.55; IgG, 288.29 ± 346.95, IgM, 201.82 ± 64.132). Higher complement titres were also observed in the various clinical stages of BL C3 (stage:In=22, 177.25 ± 5172; stage II:n=11, 197.36 ± 65.40; stage III:n=20, 190.05 ± 60.91; stage IV:n = 4, 142.25 ± 81.40); C4 (stage I:n=22,288.68 ± 181.84, stage II:n=11, 207.91 ± 67.64; stage III:n=20,243.30 ± 164.70, stage IV:n=4, 331.75 ± 275.59). The complement values for control subjects were significantly lower than the BL patients; (C3:157.04± 37.76; C4 256.39 ± 181.93). These results show relative higher immnoglobulins and complement responses in Nigerian children with BL.

Key words: Immunoglobulin, Burkitt's lymphoma, Nigeria

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INTRODUCTION

The etiology of Burkitt's lymphoma (BL) has been reported by several authors [1,2,3,4]. One of Burkitt's early papers discussed the potential role of host immunity to BL. He reported cases of spontaneous remission, and those who transiently responded to serotherapy with sera from BL patients who had undergone complete remission[5]. Other investigators who pursued this initial documented idea observed evidence of serum cytotoxicity *in-vitro*. The development of hypersensitivity reactions to extracts of autologous tumour and impaired immune reactions associated with the presence of large tumours further support the potential role of immune failure in the pathogenesis of BL.[6,7,8]. Clinical trials of immunotherapy were started in the early 1970's based on Burkitt's hypothesis of immunosurveilance using sera from patients who had undergone complete tumour regression, and immunostimulators like Bacillus Calmette Guerin (BCG)[9]. Depressed host immunity measured by mitogen stimulation and delayed type hypersentivity in BL patients bear direct and indirect relationship to the tumour burden[10,11]. The immunology of BL is further complicated by the presence of Epstein Barr viral particles in the tumour cells which express a variety of virally determined antigens that provoke complementary antibody and cellular responses[4]. In addition, as a B-lymphocyte tumour, the cells may be subjected to immunoregulatory influences such as T-helper cells or T-suppressor cells and antidiotype antibody responses.

The aim of this work is to measure the concentration of serum immunoglobulins IgA, IgG, IgM and complements (C_3, C_4), in the various clinical stages of BL, and to determine their relevance in BL pathogenesis in Nigerian children with BL.

MATERIALS AND METHODS

Patients' Selection
Fifty-seven (57) Burkitt's lymphoma patients (4-14yrs)of age attending Paediatric clinic of Ahmadu Bello University Teaching Hospital (ABUTH) Zaria and Kaduna, Nigeria, between 1994 and 1996, were enlisted for the study. These cases were diagnosed clinically and histopathologically using the established WHO criteria[12]. Human experimentation conduct protocol was approved by the ABU-Hospital Ethical Committee (Ref. No. F-MED/com.19/16/6/95 and the project number was ESC/95/00075).

Quantitation of serum Immunoglobulin and complement
The single radial immunodiffusion technique[13] was used. Serial dilution (nondil; 1:2, 1: 4, and 1:8) of standard serum (WHO recommended) were used to obtain a standard calibration curve. Samples were then assayed, and the immunoglobulin and complements were estimated using the calibration curve. Commercial test kits supplied by Sigma, Chemicals, United Kingdom were used for the assays.

Statistical Analysis
The data collected were subjected to statistical analysis using the E.P.I computer package. The various levels of significant differences were computed using analysis of variance (ANOVA), Bartlet test of homogeneity and Kruskal-Wallis one-way analysis of variance.

RESULTS

There was significant increase (P<0.05) in the concentration of IgA in all the clinical stages of BL when compared with control subjects. Table 3 shows significant different in *AP*, *P<0.005*. A relatively higher IgG titre is seen in Table 3 with significant difference in *BP* and *KP* with *P<0.005*. IgM has significantly higher titre in all the clinical stages when compared with the titres for the control subject. Table 3, also shows significant difference in *BP* and *KP (P<0.05).* The various concentrations of the serum complement (C_3 and C_4) in all the clinical stages were not significantly higher than the levels in the control subjects.

Relative comparison of the various immunoglobulin show that IgA has the highest

Table 1: Sex distribution of the BL patients and control subjects

Sex	B.L	Controls
Male	42	19
Female	15	9
Male/female	2.8:1	2.1:1

BL = Burkitt's lymphoma

Table 2: Mean age and clinical stages of the 57 BL patients and 28 control subjects

	Age distribution (yr)	
Clinical stage	BL	Control
I (n= 22)	8.33 (5.1) (5 –13)	9.21 (3.2) (4 – 14)
II (n= 11)	7.4 (2.2) (4 – 11)	
III (n= 20)	9.1 (2.4) (4 – 14)	
IV (n= 4)	8.8 (1.3) (7 – 10)	

Values are expressed in means (SD). Figures in parenthesis are the age ranges.

Table 3: Serum Immunoglobulin and complement concentrations in Burkitts lymphoma patients and control subjects

	Serum immunoglobulins (iu/mL)			Complements (iu/mL)	
Clinical stage	IgA	IgG	IgM	C_3	C_4
I (n=22)	667.27 ±509.16 (139 – 1625)	260.59 ± 232.56 (42 – 1134)	253.55 ± 104.99 (130 – 520)	177.23 ± 51.72 (83 – 239)	288.59 ± 161.18 (81 – 523)
II (n = 11)	616.82 ± 4.08.55 (95- 1625)	195.18 ± 102.86 (63 – 384)	217.82 ± 77.80 (146 – 412)	197.36 ± 65.40 (75 – 304)	20.3.91 ± 67.64 (96 – 313)
III (n=22)	695.65 ± 699.36 (139 – 2198)	309.65 ± 276.43 (76 – 1321)	419.80 ± 74.24 (112 – 406)	196.05 ± 60.09 (75 – 304)	243.3 ± 164.70 (10 – 714)
IV (n=4)	485.35 ± 289.90 (115 – 789)	244.85 ± 400.10 (50 – 9346)	233-25 ± 108.84 (105 – 348)	142.25 ± 8.46) (69 – 258)	331.75 ± 235.59 (56 – 714)
Control (n=28)	330.11 ± 260.65 (66 – 1006)	288.23 ± 346.95 (76 – 1260)	201.52 ± 64.13 (94 – 320)	157.04 ± 37.76 (92 – 236)	225.39 ± 181.93 (122 – 345)
P – value	*A – P=0.0011 B –P=0.00000 K – P=0.529	A – P = 0.63 *B – P = 0.0111 *K – P = 0.042	A – P = 0.365 *B – P = 0.0111 * K – P = 0.042	A – P = 0.365 *B – P = 0.000 K – P = 0.786	A – P = 0.093 B – P = 0.3085 K – P = 0.1397

Values are expressed as mean ± SD
Values in parenthesis indicate the range
The stars indicate degree of statistical significant differences using the various statistical models indicated.
A – values using ANOVA Test for normal distribution
B – values using Bartlets Test for homogeneity of variance
K – value using Kruskal – wallis one way analysis of variance.

titre and the class III clinical stage had the highest concentration.

DISCUSSION

The central role of antibody mediated immune response in BL has not been clearly documented. Early studies using cultured cell-lines from BL biopsy were able to recognize, some BL specific antibodies like the cardio antigens and their specific antibodies (VCA), membrane antigens (MA) with their specific antibodies, and predictions were made based on these findings[14,8]. The presence of these antibodies in very high titres have been of prognostic importance;[14,15].

This *in-vitro* finding using human sera has been able to demonstrate a significantly high immunoglobulin response in BL patients which correlate with disease activity. For example, the highest concentration of IgA was noticed among patient with tumour in multiple sites (Table 3). This observation tends to agree with earlier suggestions[14,15]. This study shows that in BL patients the major Immunoglobulin response was IgA followed by IgM and IgG.

Table 3, also shows significantly high level of the complement components, C_3 and C_4; C_3 levels from stage I to III were relatively higher than control group and C_4 levels from stage I to IV were all higher than control group but not to a statistically significant level ($P>0.05$). The actual role played by these high concentration of antibodies and complement components could be suggestive (from some clinical observations) of the tumour site. Clinically, the oral tumours are oedematous and bleed easily, indications of inflammatory activities, which may be due to complement activation. Usually the oral hygiene were poor with dental anarchy and offensive odour. These are suggestive of the role played by high complement (C_3 and C_4) titres in BL.

High level of 1gA, IgM and IgG have also been reported in some tropical diseases like malaria, and in HIV infected patients [16,17]. In these cases, it was observed that there was hypergammaglobulinaemia in Nigerian patients with HIV [17] and IgG subclass levels in malaria

infected Nigerian[16]. Previously, the etiological correlation between malaria infestation and BL has been established, and hypergammaglobulinaemic condition may induce suppressions including immune suppression that might result in BL[3].

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