

Long-term Follow-up of Patients Treated in a Phase 2 Trial With MyVax® Personalized Immunotherapy (Recombinant Id-KLH Plus GM-CSF) After Chemotherapy as Initial Treatment for Follicular Non-Hodgkin's Lymphoma (NHL)

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ABSTRACT

Background: The tumor-specific variable regions of the clonal immunoglobulin (idiotype or Id) expressed by malignant B-cell NHL can be used as a target for active personalized immunotherapy. MyVax® Personalized Immunotherapy is a patient-specific, recombinant Id protein conjugated to keyhole limpet hemocyanin (KLH) and administered with granulocyte-macrophage colony-stimulating factor (GM-CSF). Several phase 2 clinical trials have been conducted to determine the immunogenicity of MyVax® Personalized Immunotherapy in NHL patients.

Patients and Methods: Patients must have had previously untreated follicular NHL and achieved at least a PR to either CVP chemotherapy (17 patients) or CVP + CHOP chemotherapy (5 patients) to be eligible to receive 5 immunizations with MyVax® Personalized Immunotherapy. Each immunization consisted of MyVax® Personalized Immunotherapy plus GM-CSF administered SQ on Day 1 and GM-CSF alone on Days 2-4. Immunizations were administered over 24 weeks. The first 4 immunizations were given monthly, and the 5th was given 12 weeks later.

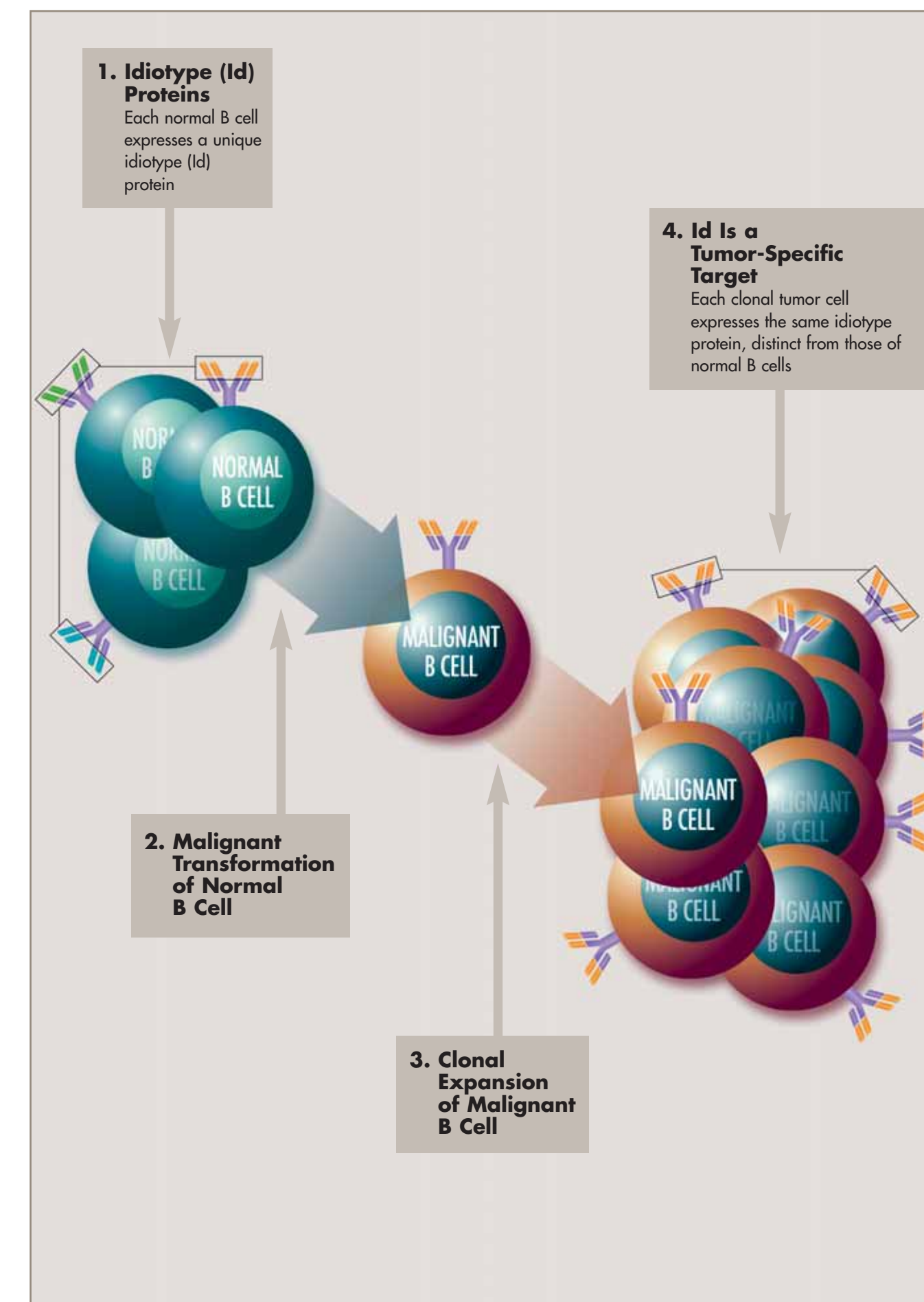
Results: Of 22 patients immunized, 21 were evaluable for immune responses (one received rituximab): 13 (62%) of 21 patients mounted idiotype-specific immune responses, 10 (48%) of 21 patients mounted humoral anti-Id responses, 7 (44%) of 16 patients evaluated mounted cellular anti-Id responses, and 4 (25%) of 16 mounted both. Humoral and T-cell immune responses were specific and recognized tumor-derived Id protein. Immunizations were safe and well tolerated, with adverse events limited to mild-to-moderate local injection-site reactions and flu-like symptoms. Follicular Lymphoma International Prognostic Index (FLIPI) scores indicated that 19 of the 21 patients (90%) were in the intermediate or high-risk categories. The median follow-up to date is more than 5.5 years, and median time to progression is 38 months from the end of chemotherapy. Nine of the 21 patients remain progression free between 57 months and 78 months postchemotherapy, 4 are in the high-risk FLIPI category.

Conclusions and Discussion: These results suggest that patients with a poorer prognosis according to their FLIPI score can achieve significant remissions following treatment with MyVax® Personalized Immunotherapy. MyVax® Personalized Immunotherapy is currently in a late-stage phase 3 trial.

INTRODUCTION

Tumor-specific immunoglobulin (idiotype, Id) represents a potential target for the immunotherapy of B-cell malignancies.¹

Figure 1. The idiotype protein is a tumor-specific target.



Traditionally, Id proteins have been produced by "rescue hybridization" or "heterohybridization production" (involving the fusion of lymphoma cells from the patient with a myeloma cell line). In studies in which patients with follicular NHL were immunized with first-generation personalized immunotherapy produced by rescue hybridization, anti-Id immune responses (humoral and T cell) were correlated with improved disease-free and overall survival,^{2,3} with tumor regression,^{4,5} and with complete molecular remissions.⁶

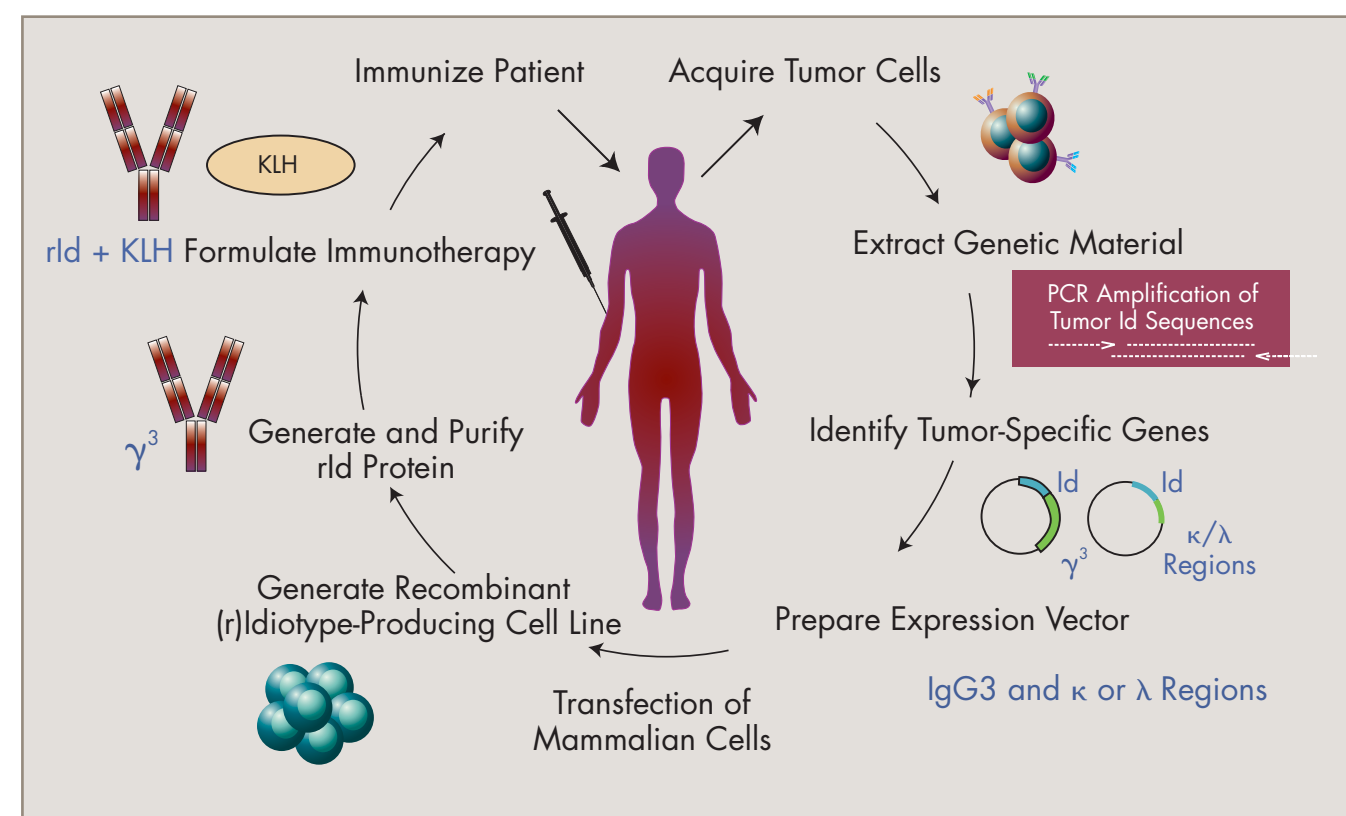
- Limitations of "rescue hybrid" method for Id protein production
 - Requires excisional biopsy
 - Requires viable tumor cells
 - Hybrids yield IgG or IgM form of Id (non-uniform)
 - Hybrids can lose Id production over time

Production of MyVax® Personalized Immunotherapy

The MyVax® Personalized Immunotherapy manufacturing process is based on recombinant gene amplification technology.

- Advantages of "molecular rescue" for patient-specific Id production
 - Requires only a tiny amount of tumor cells (FNA, core biopsy, bone marrow)
 - Does not require viable cells
 - Enables stable production of a uniform Id product

Figure 2. MyVax® Personalized Immunotherapy process.



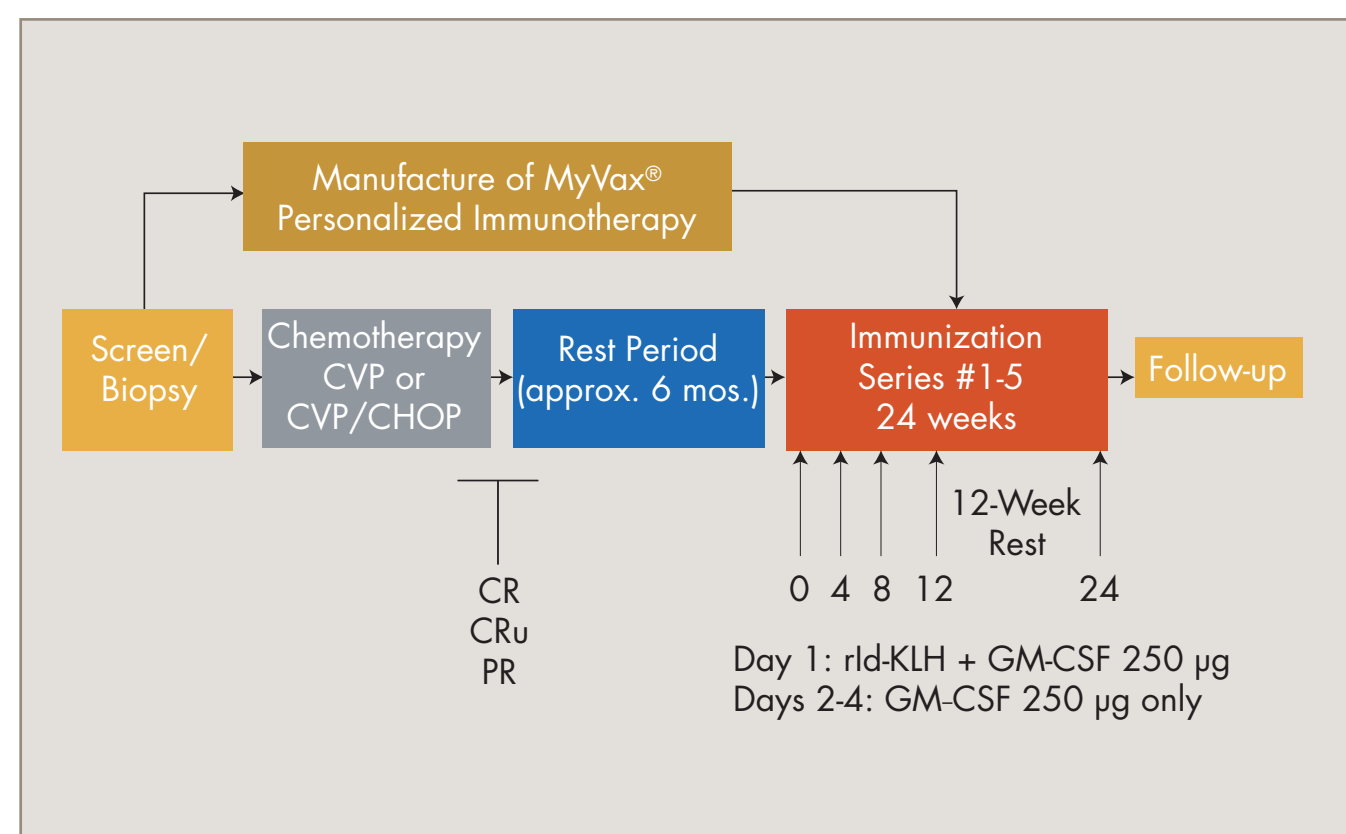
The purified rId protein is conjugated to KLH and administered with granulocyte-macrophage colony-stimulating factor (GM-CSF).

STUDY OBJECTIVES

- To determine the feasibility of producing recombinant idiotype (rId) proteins from follicular NHL specimens by recombinant gene amplification technology
- To determine the safety and immunogenicity of MyVax® Personalized Immunotherapy (rId-KLH + GM-CSF) following standard cytoreductive chemotherapy
- To characterize the immune response to rId proteins versus tumor (rescue hybrid)-derived Id proteins
- To evaluate correlation between time to disease progression (TTP), clinical prognostic factors, and immunologic outcomes

METHODS

Figure 3. Study design.



Immune Response Testing

- Humoral response
 - Serum anti-Id antibodies measured by EUSA
 - Post-immunization sera tested for antibodies against autologous tumor by flow cytometry
 - FcγRIII genotype determined by real-time PCR
- T-cell response
 - Cellular proliferative responses measured by proliferation (³H-thymidine incorporation) of peripheral blood mononuclear cells in response to relevant Id proteins

RESULTS

Patient Characteristics

- A total of 22 patients were treated with MyVax® Personalized Immunotherapy. One patient received rituximab before, during, and after immunization and was not evaluated for efficacy

| Characteristic | Patients (N=21) |
|---|-----------------|
| Age, years | 49.4 |
| Mean Range | 32-73 |
| Sex, n (%) | |
| Male | 11 (52%) |
| Female | 10 (48%) |
| Type of NHL, n (%) | |
| Follicular small cleaved cell (Grade 1) | 15 (71%) |
| Follicular mixed (Grade 2) | 6 (29%) |
| Disease stage, n (%) | |
| Stage III | 3 (14%) |
| Stage IV | 18 (86%) |
| FcγRIII genotype, n (%) | |
| V/V | 17 (81%) |
| V/V | 4 (19%) |
| FLIPI score | |
| 1 | 2 (10%) |
| 2 | 11 (52%) |
| 3 or 4 | 8 (38%) |

Patient Characteristics: Individual Subjects

| Patient | Age/Sex | Histology/Stage | FLIPI Score | Pre-Immuno-ization ChemoRx | Clinical Response Post-ChemoRx | Pre-Immuno-ization Clinical Status | Post-Immuno-ization Clinical Status (D=dead) | Time to Progression (months) | FcγRIII Genotype | Anti-Id Humoral Immune Response | Anti-Id Cellular Immune Response |
|---------|---------|-----------------|-------------|----------------------------|--------------------------------|------------------------------------|--|------------------------------|------------------|---------------------------------|----------------------------------|
| 1 | M/53 | FM/IVA | 1 | CVPx6 | CR | CR | CR | 78.7+ | V/V | Positive | Positive |
| 2 | F/44 | FSC/IVA | 3 | CVPx8, CHOPx2 | PR | CR | CR | 76.6+ | F/F | Positive | Negative |
| 3 | F/49 | FSC/IVA | 2 | CVPx8 | CR | CR | CR | 38 | V/V | Negative | Negative |
| 4 | F/37 | FSC/IVA | 3 | CVPx6 | PR | PR | PD | 12.5 | V/V | Negative | Negative |
| 5 | M/47 | FSC/IVA | 2 | 4 CVP, 4 CP | CR | CR | CR | 30.3 | F/F | Positive | Positive |
| 6 | M/32 | FSC/IVA | 2 | 5 CVP, 6 CHOP | CR | CRu | PD | 12.1 | V/V | Negative | Positive |
| 7 | M/51 | FSC/IVA | 2 | 7 CVP | CRu | CRu | CR | 68.7+ | V/V | Negative | Negative |
| 8 | F/51 | FSC/IVA | 2 | 8 CVP | CR | CRu | CR | 74.2+ | V/V | Positive | Negative |
| 9 | M/49 | FSC/IVA | 2 | 6 CVP | CR | CR | CR | 19.5 | F/F | Positive | Negative |
| 10 | M/55 | FSC/IVA | 3 | 6 CVP | CRu | CRu | CRu | 22.5, 0 | V/V | Positive | Positive |
| 11 | M/57 | FM/IVA | 3 | 8 CVP | CRu | CRu | PD | 16.4, 0 | V/V | Negative | Negative |
| 12 | M/55 | FSC/IVA | 2 | 8 CVP, 4 CHOP | PR | CRu | PD | 15.2 | F/F | Positive | Positive |
| 13 | F/52 | FSC/IVA | 2 | 6 CVP, 2 CHOP | PR | PR | PR | 22.1, 0 | F/F | Negative | Positive |
| 14 | M/48 | FSC/IVA | 2 | 6 CVP | PR | PR | CR | 68.6+ | V/V | Negative | Negative |
| 15 | F/67 | FSC/IVA | 3 | 4 CVP, 2 VP | PR | PR | PR | 64.9+ | V/V | Positive | Negative |
| 16 | M/44 | FSC/IVA | 3 | 8 CVP | PR | CRu | CR | 66.9+ | V/V | Negative | Positive |
| 17 | F/73 | FSC/IVA | 4 | 6 CVP | CR | PR | CR | 65+ | F/F | Negative | ND |
| 18 | F/39 | FM/IVA | 1 | 6 CVP | CR | CR | PD | 15 | V/V | Negative | ND |
| 19 | M/68 | FM/IVA | 3 | 6 CVP | CR | CR | CR | 60.9 | F/F | Positive | ND |
| 20 | M/40 | FM/IVA | 2 | 6 CVP, 4 CHOP | PR | CRu | CRu | 57.5+ | V/V | Negative | ND |
| 21 | F/41 | FM/IVA | 2 | 6 CVP | CR | CR | PD | 12.7 | V/V | Positive | ND |

Safety

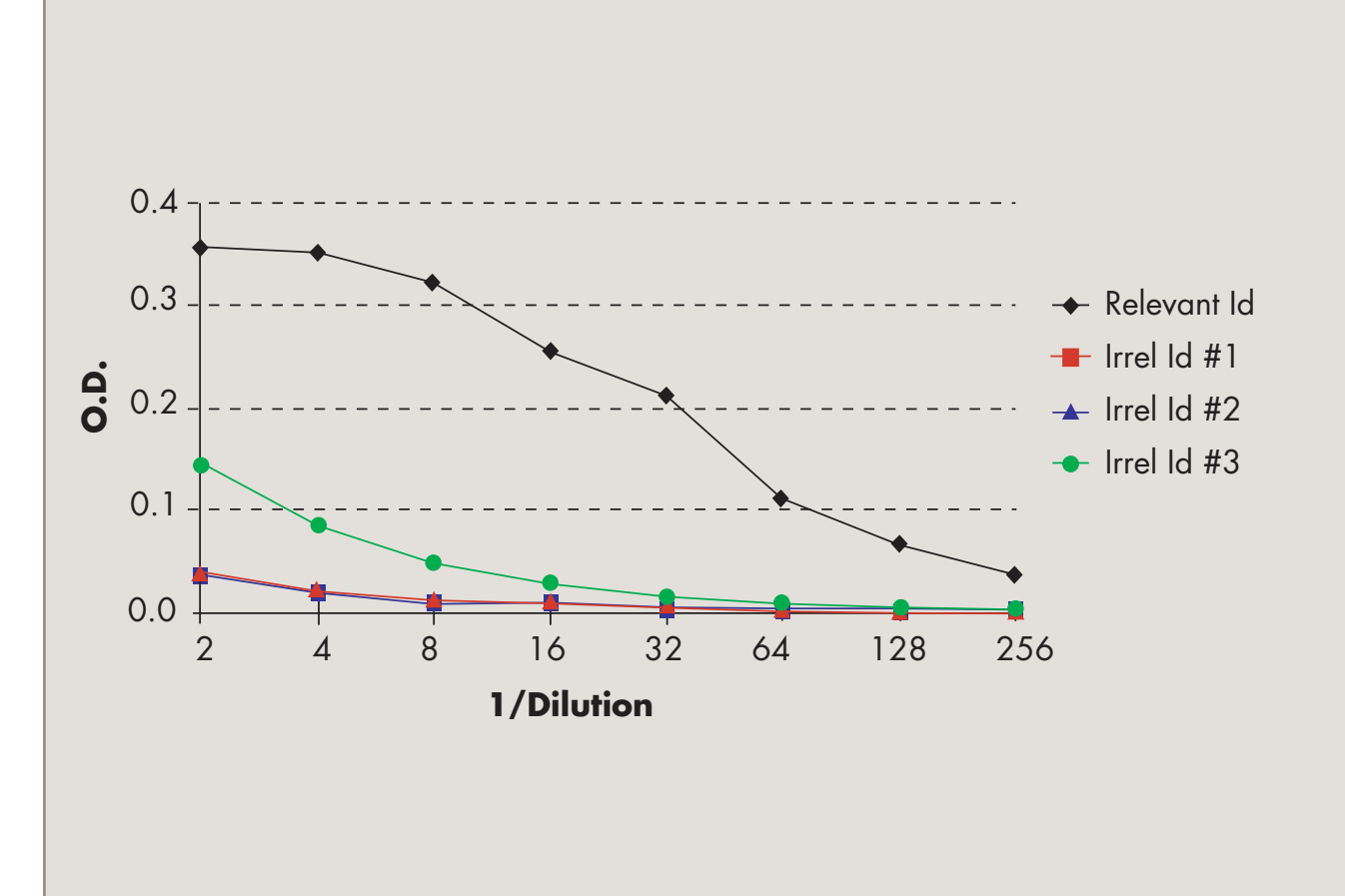
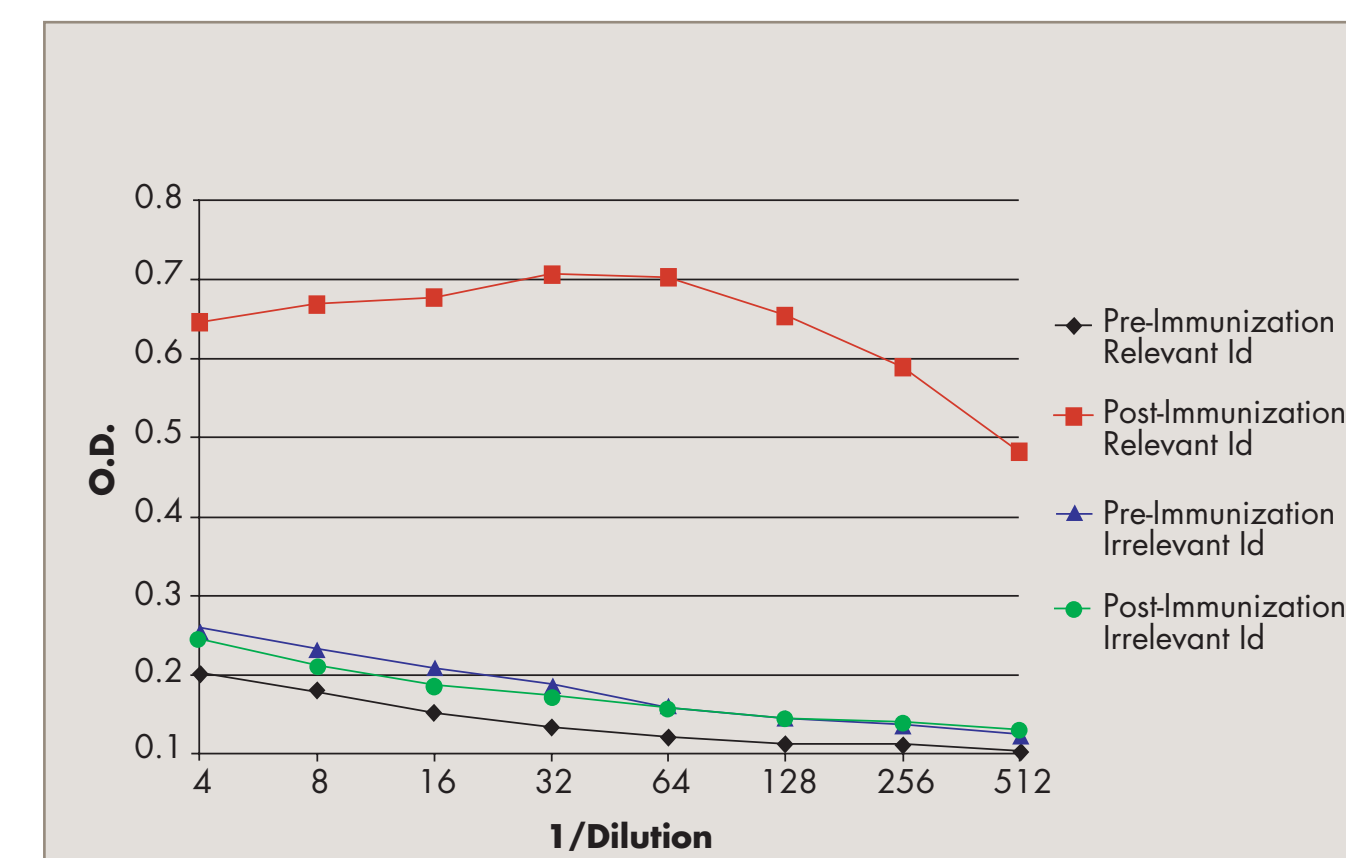
- Grade 1 or 2 injection-site reactions were reported by all 22 patients
- The majority of the adverse events were mild to moderate in severity

| Adverse Events | Patients (N=22) |
|--------------------------|-----------------|
| Injection-site reactions | |
| - Erythema | 100% |
| - Induration | 86% |
| - Pain | 86% |
| - Pruritus | 64% |
| - Bruising | 32% |
| - Inflammation | 27% |
| Headache | 55% |
| Fatigue | 45% |
| Back pain | 36% |
| Pain NOS | 32% |
| Nasopharyngitis | 32% |
| Arthralgia | 27% |
| Influenza-like illness | 23% |
| Myalgia | 23% |

Immune Responses

- 21 patients were evaluable
- Immune responses (humoral, T cell, or both): 13 (62%) patients
 - Humoral response: 10/21 patients (48%)
 - T-cell response: 7/16 patients (44%)
 - Both humoral and cellular responses: 4/16 patients (25%)
 - Immune responses by preimmunization tumor status
 - CR or CRu: 11/17 responders (65%)
 - PR: 2/5 responders (40%)
- Immune responses were specific
 - Serum anti-Id IgG antibodies recognized tumor-derived Id (measured by EUSA)

Figure 4. Specific anti-Id humoral response.



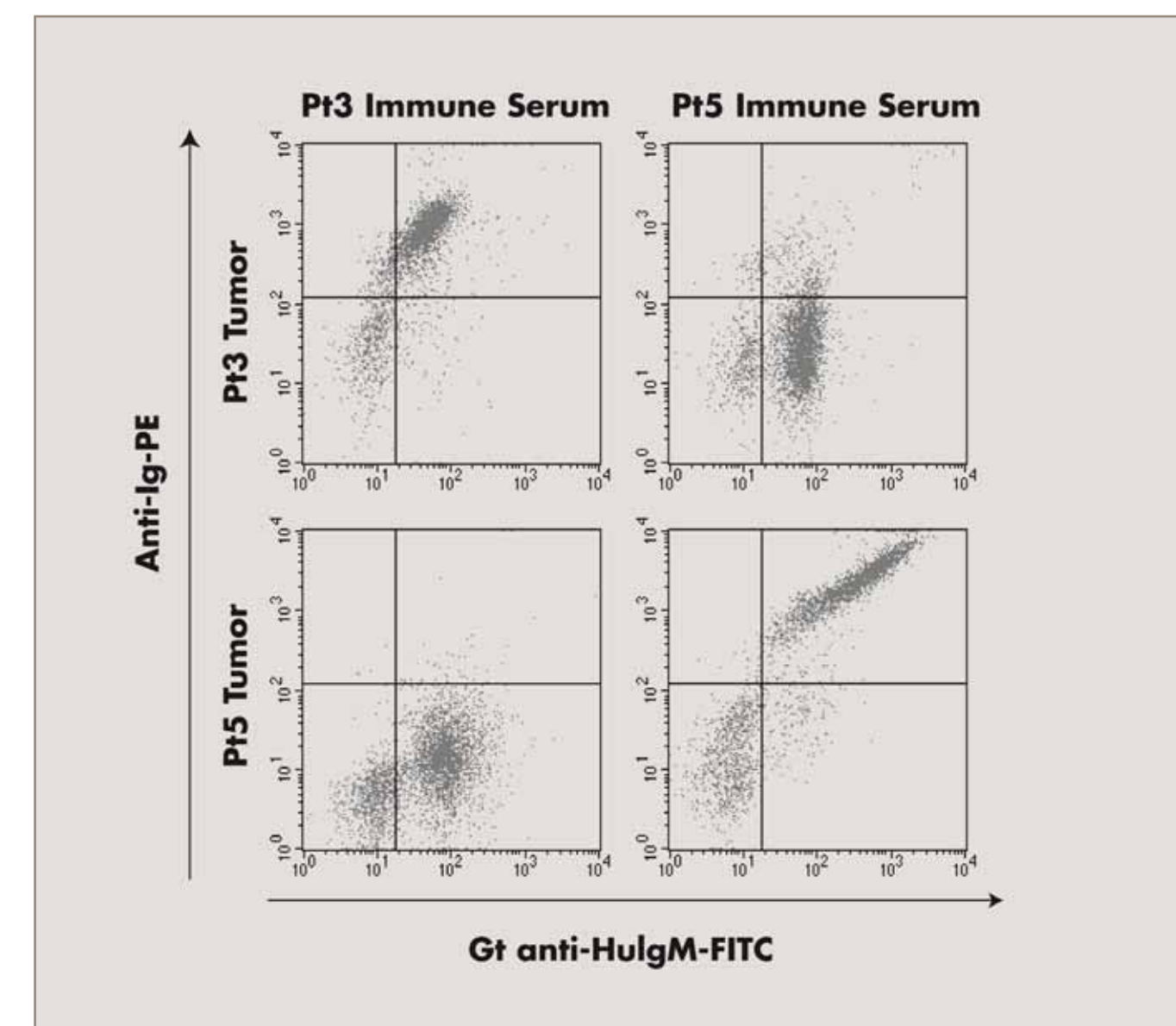
- Id proteins derived from traditional tumor rescue hybridomas were available for some patients in order to verify that immune responses to rId were relevant to the native Id protein as present on tumor cells. In patients with positive humoral reactivity to rId, reactivity to rescue hybridoma-derived Id was also demonstrated

Figure 5. The humoral response to rId reacts with native Id protein derived directly from tumor cells (rescue hybridomas).

| Pt | Reactivity to rId | Reactivity to Tumor Id |
|----|-------------------|------------------------|
| 2 | Positive | Positive |
| 7 | Positive | Positive |
| 10 | Positive | Positive |
| 11 | Positive | Positive |
| 15 | Positive | Positive |

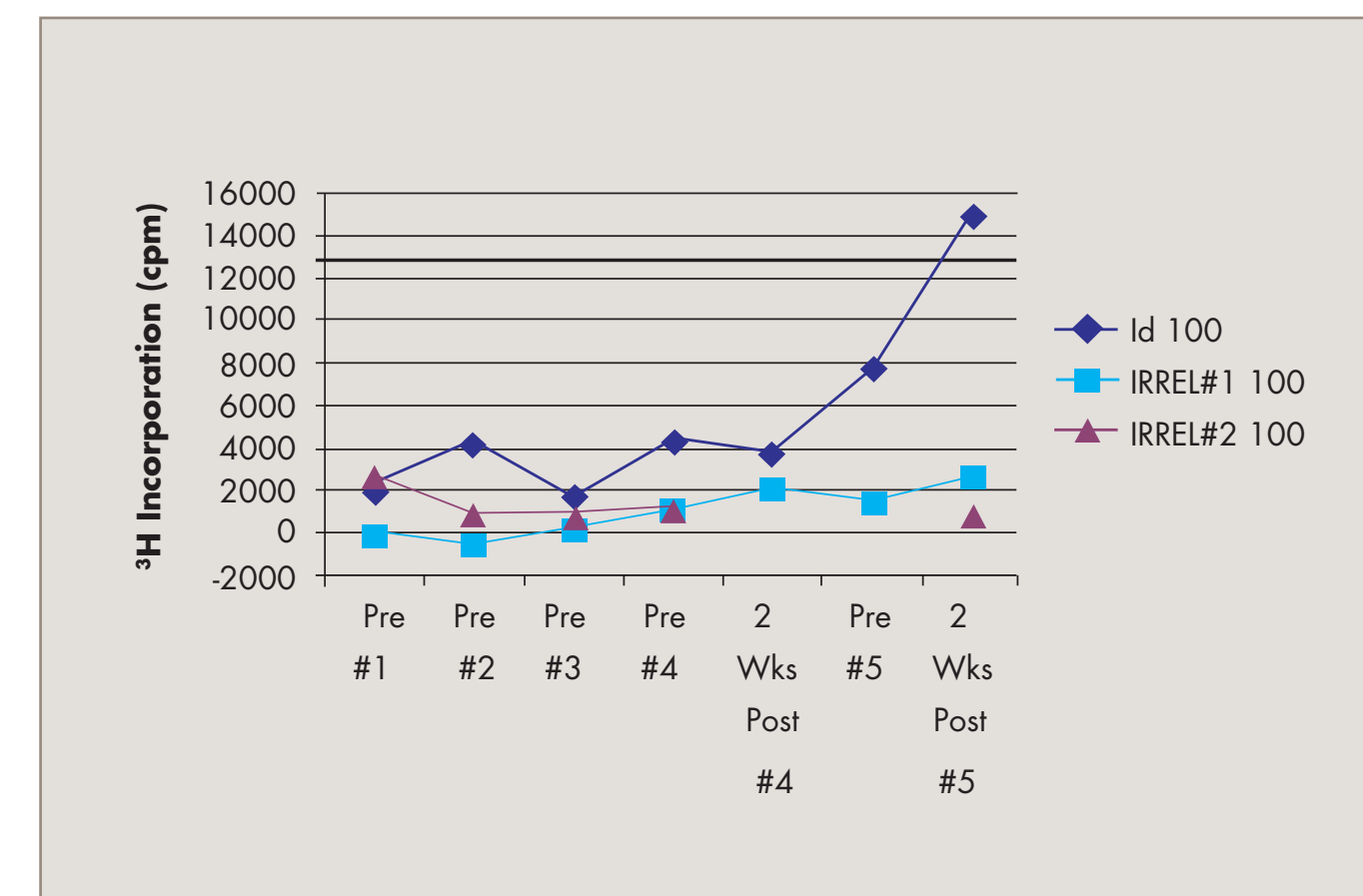
- Postimmunization sera specifically stained autologous tumor cells (measured by flow cytometry)

Figure 6. Anti-Id antibodies induced by rId-KLH immunization specifically recognize autologous tumor cells.



- Postimmunization anti-Id T-cell proliferative response was specific to relevant Id protein

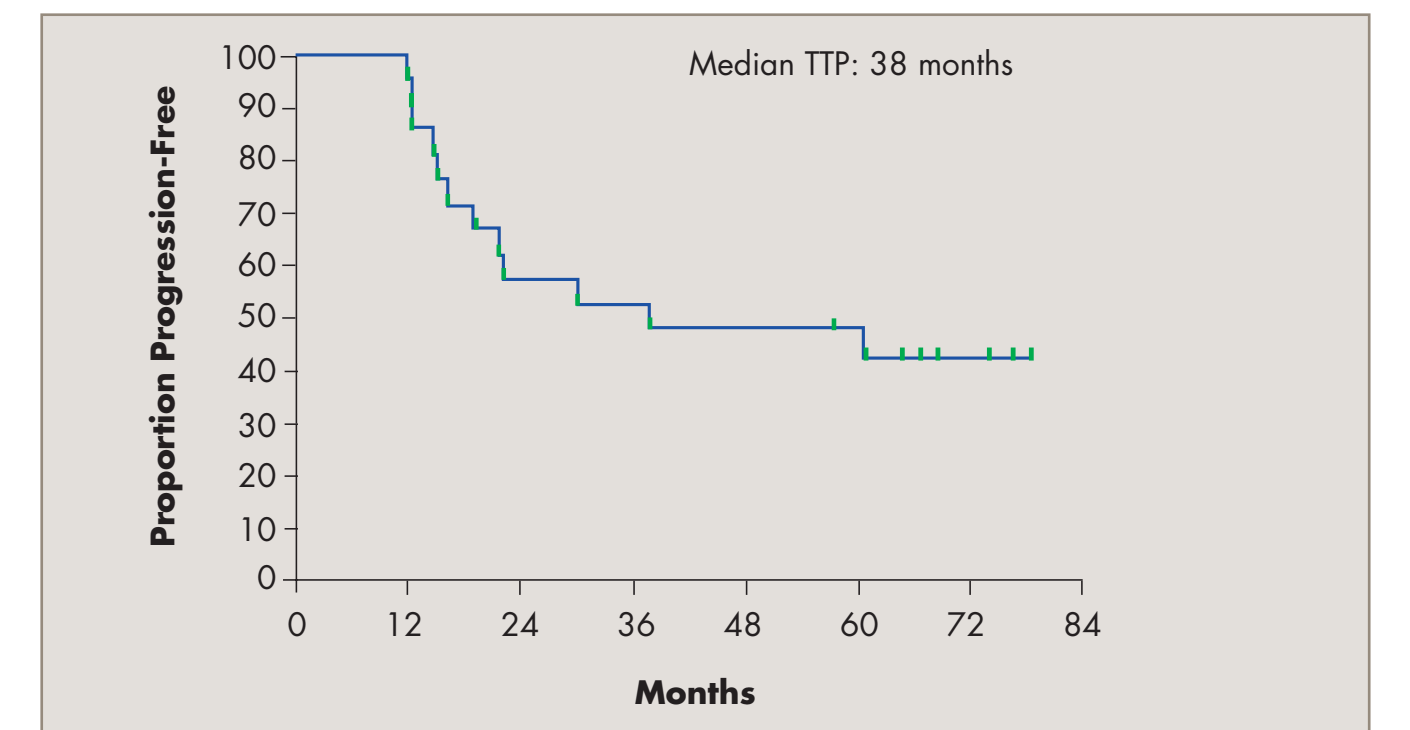
Figure 7. T-cell proliferative anti-Id response post-immunization.



Time to Disease Progression

- Median time to progression (calculated from the end of chemotherapy): 38 months

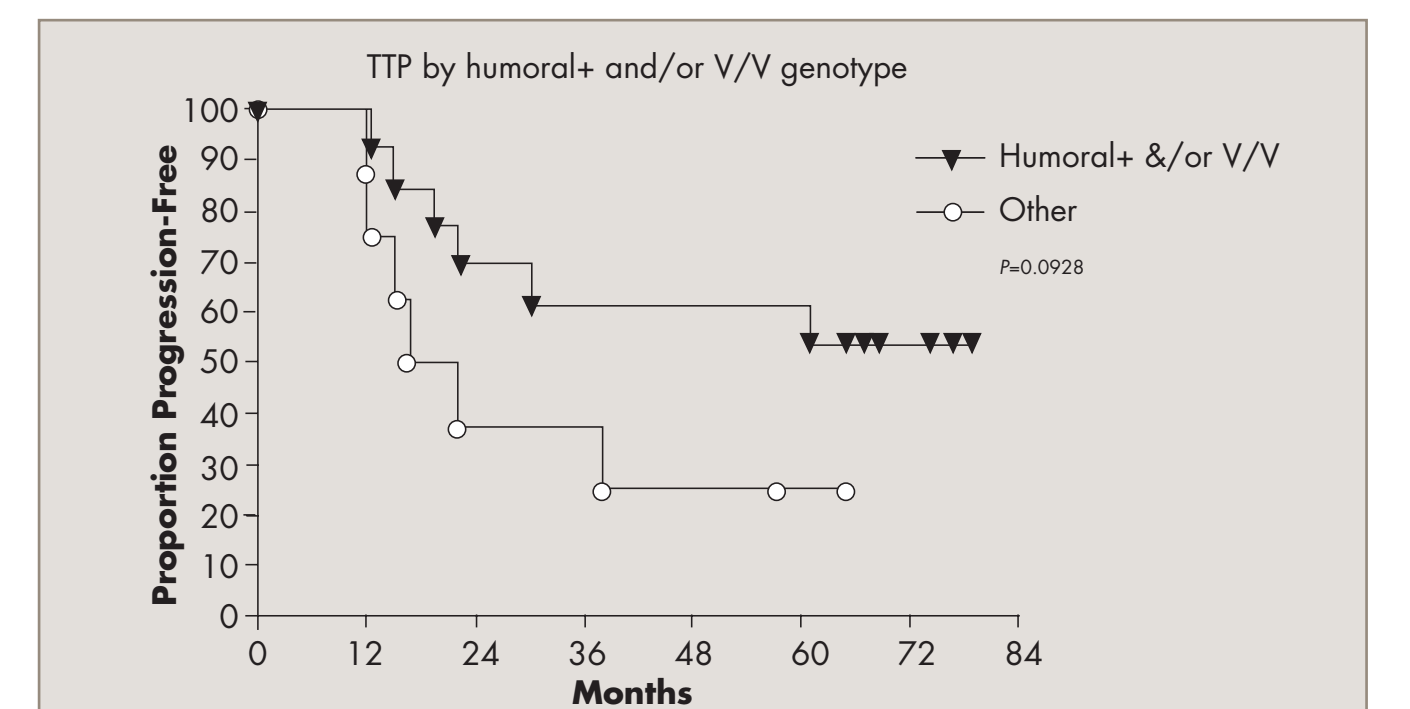
Figure 8. Time to disease progression.



- 9 of the 21 patients (43%) have remained in remission and have not required any additional therapeutic interventions for their lymphoma
 - Median time of follow-up: more than 5.5 years
 - 4 of the patients that remain in remission are high-risk (FLIPI score ≥3)

- Increased time to progression was correlated with humoral anti-Id responses and/or the V/V polymorphism of the Fc receptor γIII (FcγRIII; associated with enhanced antibody-dependent cellular cytotoxicity (ADCC))⁷

Figure 9. Humoral anti-Id immune response and favorable FcγRIII genotype correlated with remission duration.



CONCLUSIONS

- MyVax® Personalized Immunotherapy can be routinely produced from various types of follicular lymphoma specimens
- Immunization with MyVax® Personalized Immunotherapy + GM-CSF:
 - Demonstrated a favorable safety profile
 - Induced specific anti-Id immune responses in 62% of immunized patients
- Immunization with MyVax® Personalized Immunotherapy + GM-CSF can convert patients with PR after chemotherapy to CR after immunization
- Immunization with MyVax® Personalized Immunotherapy + GM-CSF can achieve significant remissions after immunization in patients with poor prognosis by FLIPI score
- A second planned interim analysis of data from a pivotal phase 3 trial of MyVax® Personalized Immunotherapy in previously untreated follicular NHL patients will be performed in mid 2006

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