Bone Marrow Transplantation, (1999) 23, 151–155

http://www.stockton-press.co.uk/bmt

Treatment of acute graft-versus-host disease with PUVA (psoralen and ultraviolet irradiation): results of a pilot study

A Wiesmann[1], A Weller[2], G Lischka[2], T Klingebiel[3], L Kanz[1] and H Einsele[1]

Departments of [1]Hematology and Oncology, [2]Dermatology and [3]Pediatrics, University of Tübingen, Germany

Summary:

Acute graft-versus-host disease (aGVHD) is a frequent and major complication after allogeneic stem cell transplantation. For many years psoralen and ultraviolet (UV)-A light have been used in the treatment of chronic cutaneous graft-versus-host disease, but few patients have received PUVA therapy for aGVHD. We assessed 20 patients who received PUVA therapy for acute cutaneous GVHD (grade 2–4). Seven patients showed additional organ manifestations (liver, gut). To better quantify the cutaneous lesions, a new scoring system was introduced: intensity of erythema (0–3) × %body surface + size of bullae (4–5) × %body surface affected. All patients received prednisolone and PUVA for treatment of aGVHD. Fifteen patients (75%), 12 with manifestations restricted to the skin, responded by score classification (average time to a 50% score reduction: 39 days) and reduction of the dosage of prednisolone (average time to a 50% prednisolone reduction: 35 days). PUVA treatment was well tolerated and might play a role in the therapy of acute cutaneous GVHD.
Keywords: allogeneic stem cell transplantation; acute GVHD; PUVA

Allogeneic bone marrow transplantation is the only curative treatment for a variety of hematological malignancies. One frequent and major complication of this treatment modality is the occurrence of acute graft-versus-host disease (aGVHD).

Management of acute and chronic GVHD often requires immunosuppressive therapy over a prolonged time period, eg cyclosporin A and prednisolone. Due to immunosuppression, patients have an additional risk of infectious complications associated with increased morbidity and mortality.

One main localization of acute GVHD is the skin, characterized in its mild form by an erythematous morbilliform eruption, and in its extensive form as epidermal necrolysis, developing up to 100 days after allogeneic stem cell transplantation. For many years, 8-methoxypsoralen and ultraviolet-A light have been used in the treatment of a variety of skin diseases, such as psoriasis, cutaneous T cell lymphoma and vitiligo. There are several studies using PUVA therapy in the treatment of chronic cutaneous GVHD,[1–9] but few patients have received PUVA therapy for treatment of acute cutaneous GVHD.[6–11]

At our institution 20 patients were studied who received PUVA therapy for acute cutaneous GVHD, to evaluate the role of photochemotherapy in the treatment of acute graft-versus-host disease.

Correspondence: Dr H Einsele, Department of Hematology and Oncology, University of Tübingen, Otfried Müller Str 10, 72076 Tübingen, Germany
Received 16 April 1998; accepted 20 August 1998

Patients and methods

Patients

Twenty patients (10 male, 10 female) received PUVA therapy as treatment for acute cutaneous GVHD after allogeneic bone marrow transplantation (nine patients with CML, five AML, two ALL, two MDS, one CLL, one chronic myeloproliferative syndrome). Thirteen patients had undergone allogeneic transplantation from a matched sibling donor and seven from a matched unrelated donor between 1995 and 1997. All patients developed cutaneous acute GVHD; six out of these 20 patients showed additional organ manifestations (liver, gut).

Conditioning regimens

Eleven patients received busulfan (16 mg/kg) and cyclophosphamide (120 mg/kg), and in addition one patient received melphalan (140 mg/m^2), and one patient received cytarabine (2×1 g/m^2 for 2 consecutive days).

Five patients underwent TBI (12 Gy) with cyclophosphamide (120 mg/kg), and in addition one patient received VP-16 (40 mg/kg),and another received thiotepa (10 mg/kg).

Supportive care

Patients were admitted to the hematologic intensive care unit and transferred to a laminar air unit on the day of transplantation. Supportive infusion therapy consisting of parenteral feeding and vitamins was administered through a tunneled Hickman catheter. For antimicrobial prophylaxis, patients received antibiotics (ciprofloxacin, $2 \times$ 500 mg/day), antifungal (fluconazol, 400 mg/day) and antiviral (acyclovir, 4×400 mg p.o./day) medication and immunoglobulin infusions (0.25 mg/kg polyvalent immunoglobulin every 3 weeks).

GVHD prophylaxis

All patients received CsA (5 mg/kg/day intravenously for the first 20 days followed by oral medication) from day −1 until at least day +100; in addition, four patients received a short course of methotrexate (15 mg/m^2 day 1, 10 mg/m^2 day 3 and 6); one patient received prednisolone (1 mg/kg/day) from day +3 and five patients received antithymocyte globulin (ATG Fresenius, 20 mg/kg day −4 to day −2).

GVHD staging

Clinical staging of acute GVHD was performed as described previously.[12] In two patients, cutaneous GVHD was confirmed by histopathology of the skin and in three patients by histopathology of intestinal and/or liver biopsy. In addition, autopsy confirmed acute GVHD in another three patients.

To better quantify the cutaneous lesions prior to PUVA therapy and thus to better assess local control of acute GVHD, a new scoring system was introduced. Extent of cutaneous manifestations of aGVHD was quantified by: intensity of erythema (0, no erythema; 1, slight erythema; 2, clearly noticable erythema; 3, marked erythema) × %body surface + size of bullae (0, no bullae; 4, small bullae; 5, big bullae) × %body surface affected.

GVHD therapy

For initial therapy of acute GVHD, all patients received prednisolone (2 mg/kg). In patients with progression of cutaneous GVHD after 3 days of standard dose prednisolone (2–5 mg/kg) additional PUVA therapy was initiated. Two hours after oral administration of 8-methoxypsoralen (0.6 mg/kg), UVA treatment was started and given four times/week. The initial dose varied between 0.3 and 0.5 J/cm^2. According to the response of cutaneous lesions, the dose was subsequently increased by 0.5 J/cm^2 increments to a maximal dose of 8.0 J/cm^2. After resolution of cutaneous symptoms and cessation of prednisolone treatment, PUVA therapy was administered twice a week for another 4 weeks until discontinuation. Patients not responding to PUVA and prednisolone therapy (2–5 mg/kg) received increasing doses of steroids (10 mg/kg–20 mg/kg). Non-responders to high-dose steroid therapy treatment received OKT3 therapy (5 mg/day for 10 days).

Results

Twenty patients with acute cutaneous GVHD were treated with PUVA therapy. GVHD was diagnosed by typical clinical manifestations, and additionally, by histopathological analysis in eight patients. Demographic data are shown in Table 1. Fourteen patients developed acute GVHD grade II, five patients grade III and one patient grade IV. Six out of 20 patients showed involvement of the liver and gut by acute GVHD.

Two patients with rapid progression of acute GVHD when receiving high dose corticosteroid treatment (20 mg/kg) were treated with OKT3 therapy. One patient with an initial response to corticosteroids (2 mg/kg) plus PUVA therapy had a severe relapse of cutaneous acute GVHD when PUVA was stopped. This patient refused further PUVA therapy and was treated with FK 506.

To better quantify the skin lesions, they were assessed by a new scoring system. The mean score at the beginning of PUVA therapy was 168 (range, 30–388). The mean maximal dose of prednisolone for treatment of acute GVHD was 8 mg/kg/day (range, 2–20 mg/kg/day).

PUVA therapy was well tolerated, with only two patients developing erythema following UV-A light exposure. Irradiation data are shown in Table 2.

Fifteen of 20 patients (75%) responded by score classification, clinical GVHD staging, and prednisolone reduction. The response rate was 92% (12/13 patients) in patients with acute GVHD confined to the skin. In contrast, only 33% of the patients (2/6) with additional organ manifestations (liver/gut) showed a response to PUVA therapy and prednisolone. The average time to a 50% prednisolone reduction was 35 days (range, 5–133 days). The average time to a 50% reduction according to the scoring system described above was 39 days (range, 10–86 days; Table 3). Nine of 20 patients died of relapse, infectious complications, or progression of hepatic or intestinal acute GVHD.

Discussion

For many years psoralen and UV-A light have been used in the treatment of chronic cutaneous GVHD after allogeneic transplantation.[1–10] Few patients have received PUVA therapy for acute cutaneous GVHD.[6–11] Furthermore, there are few reports of successful treatment of acute GVHD with extracorporal photopheresis.[13,14] At our institution, 20 patients with acute cutaneous GVHD after allogeneic stem cell transplantation were treated with PUVA therapy in an uncontrolled and unmatched study, with a response rate of 76%.

Acute GVHD is considered as a multi-step process that consists of an initial afferent phase (antigen recognition and T cell activation), followed by an amplification phase (clonal proliferation/differentiation and cytokine release) resulting in an efferent phase of tissue damage.[15,16] Transplant conditioning regimens cause tissue damage (e.g. intestinal mucosa), leading to release of inflammatory cytokines such as TNF-alpha and Il-1 by activated host cells. It has been shown that an increase of the TBI dose or high TNF-alpha levels during conditioning is associated with an increased risk of GVHD development and mortality.[17,18] Secretion of inflammatory cytokines may upregulate MHC antigens and further facilitate host recognition by donor T cells. T cell activation results in a proliferation of Th1 T cells with secretion of IL-2 and IFN-gamma, which induce further T cell expansion, cytotoxic T cell and natural killer cell response, resulting in the effector phase of target organ damage.

Although the exact mechanism of PUVA therapy remains unclear, there are *in vivo* and *in vitro* data which show immunological effects that might influence the cascade of events and the cytokine dysregulation during the development of acute GVHD.

Table 1 Characteristics of patients receiving PUVA therapy as a treatment for acute GVHD

PN	*Age*	*Sex*	*Diagnosis*	*Conditioning regimen*	*Donor*	*GVHD prophylaxis*	*Stage of GVHD liver/gut: +*	*GVHD therapy*	*Max. prednisolone dose (mg/kg/day)*
1	21	M	AML	TBI/CY	id sib	CsA	II	Pred	6
2	57	M	CML	BU/CY	id sib	CsA + MTX	II	Pred	5
3	49	F	CML	BU/CY	id sib	CsA	III+	Pred	5
4	31	F	AML	BU/CY	id unrel	CsA + ATG	II	Pred	7
5	38	M	CML	BY/CY	id sib	CsA	II+	Pred	2
6	20	M	ALL	TBI/CY	id unrel	CsA	III+	Pred + OKT3	20
7	18	M	CML	BU/CY	id unrel	CsA + MTX	IV+	Pred + OKT3	20
8	35	M	CML	BU/CY	id sib	CsA + MTX	II	Pred	5
9	34	F	CML	BU/CY	id unrel	CsA + MTX	II	Pred	10
10	10	F	AML	BU/CY/Melph	id sib	CsA	II	Pred	15
11	36	M	CMPS	TBI/CY	id unrel	CsA	III+	Pred	10
12	30	M	CML	BU/CY	id sib	CsA	II	Pred	20
13	28	F	AML	TBI/VP-16/CY	id unrel	CsA + Pred	III	Pred	7
14	8	F	MDS	TBI/Thiotepa/CY	id unrel	CsA + ATG	III+	Pred	5
15	30	F	MDS	BU/Alexan/CY	id sib	CsA	II	Pred	2
16	42	F	CML	BU/CY	id sib	CsA	II	Pred	2
17	45	F	AML	BU/CY	id sib	CsA	II	Pred	2
18	42	M	ALL	TBI/CY	id sib	CsA/ATG	I	Pred	2
19	45	M	CLL	TBI/CY	id sib	CsA/ATG	II	Pred + FK506	9
20	40	F	CML	BU/CY	id sib	CsA/ATG	II	Pred	5

PN = patient number; CMPS = chronic myeloproliferative syndrome; pred = prednisolone; ATG = antithymocyte globulin; melph = melphalan.

Table 2 Irradiation data of patients receiving PUVA therapy as a treatment for acute GVHD

PN	*UVA dose (J/cm^2)*	*PUVA treatments*	*Total UVA dose (J/cm^2)*	*Comments*
1	0.5–7.0	51	239	
2	0.3–5.0	68	117	
3	0.5–8.0	41	169	
4	0.3–8.8	>75	>103	Interruption due to pneumothorax, ongoing treatment
5	0.3	6	1.8	
6	0.5–3.5	12	24	
7	0.5–1.0	3	2.5	
8	0.3–1.0	8	6	
9	0.3–?	>8	>7	Continuation of PUVA at another institution
10	0.5–7.0	30	119	
11	0.5–1.0	3	2.5	
12	0.5–4.5	45	153	
13	0.3–6.5	86	416	Cessation due to relapse
14	0.5–7.0	53	211	
15	0.3–3.0	114	188	PUVA induced erythema, cessation due to relapse
16	0.5–3.5	67	193	
17	0.5–4.0	91	342	
18	0.5–1.0	49	49	
19	0.5–4.0	18	42	
20	0.5–	>71	>319	Ongoing PUVA therapy

PN = patient number.

Several experimental models have been used to examine the influence of PUVA therapy on lymphocyte function. Perez *et al*[19] have shown that infusion of PUVA-treated splenocytes from BALB/c mice grafted with CBA/J mouse skin into syngeneic mice induces tolerance to CBA/J major histocompatibility complex (MHC) antigens. Recipients were unresponsive in mixed lymphocyte reactions and cytotoxicity assays. In addition, they had no delayed-type hypersensivity against CBA/J cells, and rejection of CBA/J skin graft was delayed.[19] Laskin and co-workers[20] found that PUVA treatment of mouse T cells induces inhibition of mitogen-induced, as well as antigen-specific T cell proliferation. Bredberg and Forsgren[21] showed inhibition of chemotaxis of leukocytes following PUVA treatment.

Furthermore, modulation of the cytokine network following PUVA treatment has been documented in animal studies, as well as in cell culture. Aubin *et al*[22] reported that the activation of mouse keratinocytes with psoralen and UV-A irradiation induced the release of a soluble factor depressing alloreactivity. In a mouse model, Okamoto *et al*[23]

Table 3 Response to PUVA therapy in patients with acute GVHD

PN	*GVHD score at beginning of PUVA therapy*	*Maximal GVHD score*	*Stage of GVHD: skin (overall) liver/gut involvement: +*	*Time to 50% prednisolone reduction (days)*	*Time to 50% score reduction (days)*	*Follow-up (cause of death)*
1	165	165	3 (II)	24	18	Died (relapse)
2	140	140	2 (II)	17	28	Died (IA)
3	191	316	3 (III) +	133	60	Died (chronic GVHD)
4	54	192	2 (II)	5	86	Alive (chronic GVHD)
5	150	150	3 (II) +	—	—	Died (GVHD)
6	201	201	3 (III) +	—	—	Died (IA)
7	208	208	4 (IV) +	—	—	Died (GVHD, IA)
8	160	160	2 (II)	33	—	Died (listeria sepsis)
9	30	30	3 (II)	not evaluable	30	Alive (chronic GVHD)
10	180	180	2 (II)	12	13	Alive
11	300	300	3 (III) +	—	—	Died (IA)
12	100	150	3 (II)	66	69	Alive
13	388	388	3 (III)	35	28	Died (relapse)
14	200	200	3 (III) +	49	65	Alive
15	60	60	2 (II)	25	14	Alive (relapse)
16	94	94	2 (II)	6	30	Alive
17	180	180	3 (II)	64	30	Alive
18	40	40	1 (II)	14	10	Alive (relapse)
19	240	252	3 (II)	28	60	Alive
20	270	270	3 (II)	7	42	Alive (chronic GVHD)

PN = patient no; IA = invasive aspergillosis.

showed markedly suppressed production of IL-2 on day 3 with normalization of serum levels not before day 7 post-irradiation. Rivas and co-workers[24] demonstrated that exposure of murine keratinocyte cultures to UV irradiation caused a systemic suppression of delayed-type hypersensitivity by release of IL-10. Several studies show a marked influence of PUVA therapy on Langerhans cells, not only by a reduction of the cell number, but by a down-modulation of antigen-presenting function and a reduction of cytokine release.[25–30] UV-B light exposure has been reported to convert Langerhans cells from immunogenic to tolerogenic antigen-presenting cells, defective in their autocrine Il-2 production.[30]

Due to PUVA induced reduction of the number of Langerhans cells in the skin, the alteration of their antigen-presenting function and the modulation of cytokine secretion, PUVA therapy might be effective not only in chronic, but also in acute GVHD.

As documented by clinical improvement, GVHD score, and reduction in the dose of steroids administered, 92% of our patients with acute cutaneous GVHD responded to PUVA therapy. In all but two patients with additional organ manifestations of acute GVHD, no significant response and thus no reduction of the dose of corticosteroids could be achieved. Thus, in our opinion PUVA therapy should be restricted to patients with predominantly cutaneous manifestations of acute GVHD.

The newly established scoring system allows a better assessment of cutaneous GVHD and its response to therapy. PUVA therapy was well tolerated and effective in 92% of patients with acute cutaneous GVHD. No late complications, such as cutaneous squamous cell carcinoma or severe hyperlipidemia[31,32] were observed in our patients, but the brief follow-up period does not allow firm conclusions. No severe side-effects occurred during or following PUVA treatment.

Clearly, randomised prospective studies are warranted and are now underway to further assess the value of PUVA therapy for acute cutaneous GVHD.

References

1 Hymes SR, Morison WL, Farmer ER *et al*. Methoxalen and ultraviolet-A radiation in treatment of chronic cutaneous graft-versus-host reaction. *J Am Acad Dermatol* 1985; **12**: 30–37.
2 Volc-Platzer B, Hönigsmann H, Hinterberger W, Wolff K. Photochemotherapy improves chronic cutaneous graft-versus-host disease. *J Am Acad Dermatol* 1990; **23**: 220–228.
3 Jampel RM, Farmer ER, Vogelsang B *et al*. PUVA therapy for chronic cutaneous graft-versus-host disease. *Arch Dermatol* 1991; **127**: 1673–1678.
4 Kapoor N, Pellegrini AE, Copelan EA *et al*. Psoralen plus ultraviolet (PUVA) in the treatment of chronic graft-versus-host disease: preliminary experience in standard treatment resistant patients. *Semin Hematol* 1992; **29**: 108–112.
5 Vogelsang GB, Wolff D, Altomonte V *et al*. Treatment of chronic graf-versus-host disease with ultraviolet irradiation and psoralen (PUVA). *Bone Marrow Transplant* 1996; **17**: 1061–1067.
6 Eppinger T, Ehninger G, Steinert M *et al*. 8-Methoxypsoralen and ultraviolet-A therapy for cutaneous manifestations of graft-versus-host disease. *Bone Marrow Transplant* 1990; **50**: 807–811.
7 Reinhauer S, Lehmenn P, Plewig G *et al*. Photochemotherapie (PUVA) der akuten Graft-versus-Host Erkrankung. *Hautarzt* 1993; **44**: 708–712.
8 Torras H, Martin-Ortega E, Carreras E *et al*. UVA and PUVA therapy in the treatment of cutaneous graft-versus-host disease (GVHD). *Eur J Dermatol* 1993; **3**: 447–451.

9 Aubin F, Brion A, Deconinck E *et al*. Phototherapy in the treatment of cutaneous graft-versus-host disease. *Transplantation* 1995; **59**: 151–155.
10 Atkinson K, Weller P, Ryman W, Biggs J. PUVA therapy for drug-resistant graft-versus-host disease. *Bone Marrow Transplant* 1986; **1**: 227–236.
11 Aschan J. Treatment of moderate to severe acute graft-versus-host disease: a retrospective analysis. *Bone Marrow Transplant* 1994; **14**: 601–607.
12 Glucksberg H, Storb R, Fefer A *et al*. Clinical manifestation of graft-versus-host disease in human recipients of marrow from HLA-matched sibling donors. *Transplantation* 1974; **18**: 295–304.
13 Rossetti F, Dall'Amico R, Crocetti G *et al*. Extracorporal photochemotherapy for the treatment of graft-versus-host disease. *Bone Marrow Transplant* 1996; **18** (Suppl.) 175–181.
14 Owsianowski M, Gollnick H, Siegert W *et al*. Successful treatment of chronic graft-versus-host disease with extracorporal photopheresis. *Bone Marrow Transplant* 1994; **14**: 845–848.
15 Ferrara JLM, Cooke KR, Pan L *et al*. The immunopathophysiology of acute graft-versus-host disease. *Stem Cells* 1996; **14**: 473–489.
16 Klingebiel T, Schlegel PG. GVHD: overview on pathophysiology, incidence, clinical and biological features. *Bone Marrow Transplant* 1998; **21** (Suppl. 2): 45–49.
17 Hill GR, Crawford JM, Cooke KR *et al*. Total body irradiation and acute graft-versus-host disease: the role of gastrointestinal damage and inflammatory cytokines. *Blood* 1997; **90**: 3204–3213.
18 Holler E, Kolb HJ, Mittermuller J *et al*. Modulation of acute graft-versus-host disease after allogeneic bone marrow transplantation by tumor necrosis factor alpha (TNF-α) release in the course of pretransplant conditioning: role of conditioning regimens and prophylactic application of a monoclonal antibody neutralizing human TNF alpha (MAK 195F). *Blood* 1995; **86**: 890–899.
19 Perez M, Edelson R, Laroche L *et al*. Inhibition of antiskin allograft immunity by infusions with syngeneic photoinactivated effector lymphocytes. *J Invest Dermatol* 1989; **92**: 670–676.
20 Laskin JD, Mermelstein FH, Heindel ND, Ron Y. Selective inactivation of lymphocytes after psoralen/ultraviolet light (PUVA) treatment without affecting systemic immune responses. *J Leukocyte Biol* 1993; **54**: 138–144.
21 Bredberg A, Forsgren A. Effects of *in vitro* PUVA on human leukocyte function. *Br J Dermatol* 1984; **111**: 159–168.
22 Aubin F, Kripke ML, Ullrich SE. Activation of keratinocytes with psoralen plus UVA radiation induces the release of soluble factors that suppress delayed and contact hypersensitivity. *J Invest Dermatol* 1991; **97**: 995–1000.
23 Okamoto H, Horio T, Maeda M. Alteration of lymphocyte functions by 8-methoxypsoralen and long-wave ultraviolet radiation. II. The effect of *in vivo* PUVA on IL-2 production. *J Invest Dermatol* 1987; **89**: 24–26.
24 Rivas JM, Ullrich SE. Systemic suppression of delayed-type hypersensitivity by supernatants from UV-irradiated keratinocytes. An essential role for keratinocyte-derived IL-10. *J Immunol* 1992; **149**: 3865–3871.
25 Ashworth J, Kahan MC, Breathnach SM. PUVA therapy decreases HLA-DR$^+$ CD1a$^+$ Langerhans cells and epidermal cell antigen-presenting capacity in human skin, but flow cytometrically sorted residual HLA-DR$^+$ CD1a$^+$ Langerhans cells exhibit normal alloantigen-presenting function. *Br J Dermatol* 1989; **120**: 329–339.
26 Okamoto H, Horio T. The effect of 8-methoxypsoralen and long-wave ultraviolet light on Langerhans cells. *J Invest Dermatol* 1981; **77**: 345–346.
27 Friedmann PS. Disappearance of epidermal Langerhans cells during PUVA therapy. *Br J Dermatol* 1981; **105**: 219–221.
28 Ree K. Reduction of Langerhans cells in human epidermis during PUVA therapy: a morphometric study. *J Invest Dermatol* 1982; **78**: 488–492.
29 Friedmann PS, Ford G, Ross J, Diffey BL. Reappearance of epidermal Langerhans cells after PUVA therapy. *Br J Dermatol* 1983; **109**: 301–307.
30 Simon JC, Tigelaar RE, Bergstresser PR *et al*. Ultraviolet B radiation converts Langerhans cells from immunogenic to tolerogenic antigen-presenting cells. Induction of specific clonal anergy in CD4$^+$ T helper cells. *J Immunol* 1991; **146**: 485–491.
31 Altman JS, Adler SS. Development of multiple cutaneous squamous cell carcinoma during PUVA treatment for chronic graft-versus-host disease. *J Am Acad Dermatol* 1994; **31**: 505–507.
37 Maravic N, Bogdanic V, Aurer I *et al*. Severe hyperlipidemia following treatment with PUVA for acute skin GVHD. *Bone Marrow Transplant* 1994; **14**: 173–181.