Torque-Teno Viral Load – A New Tool to Predict Donor-Specific Antibodies in Pediatric Heart Transplantation Recipients

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Background and Aim

In pediatric solid organ transplantation, the individual clinical effect of immunosuppressive medication varies despite standardized and trough level controlled immunosuppressants.
The quality of the immunosuppressive regime is the pivotal point of graft survival as inadequately low immunosuppression poses the risk of graft rejection or vasculopathy and inadequately high immunosuppression leads to recurrent infections and risk of post-transplant lymphoproliferative disease.
The implementation of immunosuppressive standards is mainly guided by medication trough levels. However, there seems to be high interindividual variance of clinical immunosuppression in the presence of equal trough levels, leading to the need for further comprehensive markers for immunosuppression.
Torque-Teno Virus (TTV) is part of the human virome in up to 90% of cases, without pathogenicity and insensitive to antiviral drugs. Viral load can be easily quantified via PCR and can be linked to the immunological changes of individual patients after introduction of immunosuppressive medication. Assessment of TTV has recently been proposed as a surrogate parameter for this purpose. We report the first worldwide experience with TT viral load in pediatric heart transplant recipients.

Patients and Methods

We retrospectively evaluated TT viral load in pediatric heart transplant recipients over the course of 6.5 years at our pediatric heart transplant center regarding the development of donor-specific antibodies (DSA) and biopsy-proven rejection.

Stadium of Immunosuppression	**I** (0-3 months)	**II** (4-6 months)	**III** (6-12 months)	**IV** (12-24 months)	**V** (>24 months)
Tacrolimus Target Range (ng/mL)	10 - 15	10-12	8-10	6-8	4-8
Tacrolimus Trough Level (ng/mL, median)	11.4 ± 4.75	10.55 ± 2.83	9.00 ± 3.04	7.4 ± 2.78	6.5 ± 2.87
TTV log10 (median)	7.00 ± 2.39 (range 0-9)	8.00 ± 1.88 (range 0 - 10)	8.00 ± 0.93 (range 6-10)	8.00 ± 1.58 (range 4-10)	7.00 ± 1.64 (range 4-9)

Table I: Tacrolimus trough levels compared to TT viral load in relation to time after transplantation and target immunosuppression

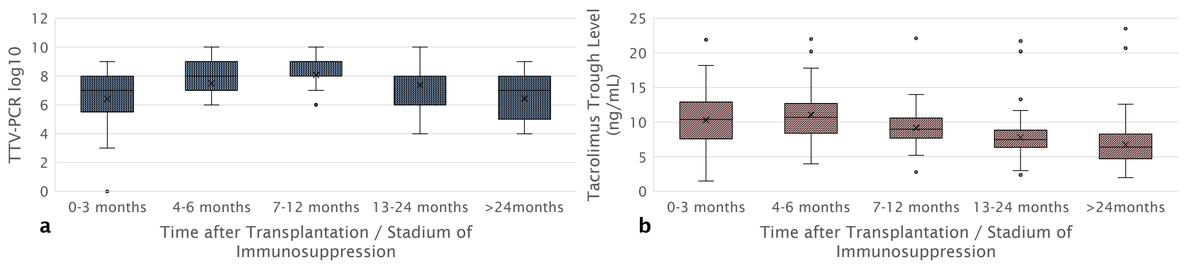

Figure I: **a** TTV load relative to time after transplantation. **b** Tacrolimus trough levels relative to time after transplantation

Results

365 TTV measurements from 44 pediatric heart transplant recipients were analyzed. Median viral load was 10^7 copies/mL ± 1.8 (IQR 5-8, range 0-10).

For all patients, per the center's standard protocol, induction with antithymoglobuline and intraoperative start of corticosteroids was administered, and postoperative therapy consisted of tacrolimus, mycophenolate mofetile and prednisone, the latter of which was tapered six months after transplantation.

Tacrolimus target and real-life trough levels are depicted in *Table I*, the development of Tacrolimus trough levels and TT viral load after transplantation is shown in *Figure I.*
While Tacrolimus levels decreased according to the adaptation of target ranges, TT viral load increased in the first months after transplantation, concurring with the delayed response after initiation of immunosuppression reported in the literature, and reached a plateau three months after transplantation. Starting 12 months after transplantation, TTV levels started to decrease, with high interindividual differences and widening of interquartile ranges. Accordingly, there was no significant correlation of Tacrolimus levels and TT viral load even after exclusion of recently transplanted patients (<3 months).

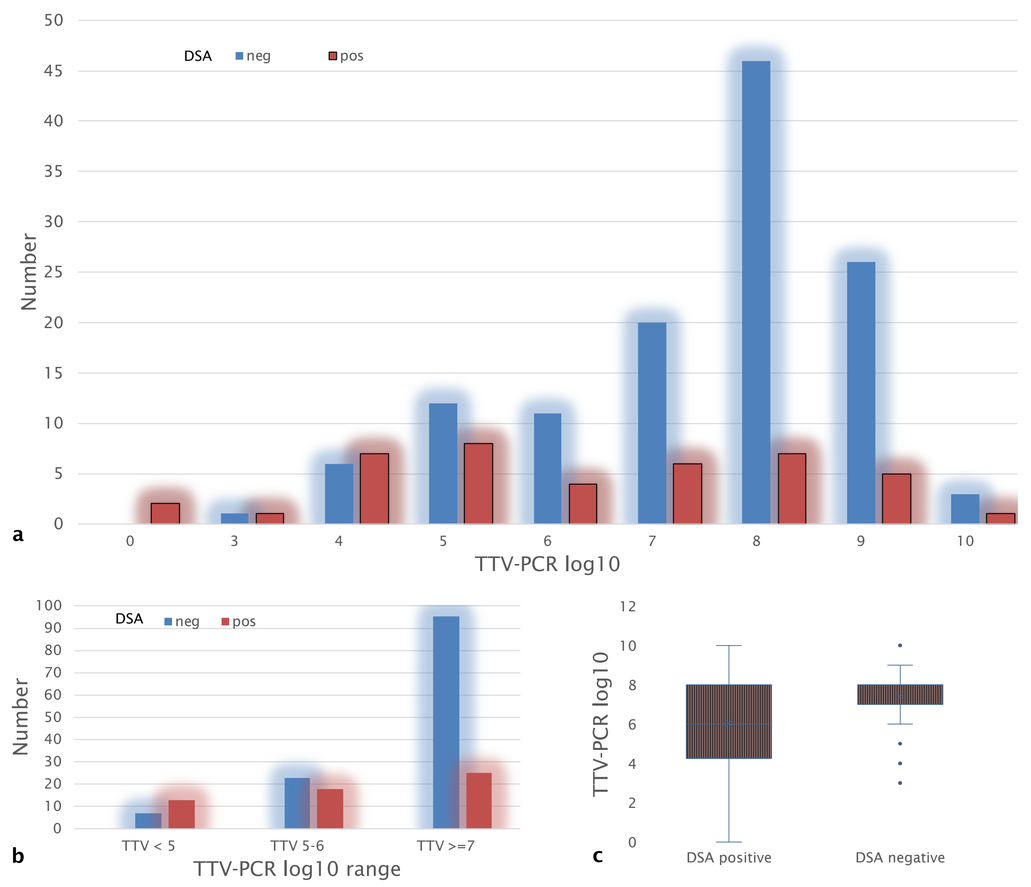

Figure II: **a** Number of DSA positive patients with regard to TTV load. **b** Number of DSA positive patients with regard to TTV load ranges. **c** Boxplot of TTV log10 quartiles with regard to DSA status

Patients who developed donor-specific antibodies had significantly lower TTV values (median log10 of 6 ± 2.3 compared to 8 ± 1.52, p<0.001 *(Figure II)*).
This was also true for those with biopsy-proven humoral or cellular rejection (p=0.002). Multivariate regression uncovered TTV levels as the leading predictor for DSA development. Patients with TTV log10 levels of 5 or less had a threefold increased relative risk for DSA development compared to those with TTV of log10 of 7 or more (Odds Ratio 7.15).

Discussion

We evaluated TTV as a possible surrogate parameter for individualized guidance of immunosuppression over a period of 6.5 years in our pediatric heart center in an observational study. To our knowledge, the study at hand is the first to report the normal course of Torque-Teno viral load in the presence of standardized immunosuppression in a sizable pediatric heart transplant recipient collective.
Our data suggests that TTV log10 levels of 6 and less are associated with a significantly increased risk of DSA development. Thus it might be warranted to individually adjust the standard immunosuppressive regime in case of low TTV despite correct and compliant use of medication. Larger prospective multicenter studies are needed to evaluate the use of TTV to control immunosuppression.