

## **T-SUPPRESSOR CELL ABNORMALITIES IN TYPE I MEMBRANOPROLIFERATIVE GLOMERULONEPHRITIS**

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### **Summary**

Twelve patients with type I membranoproliferative glomerulonephritis (MPGN) have been studied. All had normal renal function, none a nephrotic syndrome, and none was on therapy. In 7/12 serum C<sub>3</sub> and/or C<sub>4</sub> were low. T cell subsets were analysed with OKT monoclonal antibodies and Ts function (TsF) was studied by the Concanavalin-A Enhancement test (Con-A E) and with a new assay where native OKT8+ cell function only is explored (OKT8-DEP PWM test). OKT4/OKT8 ratio was lower than controls ( $p < 0.025$ ). TsF, as studied by the Con-A E was unchanged, while OKT8-DEP PWM test showed a marked decrease ( $p < 0.005$ ). Hypocomplementaemic patients (H), as compared with normocomplementaemic patients (N), had lower OKT4/OKT8 ratio ( $p < 0.01$ ) while TsF was unchanged in both groups. These results are consistent with a functional defect of Ts activity in type I MPGN. Increased OKT8+ cells may be a compensatory response, and hypocomplementaemia may be a marker of this defect.

### **Introduction**

Immunologic disturbances have long been recognised in type I MPGN, as demonstrated by complement activation, immunoglobulins and complement deposition in glomerular capillary walls and detectable circulating immune complexes in some cases [1–3]. No clearcut relationship has been found between serum complement profile, circulating immune complexes, patterns of immunofluorescence on renal biopsy and disease activity [2]. The advent of monoclonal antibodies directed against T-cell subsets [4] and the availability of various T-cell functional assays [5, 6], prompted us to study the immunoregulatory status of type I MPGN patients.

## Patients and methods

Twelve patients with a histological diagnosis of type I MPGN have been investigated: six males and six females, mean age  $37 \pm 16.5$  years, with normal renal function, without nephrotic syndrome and not having received any immunosuppressive treatment in the preceding year. Duration of illness was 4–132 months. Proteinuria (ranging 0–2g/24hrs) was present in 10/12, microscopic haematuria in 7/12. Complement activity ( $CH_{50}$ ) was low in 7/12. Complement fractions  $C_3$  and  $C_4$  as measured by nephelometer analysis were respectively reduced in 5/12 and in 4/12; two patients had low values of both. Circulating immune complexes were present in 2/12 with solid phase conglutinin assay. Serum immunoglobulins showed widely scattered values. No significant relationship was found between clinical and immunological data.

T-cell subsets were analysed by indirect immunofluorescence with OKT3, OKT4, OKT8 monoclonal antibodies (Orthomune, Ortho Diagnostic Systems, Raritan, NJ) as previously described [4].

OKT4/OKT8 ratio was calculated, as well as OKT4/OKT3 and OKT8/OKT3 ratios, after subtraction of OKT4+ OKT8+ double labelled cells. OKT4/OKT3 and OKT8/OKT3 ratios were calculated to ascertain the role of OKT4 and OKT8 subsets in determining OKT4/OKT8 ratio modifications. Non-specific T-suppressor cell function (TsF) was determined by a slight modification of the Con-A E test [5] and suppressor activity was expressed as percentage inhibition. OKT8+ cell specific suppressor activity was determined with a new test (OKT8-DEP PWM), which explores the B-cell response in a Pokeweed mitogen-driven system with and without native OKT8+ cells, as described in detail elsewhere [6]. OKT8+ cell activity is expressed as a percentage of suppression.

Sixteen sex and age matched controls were studied at the same time.

## Results

In Table I, MPGN patients are compared with controls. MPGN patients showed a significantly lower OKT4/OKT8 ratio ( $p < 0.025$ ) due to an increase of OKT8/OKT3 ratio ( $p < 0.05$ ). TsF, as measured with Con-A E test was in the normal

TABLE I. OKT4/OKT8 ratio, OKT4/OKT3, OKT8/OKT3 % and suppression index % with Con-A E and OKT8-DEP PWM assays in 12 type I MPGN patients, as compared with 16 matched controls

	T4/T8 ratio	T4/T3 %	T8/T3 %	Suppression index %	
				Con-A E	OKT8-DEP PWM
Controls	$1.8 \pm 0.72$	$60.6 \pm 11.21$	$31.1 \pm 10.86$	$47.5 \pm 15$	$40.6 \pm 13.11$
Type I MPGN	$1.2 \pm 0.50$	$53.8 \pm 17.41$	$44.2 \pm 19.36$	$35.6 \pm 22.5$	$18.3 \pm 11.41$
	$p < 0.025$	$p: NS$	$p < 0.05$	$p: NS$	$p < 0.005$

range, while OKT8-DEP PWM test showed a significant reduction of native OKT8+ cell suppressor function.

In Table II, hypocomplementaemic (H) and normocomplementaemic (N) patients are compared. H patients have lower OKT4/OKT8 ratios ( $p < 0.01$ ) not only due to an increased OKT8/OKT3 ratio ( $p$ : NS), but also to a decrease of OKT4/OKT3 ratio ( $p < 0.005$ ). Similar suppressor activities were noted in both groups.

TABLE II. OKT4/OKT8 ratio, OKT4/OKT3, OKT8/OKT3 % and suppression index % with Con-A E and OKT8-DEP PWM assays in five normocomplementaemic (N) and seven hypocomplementaemic (H) type I MPGN patients

	T4/T8 ratio	T4/T3 %	T8/T3 %	Suppression index %	
				Con-A E	OKT8-DEP PWM
MPGN – N (5)	1.6 ± 0.44	68.4 ± 15.89	38.6 ± 14.9	31.3 ± 20.3	18.4 ± 14.12
MPGN – H (7)	1.0 ± 0.34	43.5 ± 9.22	48.2 ± 22.22	35.9 ± 25.7	16.5 ± 10.09
	$p < 0.01$	$p < 0.005$	$p$ : NS	$p$ : NS	$p$ : NS

## Discussion

Patients with type I MPGN, normal renal function and without a nephrotic syndrome seem to have T-cell immunoregulatory subset abnormalities. The major features are a reduction in the OKT4/OKT8 ratio, which is more pronounced in hypocomplementaemic patients, and a functional defect of Ts cells, as demonstrated by a native OKT8+ cell-dependent assay.

Our results do not completely agree with a previous report, in which, however, the cases reported were unselected and functional assays were not performed [7]. In our series the duration of illness seems to be unrelated to T cell abnormalities. The decrease of the OKT4/OKT8 ratio is due to a relative increase of OKT8+ subset, although hypocomplementaemic patients also show a reduction in OKT4+ cells. Hypocomplementaemia may thus be considered as a marker of a more pronounced immunologic disturbance, while low OKT4/OKT8 ratios and low OKT4+ cells can account for a poor overall immune responsiveness [8].

It is noteworthy that increased circulating OKT8+ cells have been demonstrated to exert a poor suppressor activity *in vitro*.

There is increasing evidence of possible discrepancies between a given T cell phenotype and its functional activity in various diseases [7–9]. The conventional Con-A E test failed to detect significant abnormalities in our patients, maybe because it explores a bulk of heterogeneous suppressor activities. On the contrary, the OKT8-DEP PWM assay seems to be more specific for the assessment of Ts cell function, without mitogen manipulations and irrespective of patients' immune status [6].

It can be suggested that the increased number of OKT8+ cells in type I MPGN patients may partially compensate for their functional defect.

## References

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