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Original Article

Prediction and Diagnosis of Renal Allograft Dysfunction By Monitoring of Several Cytokines and T4/T8 Ratio in The First Six Months After Transplantation (Tx)

Mohamed G. Saadi, Fatina Fadel*, Tayseer Eyada and Hamdy Mostafa**
Departments of Internal Medicine and Nephrology, Pediatric Nephrology and Clinical Pathology**, Cairo University, Egypt*

ABSTRACT

Background: Acute rejection with its chronic sequele may affect the overall outcome of renal Tx. It is a complex series of events involving the cytokine response, signaling the initiation and modulation of this process.

Objectives: Our aim was the early detection and management of rejection and preservation of graft function.

Methods: 7 children and 18 adult transplanted cases were followed 1, 3 and 6 months after Tx by monitoring several cytokines including Tumor Necrosis Factor alpha (TNF-α), IL-1, IL-3, IL-4, IL-10, Transforming Growth Factor Beta (TGF-β), Insulin Like Growth Factor 1 (IGF-1) and Platelet Derived Growth Factor (PDGF-AB) as well as estimation of the T-helper (T4) to the T-suppressor (T8) ratio. Fifteen of the 25 cases had an impairment of their graft function at one time of the follow up period. The results were compared with the values of 10 healthy controls.

Results: The 10 stable cases included 5 children whose cytokine levels were not significantly different from those of the 5 adults.

The mean values of the 15 cases with dysfunction were significantly higher than those of the 10 stable cases and 10 controls. There were significant positive correlations between the s. creatinine and all the studied parameters except IGF-1 whose rate of decline rather than its absolute levels which correlated with the s. creatinine.

Five/15 cases with dysfunction were already presenting with renal impairment by the time of first sampling. In the remaining 10/15 prediction prior to actual dysfunction was achieved in all of them, denoting 100% predictability. Of these TNF-α predicted 8/10, IL-4 6/10, IL-10 6/10, IL-3 3/10, and IL-1 2/10.

Conclusions: More frequent monitoring of various cytokines may increase the predictability values, and it may be limited to monitoring of TNF-α, IL-4 and/or IL-10. The value of monitoring TGF-β, IGF-1 and PDGF-AB may be more valuable later in transplantation.

INTRODUCTION

Acute cellular rejection of renal allografts remains a principal cause of graft loss in human kidney transplants. Approximately 10 - 15% of renal grafts are lost due to rejection in the first year post-transplantation. The complexity and unpredictability of renal graft rejection suggests the presence of multiple signals which initiate, modulate and affect this process. These signals do exist and are termed cytokines[1]

Meanwhile chronic graft loss may be a consequence of various factors operating separately or concomitantly. There may be mild acute rejection attacks or a slowly progressive unnoticed underlying chronic rejection process. In addition toxic effects of immunosuppressive agents, particularly cyclosporine-A (CsA) which is known to have vasculopathic and interstitial fibrotic effects, may be operating. Furthermore recurrence of the original disease, remains a

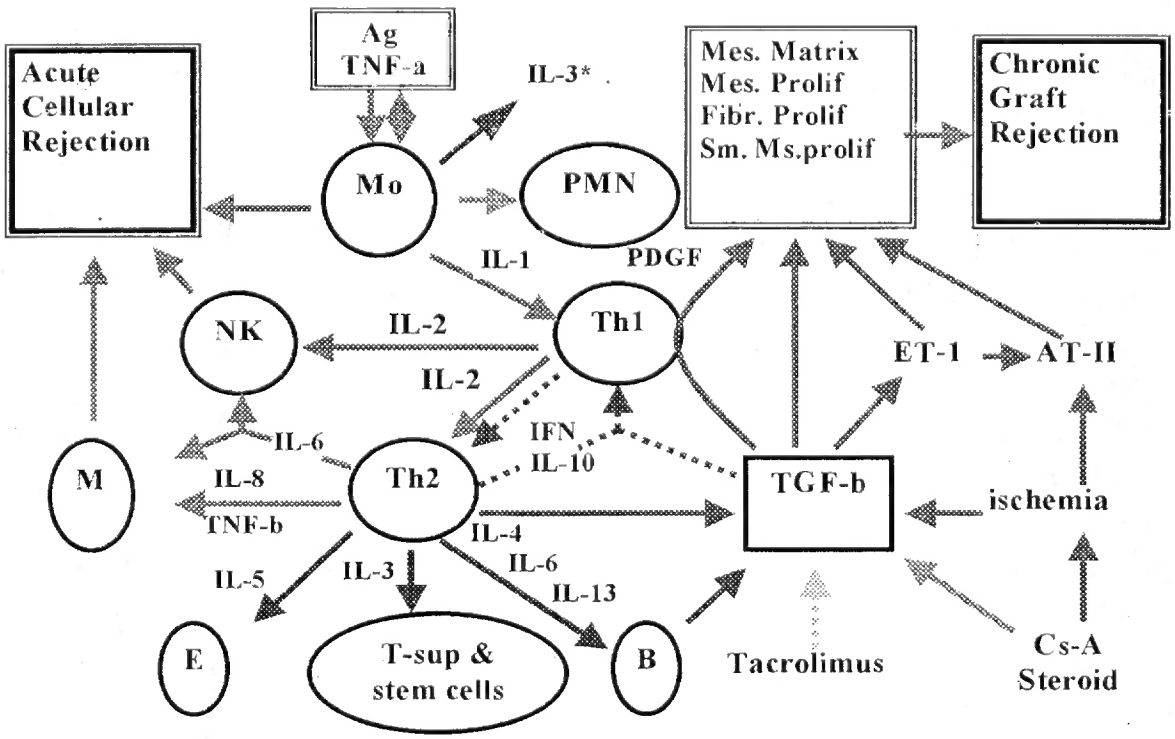
potential challenge for graft dysfunction[2].

The reactions of the immune system of the recipient demonstrated in figure (1) is under continuous immunologic stimulation by the graft antigens, mainly class II major histocompatibility antigens. These stimulate the macrophages (Mθ), which act as antigen presenting cells to the immune system. Mθ produce TNF-α which acts in an autocrine or paracrine matter to reinforce the Mθ which release several cytokines among which IL-1 is responsible for stimulation of TH1 lymphocytes which release IL-2 . The latter would stimulate the TH2 lymphocytes to release IL-6, IL-8, and TNF-β. Both IL-6 and IL-2 stimulate the activation of natural killer NK cells from the T-lymphocytes. IL-6, IL-8 and TNF-β stimulate the function of monocytes. The activated monocytes, NK cells and Mθ would attack the foreign graft antigens, mainly class I major histocompatibility antigens, leading to acute cellular rejection. The TH2 cells also release IL-4 which together with IL-6 stimulate the antibody production by the B lymphocytes. This contributes to a lesser extent to the acute rejection process[1].

The stimulated TH2 cells release IL-10 and together with the B lymphocytes release TGF-β. IL-4, IL-10 and TGF-β would act in a negative feedback mechanism to suppress the TH1 cells. The production of TGF-β may be stimulated by Cs-A and steroids which suppress TH1 activity and IL-2 receptor presentation on these cells[3].

There is debate concerning the action of both IL-4 and IL-10, where both are believed by some to have significant rejection suppressor activity[1], yet others believe that they are responsible for the rejection process and IL-4 was even blamed for enhancing the T cell functions[4].

Figure (1): PATHOGENESIS OF GRAFT REJECTION



There are as well conflicting results concerning the expression of TGF-β in histochemical studies during rejection, where it was reported to be increased[3,5], decreased[6] or unchanged[7].

The stimulated Mθ and TH2 cells produce IL-3 which stimulates the differentiation of T8 suppressor lymphocytes and would favor the reduction of the helper T4/T8 ratio[3].

Chronic graft atherosclerosis is believed to be mediated by cytokines including growth factors such as platelet derived growth factor (PDGF), fibroblast growth factor and insulin like growth factor (IGF).

TGF-β produced by TH2 cells due to either antigen stimulation or immunosuppressive agents, stimulates the production of the growth factors by the T-lymphocytes, as well as the production of endothelin (ET) by the endothelial cells.

TGF-β together with the growth factors including PDGF and IGF, as well as ET-1 potentiate excessive mesangial matrix deposition, mesangial and fibroblastic proliferation together with vascular smooth muscle proliferation, leading to transplantation associated glomerulosclerosis (Tx-AGS) and transplantation associated arteriosclerosis (Tx-AA) collectively known as graft atherosclerosis[8].

There is controversy concerning the immune response towards the grafts in young recipients, where some investigators believe it to be heightened in young patients below 5 years of age[9]. Yet others encountered excellent results in this age group[10,11].

Children may as well have different drug pharmacokinetic responses than in adults, due to more active enzyme system and would require more frequent drug dosing[12].

The significance of non-invasive techniques to the early diagnosis of acute rejection remains elusive. The central role of the cytokines in the amplification of the immune response and the relation of their plasma levels to renal allograft rejection had been high-lighted by the previously reported studies.

AIM OF WORK

The aim of the present work is to monitor some of the interleukins, growth factors, and lymphokines involved in the rejection process. Studying TNF-α, IL-1, IL-3, IL-4, IL-10, TGF-β, PDGF-AB, IGF-1, and T4/T8 ratio in 25 kidney transplant cases followed at one, three, and six months after transplantation, in order to determine the predictive, diagnostic and prognostic values of these cytokines concerning graft function and outcome. Also we tried to detect any difference in their pattern among the young cases included.

PATIENTS AND METHODS

Samples were collected at one, three and six months after renal transplantation from 55 cases. All the patients received living donated kidneys. The samples chosen for analysis were those of 15 cases (including 2 children) who showed graft dysfunction at one time of the follow-up period, and of 10 out of the remaining 40 cases with stable kidney function throughout the same period (including 5 children), as well as the samples of 10 healthy controls (including 3 children).

The 25 studied patients were 21 males

and 4 females, their mean age was 32.9 ± 18.4 years. The mean age of the adults included (18 cases) was 43.3 ± 10.7 years with a minimum age of 30 years and a maximum age of 63 years. Their original disease was hypertensive nephrosclerosis in 8 cases, obstructive uropathy in 3 cases, polycystic kidney in 2 cases, vesico-ureteric reflux in 1 case, glomerulonephritis in 1 case, while the cause was unknown in 3 cases. The mean age of the children included (7 cases) was 10.4 ± 5.1 with a minimum age of 3 years and a maximum age of 18 years. Their original disease was glomerulonephritis in 3 cases, vesico-ureteric reflux in 2 cases, while the cause was unknown in the remaining 2 cases.

The patients were given a triple immunosuppressive regimen comprising:
Prednisolone 3.5 mg/kg body weight day -1, 3.5 mg/kg x 2 intraoperative, 3.5 mg/kg day 0, 3.0 mg/kg day 1 tapered over 2 weeks to 0.5 mg/kg/day maintained over the first 3 months and reduced to 0.25 mg/kg/day over the next 3 months then 0.1 - 0.2 mg/kg/day.

The maintenance dose for children after the third post operative month were given on an alternate day regimen.
Azathioprin 1.5 mg/kg/day on days -2 and -1, 5 mg/kg on day 0, reduced over one week to 2 mg/kg/day for the first 3 months, and then maintained at 1.5 mg/kg/day, dose adjusted by the total leukocyte count.

CsA was started on day 0 or + 1 after ensuring diuresis at a dose of 6 mg/kg/day divided on 2 doses for adults and 3 doses for children, and adjusted to keep a trough blood level > 200 ng/ml in the first month, 150 – 200 ng/ml next month and then 100 – 150 ng/ml.

All the studied cases were subjected to routine clinical and laboratory assessment.

For cytokine assay serum was separated from the blood after clotting for 30 minutes at 4°C followed by centrifugation at 3000 xg for 15 minutes, and sera were stored at -70°C for 6 months after collection.

The assay of each of the cytokines was performed by the Medgnix ELISA kits employing a quantitative sandwich horse-radish peroxidase (HRP) enzyme linked immunosorbant assay, utilising cytokine specific monoclonal antibodies. The bound enzyme labeled antibodies are measured by a chromogenic reaction reading the absorbance at 450 nm.

To study the T helper (T4) to the T suppressor (T8) ratio the lymphocytes were separated from the peripheral blood by the Boyum technique. This was followed by phenotyping for the cells using specific monoclonal antibodies (OKT) by the indirect immuno fluorescence technique.

Statistical methods:

Data were summarized as the arithmetic mean (M), the average describing the central tendency of observations.

The standard deviation (SD) measured the dispersion of results around the mean.

The students t-test was used for comparing the means of 2 independent groups.

The correlation between variables was done using the regression analysis (t test)[14].

RESULTS

The important clinical and laboratory data are shown in the following tables:

Table 1: Statistical comparison of the tissue typing (matches and mismatches) and mixed lymphocytic culture (MLC) of the studied patients

		All patients N = 25	Adults N = 18	Children N = 7
HLA Matches	M	2.8	2.89	2.71
	S.D.	± 1.5	± 0.9	± 0.95
	p		N.S.	
HLA Mismatches	M	1.48	1.5	1.43
	S.D.	± 1.08	± 1.25	± 0.53
	p		N.S.	
MLC	M	4	3.36	4.24
	S.D.	± 2	± 0.87	± 1.18
	p		< 0.025	

Table 2: Follow up of the 10 kidney transplant cases who showed normal kidney functions through out the six months follow up after transplantation

	First month			Third month			Sixth month		
	All patients	Adults N = 5	Children N = 5	All patients	Adults N = 5	Children N = 5	All Patients	Adults N = 5	Children N = 5
S. cr. M	1.1	1.2	1.1	1.2	1.2	1.2	1.06	1.1	1.04
S.D.	± 0.2	± 0.2	± 0.2	± 0.2	± 0.2	± 0.2	± 0.18	± 0.19	± 0.18
p		N.S.			N.S.			N.S.	
Cs A M	235.8	249.2	228.4	182.5	199	166	135.3	148	121
S.D.	± 23.2	± 25.8	± 16.6	± 30.5	± 15.2	± 34.4	± 24.4	± 13.0	± 26.0
p		N.S.			N.S.			N.S.	
TNFα M	11.5	7.7	15.2	19.2	20.0	18.4	15.4	14.8	16.0
S.D.	± 10.0	± 8.6	± 10.8	± 7.4	± 10.1	± 4.3	± 9.1	± 8.1	± 10.9
p		N.S.			N.S.			N.S.	
IL-1 M	9.1	12.0	6.2	12.7	6.8	18.6	11.8	9.0	14.6
S.D.	± 6.7	± 7.6	± 4.8	± 12.7	± 1.6	± 16.6	± 10.6	± 1.7	± 15.2
p		N.S.			N.S.			N.S.	
IL-3 M	11.1	13.4	8.8	7.5	6.0	9.0	16.5	15.0	18.0
S.D.	± 12.3	± 16.8	± 6.5	± 1.7	± 4.4	± 5.0	± 10.0	± 6.1	± 13.5
p		N.S.			N.S.			N.S.	
IL-4 M	-1	-1	-1	-1	-1	-1	-1	-1	-1
S.D.	± 0	± 0	± 0	± 0	± 0	± 0	± 0	± 0	± 0
p		N.S.			N.S.			N.S.	

Continued

Table 2: Continued

	First month			Third month			Sixth month		
	All patients	Adults N = 5	Children N = 5	All patients	Adults N = 5	Children N = 5	All Patients	Adults N = 5	Children N = 5
IL-10									
M	-1	-1	-1	-1	-1	-1	-1	-1	-1
S.D.	± 0	± 0	± 0	± 0	± 0	± 0	± 0	± 0	± 0
p		N.S.			N.S.			N.S.	
TGF-β									
M	102.7	100.2	105.2	96.8	93.0	100.6	89.3	88.0	90.6
S.D.	± 12.8	± 13.8	± 12.8	± 20.2	± 21.5	± 20.5	± 20.6	± 19.6	± 23.8
p		N.S.			N.S.			N.S.	
PDGF-AB									
M	15350	12500	18200	22600	21200	24000	23900	24300	23500
S.D.	± 7764	± 3937	± 9991.2	± 7820.2	± 9731	± 6154.4	± 4915	± 5696.5	± 4636.8
p		N.S.			N.S.			N.S.	
IGF-1									
M	314.5	310.0	319.0	228.0	244.0	212.0	248.1	238.2	258
S.D.	± 88.5	± 28.3	± 129.5	± 70.7	± 80.2	± 64.6	± 94.8	± 115.1	± 82.0
p		N.S.			N.S.			N.S.	
T4									
M	43.4	35.2	51.6	39.5	31.0	48.0	42.0	34.0	50.2
S.D.	± 16.3	± 3.9	± 20.3	± 17.5	± 6.8	± 21.4	± 16.1	± 4.0	± 21.1
p		N.S.			N.S.			N.S.	
T8									
M	26.7	26.3	27.5	31.5	29.7	33.2	30.0	24.9	34.8
S.D.	± 5.0	± 5.2	± 5.5	± 6.2	± 8.0	± 3.8	± 11.0	± 8.8	± 11.9
p		N.S.			N.S.			N.S.	
T4/T8									
M	1.6	1.4	1.8	1.3	1.1	1.4	1.5	1.4	1.6
S.D.	± 0.36	± 0.23	± 0.33	± 0.48	± 0.4	± 0.5	± 0.53	± 0.38	± 0.69
p		< 0.05			N.S.			N.S.	

N.B.: -1 means undetectable level

Table 3: Statistical comparison of the data of the studied groups one month after transplantation

	Unstable cases (N = 15)	Stable cases (N = 10)	Controls (N = 10)
S Cr	1.5 ± 0.48	1.1 ± 0.2	1.1 ± 0.2
p # groups	< 0.05		
p # controls	0.05	N.S.	
Cs-A	167.1 ± 52.7	235.8 ± 32.2	
p # groups	N.S.		
TNF α	130.6 ± 125.6	64 ± 112	18 ± 5
p # groups	< 0.05		
p # controls	< 0.05	< 0.05	
IL-1	24.1 ± 22.7	9 ± 6.7	10 ± 3
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-3	25.06 ± 24.8	11.1 ± 12.3	11 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-4	39.6 ± 35.7	-1 ± 0000	9 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-10	13.2 ± 25.4	-1 ± 0000	7 ± 4
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
TGF-β	135.1 ± 68.3	102.7 ± 12.8	75 ± 20
p # groups	N.S.		
p # controls	N.S.	N.S.	
PDGF-AB	28,200 ± 33,300	14400 ± 8540	29,718 ± 5000
p # groups	< 0.05		
p # controls	N.S.	< 0.05	
IGF-1	293.6 ± 141	314.5 ± 88.5	270 ± 90
p # groups	N.S.		
p # controls	N.S.	N.S.	
T4/T8	1.7 ± 0.63	1.5 ± 0.27	1.5 ± 0.41
p # groups	< 0.05		
p # controls	< 0.05	N.S.	

Table 4: Statistical comparison of the data of the studied groups three month after transplantation

	Unstable cases (N = 15)	Stable cases (N = 10)	Controls (N = 10)
S Cr	1.6 ± 0.35	1.2 ± 0.2	1.1 ± 0.2
p # groups	< 0.05		
p # controls	0.05	N.S.	
Cs-A	144.3 ± 31.7	182.5 ± 30.5	
p # groups	N.S.		
TNF α	294 ± 334.9	19 ± 11	18 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-1	43.7 ± 33.2	12.7 ± 12.6	10 ± 3
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-3	63.9 ± 44.5	8.5 ± 4	11 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-4	84.9 ± 69.2	-1 ± 0000	9 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-10	22.3 ± 17.2	-1 ± 0000	7 ± 4
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
TGF-β	166.5 ± 92.3	90.8 ± 79.6	75 ± 20
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
PDGF-AB	42,300 ± 27,200	19000 ± 5000	29,718 ± 5000
p # groups	< 0.05		
p # controls	N.S.	N.S.	
IGF-1	240.8 ± 181	228 ± 70.7	270 ± 90
p # groups	N.S.		
p # controls	N.S.	N.S.	
T4/T8	2.08 ± 0.64	1.3 ± 0.37	1.5 ± 0.41
p # groups	< 0.05		
p # controls	< 0.05	N.S.	

Table 5: Statistical comparison of the data of the studied groups six month after transplantation

	Unstable cases (N = 15)	Stable cases (N = 10)	Controls (N = 10)
S Cr	1.69 ± 0.52	1.06 ± 0.18	1.1 ± 0.2
p # groups	< 0.05		
p # controls	0.05	N.S.	
Cs-A	126.7 ± 40.3	135.3 ± 24.4	
p # groups	N.S.		
TNF α	447.8 ± 511.2	15.4 ± 9.08	18 ± 5
p # groups	< 0.05		
p # controls	< 0.05	< 0.05	
IL-1	69.5 ± 66.6	11.8 ± 10.6	10 ± 3
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-3	75 ± 60.2	16.5 ± 10.01	11 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-4	95.7 ± 106	-1 ± 0000	9 ± 5
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IL-10	28.9 ± 23.1	-1 ± 0000	7 ± 4
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
TGF-β	105.7 ± 84.9	89.3 ± 20.6	75 ± 20
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
PDGF-AB	58,500 ± 87,200	24000 ± 5000	29,718 ± 5000
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
IGF-1	168.7 ± 119	238 ± 90.3	270 ± 90
p # groups	< 0.05		
p # controls	< 0.05	N.S.	
T4/T8	1.85 ± 0.6	1.5 ± 0.4	1.5 ± 0.41
p # groups	< 0.05		
p # controls	< 0.05	N.S.	

Table 6: Statistical comparison of the rate of change in IGF-I from 1-3 and 3-6 months in the studied groups

Group	Δ IGF-1 1-3 months	Δ IGF-1 3-6 months
Unstable	156.2 ± 104.4	155.4 ± 137.9
Stable	119.5 ± 87.3	44.5 ± 32.7
p value	< 0.05	< 0.05

Table 7: Correlation between serum creatinine and important studied parameters

Parameter	r value
TNF-α	0.75
IL-1	0.42
IL-3	0.5
IL-4	0.74
IL-10	0.75
TGF-β	0.43
PDGF-AB	0.64
IGF-1	0.0
ΔIGF-1	- 0.6
T4/T8	0.48

N.B.: r-value of ≥ 0.4 indicates significant correlation

Table 8: Prediction of rejection

Cytokine	Predicted cases N = 10	Alone	With other cytokines
TNF	8 cases 80%	One case	With IL-4: 2 cases With IL-10: none With IL-3 one case With IL-4 and IL-10: 3 cases With IL-4, IL-10 and IL-1: one case With IL-4, IL-10 and IL-3: one case With IL-1 and IL-3: one case
IL-4	6 cases 60%	None	With IL-10: one case With TNF: 2 cases With IL-10 and TNF: 3 cases With TNF, IL-10 and IL-3: one case With TNF IL-10 and IL-1: one case
IL-10	6 cases 60%	One case	With IL-4: one case With TNF: none With TNF, IL-4: 3 cases With TNF, IL-4, IL-3: one case With TNF, IL-4, IL-1: one case
IL-3	3 cases 30%	None	With TNF: one case With TNF and IL-1: one case With IL-4 and IL-10: one case
IL-1	2 cases 20%	None	With TNF and IL-3: one case With TNF, IL-4, IL-10: one case

DISCUSSION

As our aim was the prediction or early detection of graft rejection, we studied the cytokine responses in the serum rather than the urine. As measuring them in the urine does not detect the immunologic reaction initiating the rejection process, as the foreign graft antigens are presented systemically to the immune system which is stimulated by these antigens, and reacts to them via the various activation reactions which results in the immunologic rejection process in the graft. So to detect the undergoing systemic immune stimulation and mechanisms paving the way for a coming rejection process we need to test for these systemic reactions rather than the end organ effect. The latter would reflect the actual

rejection process not its preliminary underlying stimulatory reactions.

In a previous study for monitoring serum IL-2 receptors in the post transplantation period we demonstrated its elevation only few days before an actual rejection and can not be used for prediction on a long term follow up basis[13].

There are contradictory reports concerning the immune response in children to allografts. The results of our studied parameters of the stable cases showed that apart from the significantly higher T4/T8 ratio in children (5 cases) than adults (5 cases) there was no statistically significant difference among the different age groups. Following-up the two children among the unstable group, we noticed a 3-10 folds hampered TNF-α rise prior to manifest rejection when compared to adult cases with the same pattern of predictive rise of TNF-α. Yet due to the small number of cases we cannot conclude a reduced immunologic reaction in response to rejection in this age group as suggested by Ettenger et al.[9].

Except for IGF-1 which was lower in the unstable group than the stable group, all the tested parameters were significantly higher in the unstable group indicating a state of immune stimulation underlying the functional deterioration of the graft possibly by a rejection process. Their values were positively correlating with the serum creatinine of all the studied cases. In the unstable group the macrophage functions as denoted by the levels of TNF-α, IL-1, and IL-3, the TH1 functions as represented by its stimulatory effect on the TH2 cells as well as the level of PDGF-AB, and the TH2 functions as represented by the levels of IL-3, IL-4, IL-10, and TGF-β, were all suggestive of this state of immune stimulation whether directly mediating the rejection process or produced secondary to this state of stimulation.

And while most of these cytokines represent mediators in the afferent limb of stimulation eventually leading to the rejection process, yet IL-4, IL-10 and possibly TGF-β seem to represent the down regulating negative feed back efferent loop of this stimulated rejection process and are elevated as a reflection of it. The apparent in vitro effects of these interleukins explained by several authors[1,15] in suppressing TH1 cells as well as monocyte, and macrophage functions would be a misinterpretation of their actual stimulus for release in vivo. The T helper to the T suppressor ratio was a reflection of the balance of the immune profile and as well positively correlating with the serum creatinine.

In the unstable group 5 of the 15 cases were presenting in the time of first sampling with an impaired kidney function, and demonstrated all the previously mentioned cytokine abnormalities. The remaining 10 of this group showed one or more abnormality in their cytokine pattern prior to the elevation in their serum creatinine values by the time of the next sampling, representing 100% success in the prediction of this deterioration and by a considerable period of time.

TNF-α predicted 80% of the cases, possibly being stimulated early in the proximal part of the stimulation loop, and similar results were obtained by Keil et al., in 1994[2] and Lambert et al, in 1994[16]. Both more early and frequent analysis

would increase its predictability to 100%.

Meanwhile elevated IL-4 and IL-10 each predicted 60% of these cases. Krams et al., in 1992[17], Kutukculer et al., in 1995[18] and Mclean et al., in 1997[19] reported elevated serum IL-4 with actual rejection and undetectable values in stable cases. Also Merville et al., in 1995[20] reported this pattern for IL-10, but none encountered elevated predicting readings.

The predictive values of IL-3 (30%) and IL-1 (20%) were less contributing than those previously mentioned.

PDGF AB correlated with the serum creatinine levels, but with no pre-rejection predictive rise. Similar findings were reported by other investigators[22,23]. This suggests that it rises concomitant with the pathologic vascular changes accompanying rejection, and may play a pathogenic role in these changes, as described by Alpers et al. in 1996[24].

The absolute levels of IGF-1 were not correlating with the serum creatinine changes, and had no predictive importance. Yet its rate of change showed a significant decline with impairment of the kidney function. This suggests that there is decreased production of IGF-1 during rejection, and that its level might be influenced by various factors involving the nutritional status, and hormonal balance[25].

Our results showed a slightly higher incidence of rejection among the children included 2/7 (28.6%), as compared to 10/48 adults (20.8%). This may be related to the significantly higher MLC in children as compared to the adults, yet it may as well suggest a heightened immune response as suggested by other investigators[9], or would indicate the need for different immunosuppressive protocols.

In conclusion we believe that monitoring patients in the post transplantation period is of crucial importance to predict or detect an undergoing rejection process as well as to test the efficiency of the immunosuppressive regimen. This bears an important diagnostic and prognostic tool, for the long term out come of these patients.

We also recommend more frequent analysis of the suspected cytokines to increase their predictive value, monitoring may be restricted to fewer cytokines possibly TNF-α of the afferent limb and IL-4 of the efferent limb of the immune reaction. The benefit of monitoring growth factors may be of greater value later in transplantation to detect graft atherosclerosis.

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