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## **Organ Transplantation in Hereditary Fibrinogen A $\alpha$ -Chain Amyloidosis: A Case Series of French Patients**

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## **Abstract**

**Rationale & Objective:** Fibrinogen A  $\alpha$ -chain amyloidosis (AFib amyloidosis) is a form of amyloidosis resulting from mutations in the fibrinogen A  $\alpha$ -chain gene (*FGA*), causing progressive kidney disease leading to kidney failure. Treatment may include kidney transplantation (KT) or liver-kidney transplantation (LKT), but it is not clear what factors should guide this decision. The aim of this study was to characterize the natural history and long-term outcomes of this disease, with and without organ transplantation, among patients with AFib amyloidosis and various *FGA* variants.

**Study Design:** Case series.

**Setting & participants:** Thirty-two patients with AFib amyloidosis diagnosed by genetic testing in France between 1983 and 2014, with a median follow-up of 93 (range, 4 to 192) months, were included.

**Results:** The median age at diagnosis was 51.5 (range, 12 to 77) years. Clinical presentation consisted of proteinuria (93%), hypertension (83%), and kidney failure (68%). Manifestations of kidney disease appeared, on average, at age 57 (range, 36 to 77) years in patients with the E526V variant, at age 45 (range, 12 to 59 years) in those with the R554L variant ( $p < 0.001$ ), and at age 24.5 (range, 12 to 31) years in those with frameshift variants ( $p < 0.001$ ). KT was performed in 15 patients and LKT was performed in four. In KT patients with E526V or R554L variants, recurrence of Afib-amyloidosis in the kidney graft was less common than with frameshift variants (22% versus 83%,  $p = 0.03$ ) and led to graft loss less frequently (33% versus 100%). Amyloid recurrence was not observed in patients after LKT.

**Limitations:** Analyses were based on clinically available historical data. Small number

of patients with non-E526V and frameshift variants.

**Conclusions:** Our study suggests phenotypic variability in the natural history of AFib amyloidosis, depending on the *FGA* mutation type. KT appears to be a viable option for patients with the most common E526V variant, while LKT may be a preferred option for patients with frameshift variants.

**Key words:** amyloidosis, fibrinogen, end-stage renal disease (ESRD), genotype-phenotype correlation, liver-kidney transplantation, *FGA*, hereditary disease, kidney biopsy, amyloid recurrence, case series

## **Introduction**

Hereditary amyloidosis with decreased kidney function may be associated with mutations in eight genes, including fibrinogen A  $\alpha$ -chain (*FGA*)(1), apolipoprotein A-I (*APOA1*)(2), apolipoprotein A-II (*APOA2*)(3), lysozyme (*LYZ*)(4), gelsolin (*GSN*)(5), transthyretin (*TTR*), apolipoprotein C-II (*APOC2*)(6), and apolipoprotein C-III (*APOC3*)(7). Fibrinogen A  $\alpha$ -chain amyloidosis (AFib amyloidosis) was first described in 1993 and remains the most frequent form in Europe (1). AFib amyloidosis is an autosomal dominant disease resulting from specific mutations in *FGA*, leading to amyloidogenic variants (8). Clinically, AFib amyloidosis patients present with proteinuria or nephrotic syndrome and hypertension around the fifth decade of life and develop kidney failure rapidly after disease onset (8,9). Histologically, Afib-amyloidosis patients show massive amyloid deposition in glomeruli, with little or no vascular or interstitial amyloid deposits (10). Treatment of Afib-amyloidosis is limited to hemodialysis and kidney transplantation, with a variable risk of amyloid recurrence in the kidney allograft within the first few years after transplantation (8).

Since the first Afib-amyloidosis case (1), 17 amyloidogenic *FGA* variants have been reported and the first genotype-phenotype correlations have been made, indicating that the clinical expression, prognosis, and risk of amyloid recurrence in Afib-amyloidosis patients may be determined by mutation type (1,8,11–18). Indeed, the most common variant globally, E526V (a missense variant in which the glutamate at amino acid 526 is replaced by valine), is usually associated with late-onset disease, slow progression towards kidney failure, low penetrance, and usually excellent outcomes after kidney transplantation (KT) (8,11,14,16–19). However, *FGA* frameshift variants are

associated with more aggressive disease, with early-onset, rapid evolution, and fast amyloid recurrence after KT (15). The molecular basis for this variability in the clinical expression and prognosis of Afib-amyloidosis is unclear, but it may stem from the distinct amyloidogenic properties of *FGA* variants(15).

Considering the potential systemic nature of Afib-amyloidosis and amyloid recurrence in the kidney graft, the role of transplantation in the treatment of Afib-amyloidosis is evolving. Combined liver and kidney transplantation (LKT) or even liver transplantation alone, when performed early in the course of the disease, can prevent renal and systemic disease progression (20–22). However, the Afib-amyloidosis patients reported to date in large case-series studies with long-term follow-up have been carriers of the most common variant, E526V (8,20,23). Consequently, the natural history of Afib-amyloidosis associated with variants other than E526V remains poorly understood, precluding the full understanding of genotype-phenotype correlations, which are essential to define a consensus strategy on organ transplantation.

The aims of this study were to describe the clinicopathological presentation and the long-term outcomes of solid organ transplantation, associated with E526V, R554L (a missense variant in which the arginine at amino acid 554 is replaced by a leucine), and two different frameshift *FGA* variants, in a case series of 32 French Afib-amyloidosis patients. These additional studies with long-term follow-up of Afib-amyloidosis patients with various amyloidogenic *FGA* mutations may help to improve our knowledge of phenotype-genotype relationships, in order to optimize organ transplantation strategies.

## **Methods**

### **Study design**

We report on a case series of AFib-amyloidosis patients from several French universities and general hospitals. The study was performed in accordance with the ethical standards of the Declaration of Helsinki and all patients gave informed consent to participate. We used electronic newsletters and email to contact practitioners registered in the Société Francophone de Néphrologie, Dialyse et Transplantation. We next contacted practitioners who cared for the patients for demographic, clinical, and histological data, which were directly collected from the patients' charts by LM and MU.

### **Setting and participants**

Patients aged at least 18 years, with a genetically confirmed Afib-amyloidosis diagnosis made between 1983 and 2014 were included. The patients were followed up until 2017. Demographic, clinical, and histological data were retrospectively obtained from patients' charts and were centrally reviewed. Estimated glomerular filtration rate (eGFR) was calculated using the IDMS-traceable 4-variable Modification of Diet in Renal Disease Study (MDRD) equation. Incident kidney replacement therapy (KRT) was defined as the beginning of dialysis or transplantation.

### **Genetic analysis**

Blood samples from all patients were obtained after their written informed consent and with the local approval of the Ethics Committee of each hospital center. This study was performed according to the principles of the Declaration of Helsinki. Blood samples from patients with non-AL amyloidosis deposits in a biopsy sample and/or a previous family history of genetically confirmed Afib-amyloidosis were analyzed. The entire coding region and flanking splice sites of exon 5 of the *FGA* gene (MIM\_134820) were sequenced as previously described (15). The nomenclature of the *FGA* mutations is

based on the *FGA* transcript reference (NM\_000508.3). According to the recommendations of the Human Genome Variation Society (HGVS), amino acid numbering starts at the initiator methionine. To convert the conventional mature protein amino acid numbering to HGVS nomenclature, 19 residues would need to be added, meaning that in HGVS nomenclature, the missense variants would be denoted p.Glu545Val (E526V) and p.Arg573Leu (R554L); the 2 frameshift variants would be p.Phe540Leufs\*28 (Phe521Leufs; ie, a phenylalanine to leucine frameshift at amino acid 521, leading to a termination codon 28 residues later) and Val541Alafs\*27 (Val522Alafs; ie, a valine to alanine frameshift at amino acid 522, leading to a termination codon 27 residues later). All of these are documented pathogenic variants, including the p.Phe540Leufs\*28 variant, which we previously analyzed by mass spectrometry (15).

### **Statistical methods**

Medians and ranges are used in the description of continuous variables and these data were compared using the Mann Whitney U test. Percentages were used for the description of dummy variables and were compared using Fisher's exact test. Patient survival was censored at the time of loss to follow-up or the end of the study. Graft survival was censored at the time of death, loss to follow-up, or the end of the study. Patient and kidney allograft survival over time were plotted using Kaplan-Meier curves. STATA (version 14; StataCorp, College Station, TX, USA) and R (version 3.2.1, R Foundation for Statistical Computing, Vienna, Austria) were used for the descriptive and survival analyses, respectively.

## **Results**

### **Patient Characteristics**

### *Overall*

A total of 32 patients genetically diagnosed with Afib-amyloidosis between 1983 and 2014, with a male/female ratio of 1/3, were included in this study. In the 23 patients with the *FGA* E526V variant, we identified three families with more than one kindred. In the five R554L carriers, we identified two families with two and three kindreds, respectively. We identified four patients with frameshift variants from two different families (**Table S1**). Clinical characteristics at diagnosis are summarized in **Table 1**. The median duration of follow-up, censored by death, was 96 (range, 12 to 324) months. The median age at diagnosis was 51.5 (range, 12 to 77) years. Kidney involvement was reported in 100% of patients. At diagnosis, 83% of patients presented with hypertension (HTN) associated with glomerular-range proteinuria, leading to nephrotic syndrome in 44% and kidney failure in 68%. The median eGFR was 49.6 (range, 10 to 172.3) mL/min<sup>1.73m<sup>2</sup></sup>, **Table 1 and Table S1**). Twenty-three patients who were carriers of the E526V variant, five patients with the R554L variant, and four patients carrying the frameshift Phe521Leufs and Val522Alafs variants were included in this study. Clinically significant extra-renal biopsy-proven amyloidosis was observed in 16% of patients, even after a median follow-up of 8 years, and involved patients with the E526V genotype.

Notably, cardiovascular disease was reported in 66% of patients. Nine patients presented with myocardial hypertrophy (one of whom had proven cardiac amyloidosis); six patients presented with cardiac valvular disease; two patients underwent aortic valvular replacement, of which one underwent multiple endomyocardial biopsies during the procedure that showed no evidence of cardiac amyloidosis; two patients presented with ischemic cardiopathy; and one patient each presented with non-ischemic dilated

cardiomyopathy of undetermined cause, transient ischemic attack, lower limb arteriopathy, aortic aneurysm. Except for one case of proven cardiac amyloidosis, these cardiovascular manifestations were not documented as being a consequence of amyloidosis and were more likely to be atherosclerotic, considering the age of the patients and their cardiovascular risk factors (chronic kidney disease, n = 32 ; dyslipidemia n = 20 [63%] ; HTN, n = 19 (59%); tobacco use, n = 7 [22%]; and diabetes, n = 2 [6%]). A total of 28 patients underwent systematic carotid Doppler ultrasound, which revealed two non-stenosis atheromatous lesions, one non-significant (less than 70%) carotid stenosis, and one atheromatous plaque responsible for one transient ischemic attack.

#### *E526V carriers*

The median age at diagnosis of the 23 patients with the *FGA* E526V variant was 57 (range, 36 to 77) years. Kidney involvement was reported in 100% of these patients. At diagnosis, 22 (95%) of E526V carriers presented with HTN associated with glomerular-range proteinuria, leading to nephrotic syndrome in 44% and kidney failure in 80%. The median eGFR was 45.5 (range, 10-96) mL/min/1.73 m<sup>2</sup>. A family history of Afib-amyloidosis was reported in 94% of E526V carriers. Five E526V carriers (22%) presented with clinical extra-renal amyloidosis involvement. Of these, one patient developed amyloid cardiomyopathy responsible for stage 3 cardiac failure and arrhythmia. Cardiac amyloidosis was biopsy-proven, with anti-fibrinogen antibodies labeling amyloid deposits in subendocardial and perivascular areas. Peripheral symptomatic axonal neuropathy was detected in two patients (other causes of axonal neuropathies excluded), whereas asymptomatic neuropathy was diagnosed on systematic

electromyograms in two others. Carpal tunnel syndrome was documented in two patients. Of note, no biopsies were performed to confirm amyloidosis.

#### *R554L carriers*

The median age at diagnosis of the 5 patients with the *FGA* R554L variant was 53 (range 45 to 59) years. Kidney involvement was reported in 100% of these patients. At diagnosis, 2 of 4 (50%) patients with data available presented with HTN associated with non-nephrotic range proteinuria, leading to kidney failure in 50%. The median eGFR was 59 (range, 51.7 to 66.3) mL/min/1.73 m<sup>2</sup>. A family history of Afib amyloidosis was reported in 100% of cases with the R554L variant. No clinical extra-renal involvement was documented.

#### *Phe521Leufs and Val522Alafs frameshift carriers*

The median age at diagnosis of the 4 patients with the Phe521Leufs or Val522Alafs frameshift variants was 24.5 (range, 12 to 31) years. Kidney involvement was reported in 100% of these patients. At diagnosis, these patients presented with glomerular-range proteinuria, leading to nephrotic syndrome in 75% of cases, but it was not associated with HTN or kidney failure. The median eGFR was 132.1 (range 91.8 to 172.3) mL/min/1.73 m<sup>2</sup>. A family history of Afib-amyloidosis was reported in 100% of these cases. No clinical extra-renal involvement was documented.

#### *Clinical Characteristics by Variant*

Overall, clinical manifestations appeared later for patients with the E562V variant (n = 23) compared with those with the R554L and frameshift variants (non-E526V group, n = 9 patients). They appeared at 57 (range, 36 to 77) years in the E562V group compared to 45 (range, 12 to 59) years in the R554L group ( $p < 0.001$ ) and 24.5 (range, 12 to 31)

years in the frameshift variant group ( $p < 0.001$ ). Moreover, eGFR at the time of diagnosis was significantly lower in the E526V group (45.5 [range, 10 to 96] mL/min/1.73 m<sup>2</sup>) compared to the non-E526V groups (R554L: 79 [range, 51.7 to 172.3] mL/min/1.73m<sup>2</sup>,  $p = 0.008$ ; frameshift group: 132.1 [range, 91.8 to 172.3] mL/min/1.73 m<sup>2</sup>,  $p = 0.005$ ).

### **Renal histology**

Kidney biopsy was performed in 28 out of 32 patients, but a precise description of the biopsy was only available for 26 of these patients. A kidney biopsy was not performed in four patients, because they were related to a proband with a previous histological confirmation of disease (patients 7, 23, 27, and 28). For two patients, proteomic analysis using mass spectrometry of the amyloidosis deposits confirmed the diagnosis of Afib-amyloidosis, as previously reported for one family (15). Extensive amyloidosis deposits were almost exclusively found in glomeruli (**Fig. 1A, B**). Extra-glomerular amyloid deposits were only documented in 3/26 patients (two with the Phe521Leufs variant and one with the E526V variant) and these consisted of arteriolar deposits. Nonspecific tubulo-interstitial lymphocytic infiltrates were documented in 5/26 patients. No other tubulo-interstitial lesions were identified. Amyloid deposits were characterized by Congo red-positive staining (**Fig. 1B**) and for some patients, immunofluorescence using a primary antibody against fibrinogen showed strong positivity within the glomerulus (**Fig. 1C**). The sensitivity of routine immunohistochemistry techniques for Afib-amyloidosis diagnosis (**Table S2**) was very poor (8% for paraffin-embedded sections and 35% for frozen sections) and therefore, its specificity may also be questioned.

### **Clinical course and renal prognosis**

Afib-amyloidosis led to KRT initiation in 100% of the 32 symptomatic patients included in this study. The E526V group had older age at KRT initiation (49 [range 38 to 77] years) than did those in the non-E526V group (27 [range 12 to 63] years,  $p < 0.01$ ; **Fig. S1 A, B**). Moreover, the median time between diagnosis and KRT initiation was significantly shorter in the E526V group (24 [range 0 to 72] months) than in the non-E526V group (48 [range 12 to 84] months,  $p = 0.03$ ) and in the frameshift variant only group (36 [range 12 to 84] months,  $p = 0.2$ ).

Four patients died, all of whom were carriers of E526V. Of these, one died because of a larynx carcinoma; one died of septic shock; and two died of cardiovascular causes (aortic bioprosthesis thrombosis and cardiac arrest of undetermined cause), with no histological proof of amyloidosis (**Table S3**). The 10 and 30-year patient survival rates since diagnosis were 84% and 70%, respectively.

### **Follow-up after KT or LKT**

#### *Overall*

Eighteen patients underwent transplantation, with 15 receiving KT only and four receiving LKT (including one patient who previously received KT). The median follow-up duration post transplantation was 93 (range 4 to 192) months. For the patients who received only KT, the disease recurrence in the graft was diagnosed in 47% of cases (7/15). Recurrence of the disease was suspected by de novo or increased glomerular-range proteinuria and/or degradation of allograft function. It was then confirmed by a biopsy of the allograft revealing glomerular amyloidosis deposits with Congo red staining. The median time of recurrence was 96 (range 12 to 120) months.

Graft loss occurred in 44% (8/18) of patients. Recurrence of the disease on the

kidney graft was responsible for graft loss in 75% (6/8) of cases. The 5-, 7-, and 9-year kidney graft survival rates were 88%, 80%, and 72%, respectively (**Fig 2A**) and no significant differences were observed between the different variant groups (**Fig 2B**).

#### *E526V group*

The median time of graft loss after KT in patients with the E526V variant was 102 (range 4 to 141) months. Recurrence of the disease on the kidney graft occurred in 22% (2/9) of these patients, with a median time to recurrence of 119.5 (range 119 to 120) months. Graft loss occurred in 33% of patients (3/9; one sudden cardiac arrest, one recurrence, one cellular rejection). Only one patient experienced severe infectious complications (several acute pyelonephritis). Overall, only one patient (1/3, 11%) in the E526V group experienced graft loss due to Afib-amyloidosis recurrence.

#### *R554L group*

The median time of graft loss after KT in patients with the R554L variant was 156 (range 120 to 192) months. Recurrence of disease in the kidney graft occurred in 67% (2/3) of patients, with a median time of recurrence of 105 (range 90 to 120) months. This caused graft loss in both of these 2 patients. No graft rejection or severe infectious complications occurred.

#### *Phe521Leufs and Val522Alafs groups*

The median time of graft loss after KT in patients with frameshift variants was 84 (range 32 to 114) months. Recurrence of the disease on the kidney graft occurred in all three Afib-amyloidosis patients, with a median time of recurrence of 24 (range 12 to 96) months, leading to graft loss in each case. No graft rejection or severe infectious complications occurred.

### *Comparisons*

After KT alone, we observed that disease recurrence in the kidney graft was significantly less frequent in the E526V group than in the frameshift variant group (22% versus 100%,  $p = 0.045$ ). The time to recurrence tended to be longer in patients with the E526V variant when compared to patients with other variants, although this was not statistically significant. Graft loss due to amyloid recurrence in the transplanted kidney was observed in 11% of patients in the E526V group compared to 100% in the non-E526V group (including both R554L and frameshift variant groups,  $p = 0.1$ ) and the frameshift variant only group ( $p = 0.4$ ).

### *LKT*

Three Afib-amyloidosis patients received LKT as their first transplant and another one received LKT after failure of the first KT. As expected, no disease recurrence was observed in the kidney graft after dual transplantation, and consequently, no kidney graft loss occurred. Only one patient died (of larynx carcinoma, 18 months post-transplantation). No other severe complications occurred.

### **Discussion**

The kidney is recognized as the main organ impacted by Afib-amyloidosis and the option of kidney transplantation is inevitably considered during the course of the disease. However, because of amyloid recurrence in the transplanted kidney and the potential risk of systemic progression of the disease, the addition of liver transplantation is considered a curative treatment for Afib-amyloidosis, given that it definitively eradicates the hepatic source of the amyloidogenic *FGA* variant (20–22,24,25). However, this double transplantation procedure has the potential risk of early perioperative death and it has

often been performed only in Afib-amyloidosis patients presenting with significant extra-renal involvement or a more severe disease course (20–22,25). The lack of in-depth knowledge regarding genotype-phenotype relationships and long-term transplantation outcomes according to different *FGA* genotypes explains why transplantation strategy is still a matter of discussion.

To date, only a few Afib-amyloidosis case series have been documented (8,20,23). In the British series, the largest to date, ten patients underwent KT and nine patients underwent LKT. The loss of transplanted kidneys after KT due to amyloid recurrence was observed after a median of 7.3 years, with no recurrence of Afib-amyloidosis in cases of LKT or preemptive liver transplantation (8). In this series, no detailed information was available on the genotype of patients and thus, it was not possible to evaluate the risk of amyloid recurrence by mutation type (8). In addition, the overall survival time was shorter for patients who underwent LKT than for those who underwent KT only, due to a high burden of post-operative complications (up to 33%). This highlights that LKT may not be the best strategy for all patients, although other studies have reported improved outcomes after LKT (20). In the study by Tavares et al., only one E526V patient received KT only and 14 years later, no clinical amyloid recurrence was evident (23).

In our case series of 18 transplanted Afib-amyloidosis patients (15 receiving KT alone), with a long follow-up period of 192 months after transplantation, we observed that the risk of amyloid recurrence on the kidney graft was significantly lower in E526V carriers than in patients with the R554L or frameshift variants. This was comparable to previously reported results (26). However, although the 5-year survival rate (i.e., 85%) of the patients in our study was similar to those in the British study, the overall 10-year

survival rate was higher (71.5% versus 30% for KT and 75% versus 63% for LKT). The recurrence of Afib-amyloidosis in the kidney graft in patients with the E526V variant was rarely responsible for the rate of graft loss, and graft survival after KT only was comparable to the rates in the general French kidney transplant population (88%, 80%, and 72% in our case series at 1, 5, and 9 years post-transplantation, respectively, compared to 91.4%, 79.4%, 62.3% at 1, 5, and 10 years, respectively (27). This comparison is relevant since our patients were recruited from various kidney transplantation centers in France. However, we cannot exclude the possibility that undiagnosed Afib-amyloidosis patients were among the kidney transplant population in those centers. Nevertheless, this may not change the overall survival data of kidney transplant patients, as undiagnosed Afib-amyloidosis patients may only represent a very small percentage of transplant patients. Another limitation of our study is that some patients in our series were siblings. Even if this represents a very small percentage of our total patient population, this familial clustering may have influenced the outcomes, given that they may have shared common environments and modifier genes. Overall, these data support that KT alone may be a valid option for patients with the most common E526 variant.

In addition, as suggested by Rowczenio et al. (28), Afib-amyloidosis patients with R554L and frameshift genotypes are characterized by a more rapid course towards kidney failure than those with the E526V genotype (21,22). Another observation that supports KT rather than LKT is the low incidence of clinically significant extrarenal manifestations. Moreover, these were mostly peripheral neuropathy and carpal syndrome documented in E526V patients, except for one previously published case of cardiac

involvement (29). Of note, 66% of our Afib-amyloidosis patients had cardiovascular manifestations that were not related to Afib-amyloidosis deposits, but were likely a consequence of kidney failure and hypertension as previously reported (8). However, as recently suggested (20) those cardiac manifestations may directly result from amyloid deposition in vascular walls and in atheromatous plaques. In our series, vascular deposits were only documented within the kidney vessels in one patient with the E526V variant, but in all patients with frameshift variants. These renal amyloidosis vascular deposits may cause severe malignant hypertension, leading to acute kidney injury, as we previously reported (15).

In conclusion, our study presents the long-term follow-up of a case series of transplanted Afib-amyloidosis patients, with a spectrum of various *FGA* genotypes. Although the sample size of Afib-amyloidosis patients with non-E526V variants was small in comparison to the number of patients with the most common E526V variant, our findings support the notion that the transplantation strategy should be chosen according to mutation type, as previously suggested (28). These observations suggest that KT alone may be considered for E526V carriers with no clinical evidence of systemic manifestations, whereas LKT may be the best option for patients with frameshift or R554L genotypes, because they may be younger and fitter and those genotypes are associated with a higher risk of premature kidney allograft loss due to amyloid recurrence. These observations also suggest that, in the presence of severe systemic manifestations of biopsy-proven Afib-amyloidosis, LKT may be used regardless of mutation type.

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Authors' Contributions: Research idea and study design: HF, SV, LV, LM, MU; Data acquisition: LM, MU and all other co-authors; Data analysis/interpretation: HF, SV, LV, LM, MU; Statistical analysis: LM, MU, HF; Supervision or mentorship: HF, SV, LV. Each author contributed important intellectual content during manuscript drafting or revision and agrees to be personally accountable for their own contributions and to ensure that questions pertaining to the accuracy or integrity of any portion of the work, even those in which the author was not directly involved, are appropriately investigated and resolved, including with documentation in the literature if appropriate.

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**Table 1.** Clinical characteristics of Afib amyloidosis patients at diagnosis

	Total cases (N = 32)	E526V carriers (n = 23)	Non-E526V carriers (n = 9)		P *	P **
			R554L (n = 5)	Frameshift (n = 4)		
Age, years	51.5 (12-77)	57 (36-77)	53 (45-59)	24.5 (12-31)	<0.001	<0.001
Hypertension	26 (83%)	22 (95%)	2 (50%)****	1 (33%)****	0.02	0.04
Proteinuria, g/d	4.05 (0-12)	4.1 (0-12)	4 (1.5-6)	6 (3-6)	0.2	0.7
Nephrotic syndrome	13 (44%)***	11 (50%)****	3 (75%)****	3 (67%)****	0.6	0.2
eGFR, mL/min/1.73 m <sup>2</sup>	49.6 (10-172.3)	45.5 (10-97.5)	59 (51.7-66.3)	132.1 (91.8-172.3)	0.008	0.005
Serum creatinine, µmol/L	121.5 (60-472)	135.5 (60-472)	92 (83-101)	67 (61-73)	0.08	0.2
Extra-renal amyloidosis	7 (22%)	7 (30%)	0	0	0.1	0.5

Values for categorical variables gives as median (range); for continuous variables as count (percentage).

Conversion factors: serum creatinine in mg/dL to µmol/L, ×88.4.

eGFR: estimated glomerular filtration rate

\*p value comparing E526V carriers and non-E526V carriers

\*\*p value comparing frameshift versus other variants

\*\*\*missing data for three patients

\*\*\*\*missing data for one patient

**Table 2.** Post-transplantation follow-up of Afib amyloidosis patients

	Total cases (N = 32)	E526V carriers (n = 23)	Non-E526V carriers (n = 9)		p*	p**
			R554L (n = 5)	Frameshift (n = 4)		
<b>Transplantation****</b>	18/32 (56%)	11/23 (48%)	3/5 (60%)	4/4 (100%)	0.2	0.1
<b>KT only*</b>	15/18 (83%)	9/11 (82%)	3/3 (100%)	3/4 (75%)	0.049	0.9
Induction therapy						
Basiliximab	7	7	0	0		
Antithymocyte globulins	7	2	3	2		
Immunosuppressive regimen						
Pred/tacro/MMF	10	7	2	1		
Pred/ciclo/MMF	1	1	0	0		
Pred/Tacro/evero	1	1	0	0		
Pred/Tacro/AZA	2	0	1	1		
Recurrence of the disease	7/15 (47%)	2/9 (22%)	2/3 (67%)	3/3 (100%)	0.2	0.045
Time to recurrence, mo***	96 (12-120)	119.5 (119-120)	105 (90-120)	24 (12-96)	0.1	0.1
Graft loss	8/18 (44%)	3/11 (27%)	2/3 (67%)	3/3 (100%)	0.1	0.2
Graft loss due to recurrence	6/8 (75%)	1/3 (33%)	2/2 (100%)	3/3 (100%)	0.9	0.4
<b>LKT*</b>	3/18 (17%)	2/11 (18%)	0	1/4 (25%)	0.04	0.9
Induction therapy						
Antithymocyte globulins	1	0		1		
Basiliximab	2	2		0		
Pred/tacro/MMF IS regimen	3	2		1		
Recurrence of disease	0	0	0	0	0.2	0.9
Graft loss	0	0	0	0	0.1	0.9

KT: kidney transplantation, LKT: liver and kidney transplantation, pred: prednisone, tacro: tacrolimus, ciclo: cyclosporine, MMF: mycophenolate mofetil, evero: everolimus, AZA: azathioprine; IS, immunosuppressive.

\*pvalue comparing E526V carriers and non-E526V carriers, \*\*pvalue comparing E526V carriers and frameshift carriers

\* The denominator is the total number of patients who were transplanted (with kidney alone, or liver-kidney). The numerator includes only those patients receiving LKT as their first transplantation.

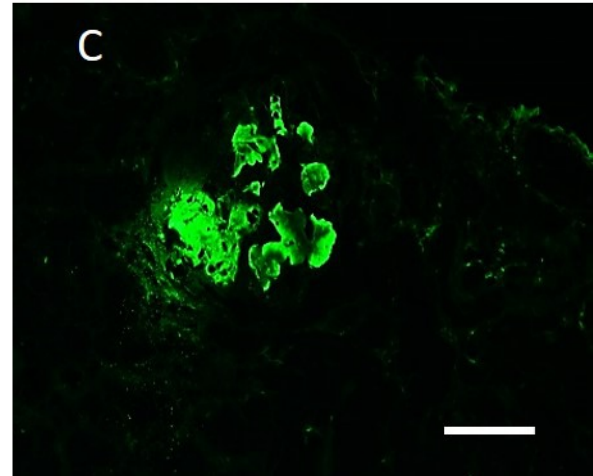
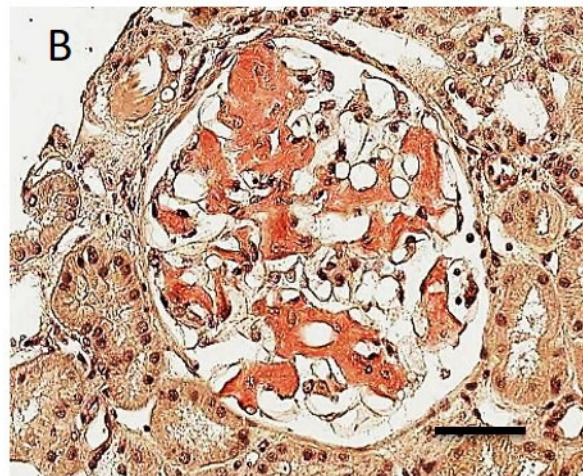
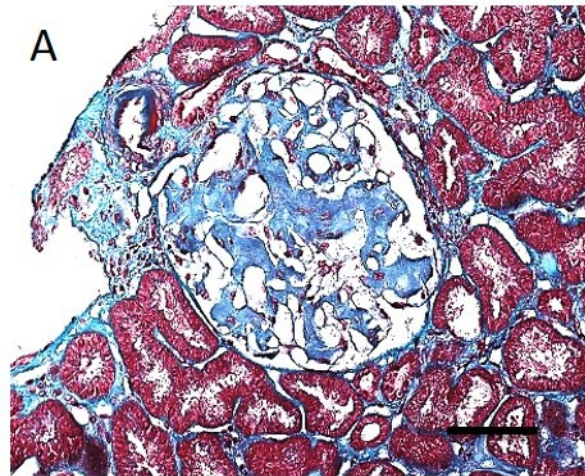
\*\*\*median (range)

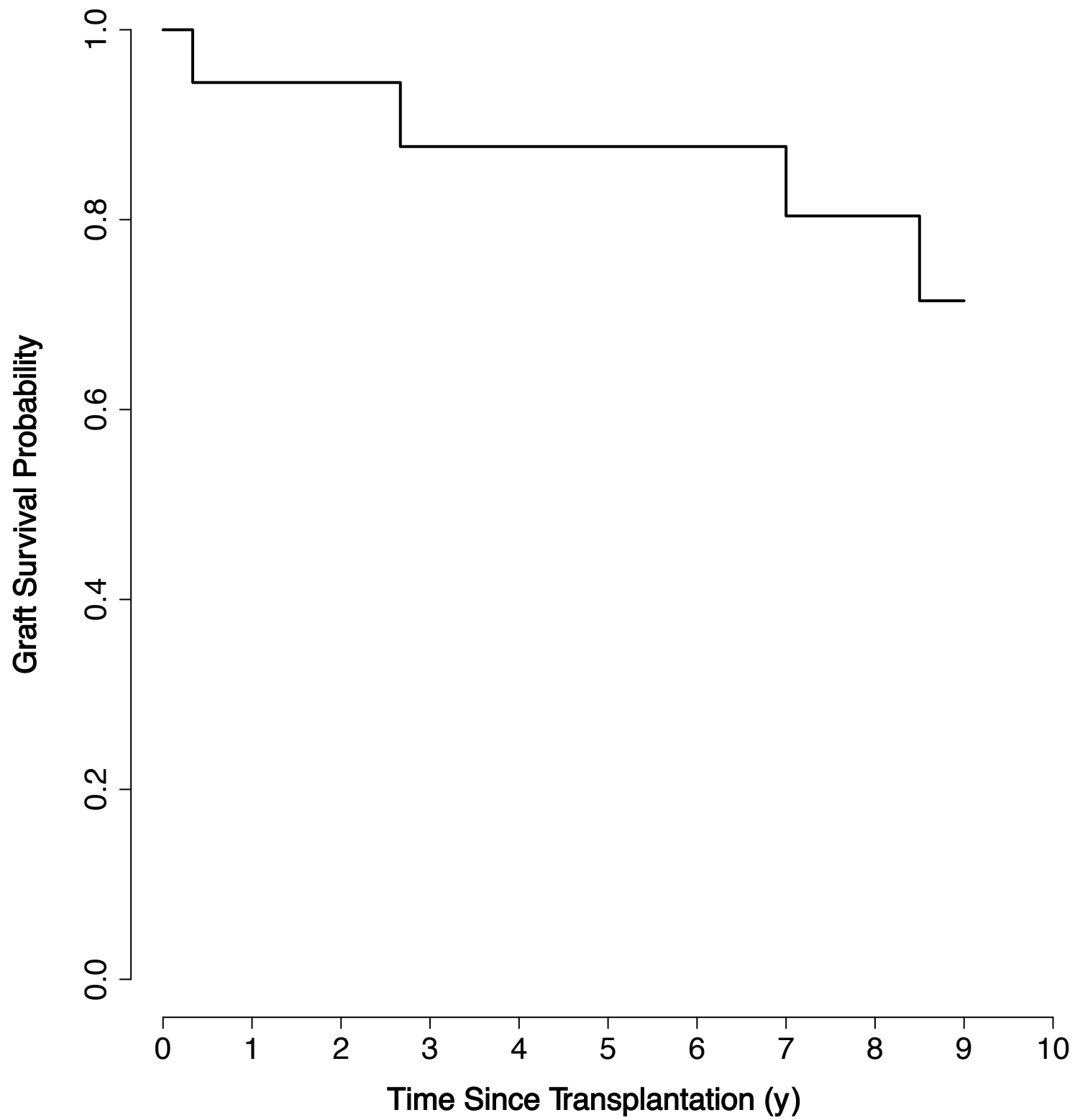
\*\*\*\*KT or LKT

### **Figure Legends**

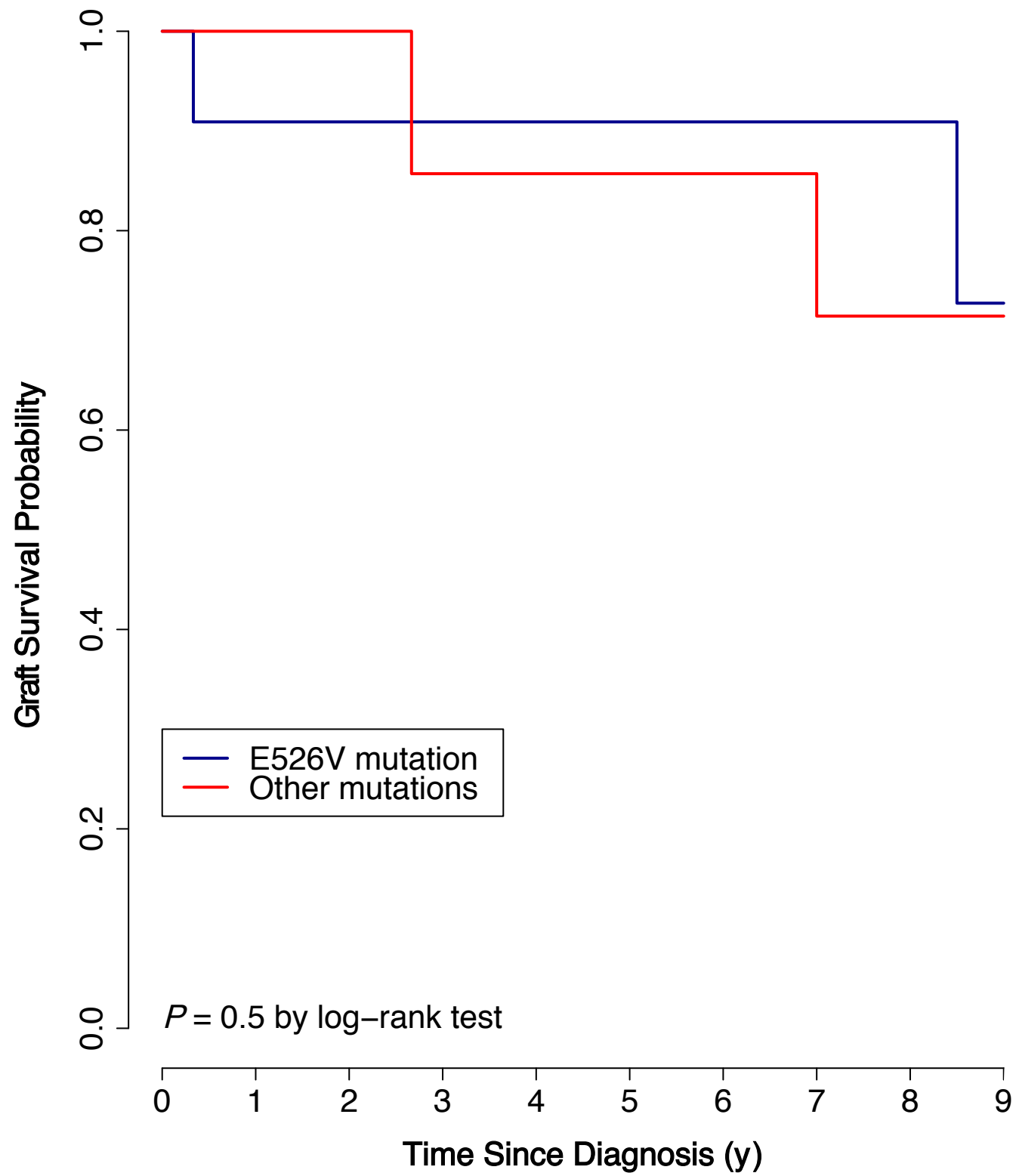
**Fig 1. Renal histology** A. Light microscopy of trichrome staining showing extensive glomerular deposits responsible for mesangial expansion, with no extra glomerular deposit. Scale, 20  $\mu$ m B. Light microscopy of Congo red staining showing Congo red-positive glomerular deposits, with no extra glomerular deposit. Scale, 20  $\mu$ m C. Immunofluorescent staining showing glomerular deposits with positive staining for fibrinogen. Scale, 20  $\mu$ m

**Fig 2. Survival data** A. Kaplan-Meier curve showing graft survival in the whole case series, censored at the time of death. Survival rates were 87.7, 80.4, and 71.5% at 5, 7, and 9 years post transplantation, respectively. B. Kaplan-Meier curve showing graft survival in the “E526V mutation” group and in the “other mutations” group, censored at the time of death. Graft survival rates were 90.9 and 72.7% at 5 and 9 years post transplantation, respectively, in the “E526V mutation” group versus 85.7 and 71.4% in the “other mutations” group, with no significant difference.





No. at  
Risk: 18 16 14 13 13 13 12 12 9 8



**No. at Risk:**

E526V mutation:	11	9	7	7	7	7	6	6	5	4
Other mutations:	7	7	7	6	6	6	6	6	4	4