Outcome in AL amyloidosis presenting with advanced CKD in relation to speed and magnitude of clonal response to chemotherapy

Running Title: Outcomes in advanced renal AL amyloidosis

Tamer Rezk Bsc[1,2], Helen J Lachmann MD[1], Marianna Fontana PhD[1], Sajitha Sachchithanantham MD[1], Shameem Mahmood MD[1], Aviva Petrie [3], Carol J Whelan MD[1], Jennifer H Pinney MD[1], Darren Foard[1], Thirusha Lane PhD[1], Taryn Youngstein Bsc[1], Ashutosh D Wechalekar MD[1], Paul Bass MD[2], Philip N Hawkins PhD[1], Julian D Gillmore PhD[1]

[1]National Amyloidosis Centre, [2]UCL Centre for Nephrology, Division of Medicine, University College London & [3]Eastman Dental Institute, University College London

Correspondence: Dr JD Gillmore, National Amyloidosis Centre, Division of Medicine, University College London, Royal Free Campus, Rowland Hill Street, London NW3 2PF, UK

Tel: +44 (0)20 7433 2726 E-mail: j.gillmore@ucl.ac.uk

Fax: +44 (0)20 74332844

Abstract: 248 words

Text: 3678 words

Abstract

Renal involvement causing progressive chronic kidney disease (CKD) is present in 70% of patients with systemic AL amyloidosis at diagnosis. Chemotherapy that substantially suppresses free light chain (FLC) production is associated with improved patient survival, but its benefit in delaying the onset of renal replacement therapy among patients who present with established advanced CKD has not been studied. Of 1000 patients who were enrolled into the prospective UK AL amyloidosis chemotherapy (ALchemy) study, 84 patients had advanced amyloid-related CKD defined by an estimated glomerular filtration rate (eGFR) <20 ml/min/1.73 m^2. We determined outcomes among these 84 patients in relation to response to chemotherapy evaluated at 3, 6 and 12 months from baseline. Median baseline eGFR was 10 ml/min/1.73 m^2. Patients who achieved a dFLC response of ≥90% within 3 months of baseline had significantly better overall survival (p=0.02), a prolonged time to dialysis (p=0.003), and a prolonged time to the composite endpoint of 'death or dialysis' (p<0.001) compared to those who achieved lesser degrees of clonal response at the same timepoint. A delay beyond 3 months in achieving a ≥90% dFLC response was also associated with worse outcomes. Cox regression analyses showed that achieving a dFLC response of ≥90% within 3 months of baseline was the only independent predictor of all three of these outcome measures (p<0.04). Renal survival among patients with systemic AL amyloidosis who present with advanced CKD is strongly dependent upon the magnitude and speed with which the underlying hematologic disorder is suppressed by chemotherapy.

Keywords: Amyloid, amyloidosis, chemotherapy, chronic kidney disease

Introduction

The amyloidoses are disorders of protein folding, in which a variety of proteins misfold and aggregate into amyloid fibrils that accumulate in tissues and disrupt organ function.[1] Immunoglobulin light chain (AL) amyloidosis is caused by deposition of fibrils derived from monoclonal immunoglobulin light chains and is the most common and serious type of systemic amyloidosis.[2] Renal involvement is present in approximately 70% of patients with systemic AL amyloidosis at diagnosis, manifesting with nephrotic syndrome and progressive renal impairment.[3] Progression to end-stage renal disease (ESRD) is one of the main determinants of morbidity in AL amyloidosis,[4] whilst presence and severity of cardiac amyloidosis is the main determinant of mortality.[5, 6]

Response to chemotherapy has been shown to be strongly and independently associated with both patient survival[3] and renal outcomes in patients with AL amyloidosis.[7, 8] Other factors associated with poor renal outcomes in those with renal AL amyloidosis include a low GFR and heavy proteinuria at diagnosis.[7, 9] However, there are no data on whether chemotherapy can delay onset of renal replacement therapy in patients with AL amyloidosis who present with established advanced CKD and whether speed of clonal response influences renal outcome.

The AL Amyloidosis Chemotherapy study (ALchemy) is a comprehensive prospective observational study opened in 2009 into which all patients newly diagnosed with AL amyloidosis at the UK National Amyloidosis Centre (NAC) are invited to participate. We report here the renal and patient outcomes among all participants in ALchemy who had advanced CKD at the time of diagnosis (and entry to study) between 2009 and 2015 in relation to the speed and depth of the hematologic response to chemotherapy.

Results

Baseline Characteristics and Patient Survival

Baseline demographics and clinical characteristics of all 84 patients are listed in Table 1. The cohort was followed for a median of 16.3 months (range 0.4-68.0) from baseline. Forty five of 84 patients had 'renal isolated' involvement and 39 had evidence of both cardiac and renal involvement (Figure 1). Fifty-seven of 84 patients had renal histology performed and all biopsies showed extensive renal infiltration by amyloid; the only other notable pathology being hypertensive arteriosclerosis in 2 cases. Median age at diagnosis in the whole cohort was 68 years with an almost equal male to female ratio. Median eGFR was 10 ml/min/1.73 m^2 with a median 24 hour urinary protein leak of 6.2 grams. Serum albumin was modestly reduced with a median of 31 g/L despite substantial proteinuria in the majority of cases. Median NT-proBNP was 550 pMol/L with 32 patients having a concentration >1000 pmol/L (i.e., Mayo stage 3b disease).[6]

A total of 47/84 (56%) patients from the whole cohort died with median time from baseline to death by Kaplan Meier analysis of 25.2 months (CI 9.4-41.1). Thirty of 39 patients with cardio-renal syndrome (due to cardiac and renal involvement by amyloid) died and 17 of 45 patients with 'renal isolated' amyloidosis died. Median overall survival was significantly longer (49.2 months [CI 34.5-undefined]) among those with 'renal isolated' amyloidosis compared to those with cardio-renal syndrome (8.4 months [CI 4.7-22.9], $p<0.001$) (Figure 2). Cause of death among those with cardio-renal syndrome was invariably from progressive cardiac amyloidosis, and in those with renal isolated amyloidosis, was from 'progressive amyloidosis' in 9 cases, and from sepsis, cerebrovascular accident, and incarcerated femoral hernia in one case each. In 5 cases the cause of death was unknown.

Chemotherapy

Chemotherapy was planned in all 84 cases, but was actually administered to 78 patients. Reasons for non-administration of chemotherapy were patient death from progressive amyloidosis in 4/6 patients, and dialysis-dependence in 2 patients who did not have significant extra-renal amyloid. Among those who did receive chemotherapy, 43 (55%) received bortezomib-based regimens first line, 22 (28%) received thalidomide-based regimens first line, and 13 (17%) received a first line regimen containing neither bortezomib nor thalidomide. Median (range) number of chemotherapy cycles administered first line was 4 (1-8) for each of bortezomib-based, thalidomide-based and non-bortezomib, non-thalidomide containing regimens. No patient discontinued bortezomib therapy, but 1 patient discontinued thalidomide, and 1 patient discontinued non-bortezomib, non-thalidomide chemotherapy due to toxicity. As-treated analysis of the 78 patients who received chemotherapy showed a dFLC response at 3 months of ≥90% in 15/43 (34%) who received bortezomib compared to 4/22 (18%) who received thalidomide (bortezomib vs thalidomide, p=0.09, Fisher's exact test) and 2/13 (15%) who received neither drug first line (bortezomib vs neither bortezomib nor thalidomide, p=0.12, Fisher's exact test).

Overall survival (OS) in relation to response to chemotherapy

Seventy-four patients were evaluable for dFLC response by consensus criteria (i.e., had a dFLC at baseline of >50mg/L). Of those 74 patients, 15 did not have a dFLC measurement at 3 months, in 11 cases due to prior death, and were therefore excluded from the analysis of survival in relation to hematologic response at this timepoint. Of the 11 patients who died, 6 did so before receiving chemotherapy, 4 died from progressive amyloidosis and one died from chemotherapy-related complications (sepsis). There was no significant difference in overall

survival between 26 evaluable patients who achieved a dFLC of <40mg/L within 3 months of baseline (median 49.2 months [CI 25.0–undefined]) and 33 evaluable patients who achieved lesser degrees of clonal response at the same timepoint (median 37.4 months [CI 11.3–undefined]) (log rank test, p=0.40) (Figure 3a). Using the 'percentage of baseline dFLC' method to calculate hematologic response enabled all patients to be considered 'evaluable' at baseline although, as described above, the 15 patients who did not have a dFLC measurement at 3 months were again excluded from this analysis. Median overall survival among 21 evaluable patients who achieved a dFLC response of ≥90% within 3 months of baseline was undefined compared to 31.8 months (CI 15.7–55.1) among 48 patients who achieved lesser degrees of clonal response at the same timepoint (log rank test, p=0.02) (Figure 3b). There was no significant difference in overall survival between those patients who achieved a <50% dFLC response at 3 months and those who achieved a dFLC response of 50-89% (log rank test, p=0.09) (Figure 3c).

By Cox regression analysis, independent factors associated with death in the whole cohort of 84 patients were elevated NT-proBNP at presentation (HR 2.72 [CI 1.451-5.088], p=0.002) and achieving a dLFC response ≥90% at 3 months (HR 0.36 [CI 0.138-0.935], p=0.036) (Supplementary Table 1). Percentage dFLC response was also highly significant when incorporated as a continuous variable (HR 0.980 [CI 0.968-0.992], p=0.001)

Renal survival in relation to response to chemotherapy

Among 68 patients who were dialysis-independent at baseline and therefore evaluable for analyses of renal survival, there were 46 patients who were evaluable for hematologic response at 3 months according to consensus criteria. Among 22/46 who achieved an absolute dFLC of <40mg/L within 3 months of baseline, median time to dialysis dependence was 9.7 months (CI

3.4–undefined) compared to 5.2 months (CI 1.9-17.1) among 24/46 patients who achieved lesser degrees of clonal response at the same timepoint (log rank test p=0.18) (Figure 4a). Using the 'percentage of baseline dFLC' method to calculate hematologic response, there were 56 patients who were evaluable for hematologic response at 3 months. Median renal survival among 18/56 patients who achieved a dFLC response of ≥90% within 3 months of baseline was 23.0 months (CI 9.7–undefined) compared to 6.1 months (CI 3.4-12.5) among 38/56 patients who achieved lesser degrees of clonal response at the same timepoint (log rank test, p=0.003) (Figure 4b). Renal outcomes were equally poor among those who achieved a delayed ≥90% dFLC response, classified as only after 6 months from baseline (n=5) or only after 12 months from baseline (n=6) (dFLC response ≥90% within 3 months vs 6 months (log rank test, p=0.001) or vs 12 months (log rank test, p<0.003) (Figure 4c).

By Cox regression analysis, the only independent factor associated with a requirement for RRT among the 68 patients who were dialysis independent at baseline was achieving a dFLC response of ≥90% within 3 months (HR 0.24 [CI 0.106-0.547], p=0.001) (Table 2). Percentage dFLC response at 3 months was also significant when incorporated as a continuous variable (HR 0.978 [CI 0.958-0.998], p=0.031). Interestingly, presenting eGFR, presenting NT-proBNP, and proteinuria at presentation did not predict progression to dialysis. Furthermore, stratification of patients by index of chronic damage on renal histology was not predictive of progression to dialysis, although it should be noted that the vast majority of patients had moderate or severe chronic damage on renal biopsy (Table 1).

Forty-five of 84 patients from the whole cohort had renal amyloidosis in the absence of cardiac involvement and were defined as 'renal isolated.' Nine such patients were on RRT at baseline and 2 died before the 3 month evaluation. In light of our previous results, the remaining 34 'evaluable' patients were stratified using the 'percentage of baseline dFLC' method to ≥90% or <90% dFLC response within 3 months of baseline. Median renal survival

among 11 patients who achieved a dFLC response of ≥90% within 3 months of baseline was 23.0 months (CI 7.3–undefined) compared to only 6.2 months (CI 3.0–12.5) among 23 patients who achieved lesser degrees of clonal response at the same timepoint (log rank test, p<0.007) (Figure 5), and 5.7 months (CI 1.9-undefined) among those who achieved a ≥90% dFLC response, but only after 12 months from baseline (log rank test, p<0.03). There was no significant difference in renal survival between those who achieved a dFLC response within 3 months of 50-89% compared to a dFLC response of <50% (log rank test, p=0.83). By Cox regression analysis, the only independent factor associated with a requirement for RRT in the 45 patients with 'renal isolated' amyloidosis was achieving a dFLC response of ≥90% at 3 months (HR 0.62 [CI 0.057-0.655], p=0.008).

Time to composite endpoint of death or dialysis in relation to response to chemotherapy

Sixteen patients, who were dialysis dependent at baseline, were excluded from all analyses of time to the composite endpoint of death or dialysis. Twelve patients did not have an FLC assay measured at 3 months from baseline, in 10 cases due to death. Of the 10 patients who died, 4 died before receiving chemotherapy, 5 died during chemotherapy from progression of their systemic amyloidosis and 1 died from chemotherapy-related complications (sepsis). The remaining 56 patients were stratified according to dFLC response of <90% or ≥90% at 3 months. Among 18 patients who achieved a ≥90% dFLC response, median time to composite endpoint of death or dialysis was 17.3 months (CI 7.3-46.1) compared to 5.3 months (CI 3.4-7.6) among 38 patients who achieved a <90% response (log rank test, p<0.001) (Figure 6). The first event was death in 9 patients and dialysis in 27 patients. There was no significant difference in median time to death or dialysis between those patients who achieved a <50%

dFLC response at 3 months and those who achieved a dFLC response of 50-89% (log rank test, p=0.53).

By Cox regression analysis, independent factors significantly associated with the composite endpoint of death or dialysis among all 68 patients who were dialysis independent at baseline were elevated NT-proBNP at presentation (HR 2.40 [CI 1.293-4.463], p=0.006) and achieving a dFLC response ≥90% at 3 months (HR 0.23 [CI 0.102-0.505], p<0.001) (Supplementary Table 2). Percentage dFLC response was also highly significant when incorporated as a continuous variable (HR 0.981 [CI 0.971-0.992], p=0.001). Neither presenting eGFR, nor proteinuria at presentation were significant predictors of the composite endpoint. Due to the high clonal response rates observed with bortezomib, a multivariable model in which dFLC response at 3 months of <90% or ≥90% was replaced by bortezomib vs no bortezomib was undertaken. The only factor independently associated with the same composite endpoint in this model was serum NT-proBNP concentration at presentation (HR 2.48 [CI 1.350-4.535], p=0.003) (Supplementary Table 2).

Discussion

Response to chemotherapy is known to be one of the main determinants of patient survival in systemic AL amyloidosis.[3, 10] Two thirds of patients with systemic AL amyloidosis have renal involvement at diagnosis and renal outcome, as well as patient survival, is known to be influenced by response to chemotherapy.[7] However, no studies have been performed to specifically investigate whether the magnitude and speed of clonal response to chemotherapy in patients who present with established advanced renal impairment influences time to requirement for RRT. Similarly, the merits of administering chemotherapy, which is invariably associated with substantial short-term morbidity, remain uncertain among AL amyloidosis

patients who present with advanced CKD but do not have clinically significant extra-renal organ involvement by amyloid. Here we show for the first time, that the speed and magnitude of clonal response in patients presenting with a GFR of <20 ml/min/1.73 m^2 due to renal amyloidosis, directly influence the clinically important outcome measures of death, dialysis and the composite endpoint of death or dialysis, with markedly extended renal and patient survival among patients who achieved a clonal response of ≥90% within 3 months of baseline. Furthermore, we show that in patients with an eGFR of <20 ml/min/1.73 m^2 who do not have cardiac amyloidosis, chemotherapy can substantially delay the requirement for RRT. The findings presented here are analogous to the effect of chemotherapy in patients with advanced (Mayo stage 3) cardiac AL amyloidosis, [11] in which the speed and depth of clonal response directly influence patient survival. Until now it has not been clear whether the same degree and speed of clonal response can salvage renal function or if not, delay RRT in those who do not have cardiac involvement and whether patients with isolated renal amyloidosis require chemotherapy with the same degree of urgency as those with cardiac AL amyloidosis.

Importantly, this study does not prove beyond all doubt that aggressive chemotherapy aimed at achieving a rapid and deep clonal response delays dialysis and/or improves survival in this cohort of patients, since there was no prospective randomisation to a placebo arm or 'low intensity' chemotherapy arm. It does not therefore take into account those whose death or requirement for RRT may have been accelerated by chemotherapy. Nonetheless, the evidence for pursuing chemotherapy that is likely to achieve a rapid and deep clonal response in such patients is compelling; among the 47 patients in the whole cohort who died, 26 did so from progressive amyloidosis, including 4 patients who died before receiving chemotherapy; with only one death of the 26 deaths attributable to complications of chemotherapy. Similarly, there was no evidence of acute kidney injury complicating CKD among those in the cohort who received chemotherapy and only 2 patients out of the 78 who received chemotherapy

required discontinuation due to toxicity, one of whom was already receiving RRT at the time of commencement of chemotherapy, and the other of whom received a total of 8 cycles (first line thalidomide switched to bortezomib) to a complete clonal response and remains dialysis independent. Given that it would probably be considered unethical to withhold chemotherapy from patients with advanced renal dysfunction due to AL amyloidosis, particularly in light of the findings reported here, a prospective randomised trial to definitively answer this question will probably never be possible.

Although bortezomib was associated with higher rates of rapid (within 3 months) and deep (≥90%) clonal response compared to non-bortezomib containing regimens, we were unable to demonstrate that administration of bortezomib was an independent predictor of outcome in this cohort. Nonetheless, we would encourage the use of bortezomib first line in patients with advanced renal impairment from AL amyloidosis due to the fact that it is generally well tolerated, no dose modification is necessary in patients with advanced renal impairment, and due to the speed and efficacy with which it can suppress the underlying clonal dyscrasia.

It is noteworthy that use of the established AL amyloidosis consensus criteria for measuring clonal response to chemotherapy, in which patients are required to have an absolute pre-treatment dFLC concentration of >50 mg/L to be evaluable, was associated with categorization of 12% patients as 'not evaluable', in accordance with the 15% patients reported in the consensus document.[12] However, it is interesting that, in this cohort of patients, the consensus criteria did not even predict patient survival, which is well known from larger studies of patients with AL amyloidosis to be associated with depth of hematologic response at 3 months.[13] However, the 'percentage of baseline dFLC' method used in our analyses, which has also been previously validated in AL amyloidosis,[7] predicted both patient and renal survival. Whilst the consensus criteria may be appropriate for determining eligibility of patients with AL amyloidosis for formal clinical trials, our 'real world' data indicates that the

'percentage of baseline dFLC' method is valid and applicable to all patients in a clinical practice setting, and may be superior in patients with established advanced CKD. This study is of insufficient size to recommend a change in the consensus criteria, but given the relative rarity of the disease, the need for a true representation of clinical practice within clinical trials, and the established difficulties associated with enrolling sufficient numbers of patients with AL amyloidosis into most clinical trials, we believe that a specific comparison of these two methods among a large cohort of AL amyloidosis patients with established advanced CKD is warranted and, depending on the findings, may merit considering a change to the consensus criteria.

In summary, chemotherapy should not be withheld from patients with advanced CKD due to renal AL amyloidosis. On the contrary, such patients should be treated urgently with the aim of achieving a rapid and deep clonal response, the result of which may be delayed dialysis and prolonged survival.

Patients and Methods

Patients

At the time of censor, 1000 patients with newly diagnosed AL amyloidosis had been enrolled into the ALchemy prospective observational study at the National Amyloidosis Centre (NAC). Renal involvement, defined as non-Bence Jones proteinuria of more than 0.5g/24 hr according to the amyloidosis international consensus criteria,[14] was present in 672 patients, of whom 84 had presented with advanced renal impairment defined by eGFR <20 ml/min/1.73 m^2. The analyses presented in this manuscript concern this cohort of 84 patients with eGFR <20 ml/min/1.73 m^2 at baseline (Figure 1; Consort Diagram).

All patients underwent protocolized assessments every 3-6 months at the NAC, each assessment comprising clinical evaluation, serum and urine biochemistry including assessment of renal and liver function, N-terminal pro-b-type natriuretic peptide (NT-proBNP), echocardiography, SAP scintigraphy,[15] and assessment of hematological disease by serum free light chain (FLC) assay, serum and urine immunofixation electrophoresis. The presence of cardiac amyloidosis was defined by echocardiography according to international consensus criteria,[14] or in cases in which there was doubt, by additional cardiac magnetic resonance imaging on the basis of native T1 and/or extracellular volume measurement, as previously reported.[16, 17]

All patients were managed in accordance with the Declaration of Helsinki and provided written informed consent for study entry (REC reference 09/H0715/58) and publication of their data.

Renal Histology

Renal biopsies were performed in 57 of 84 patients. All biopsies were routinely stained with Congo red and a panel of amyloid-fibril antibodies, as previously described.[18] Additionally, all biopsies containing sufficient cortical tissue for evaluation (n=49/57) were analysed by a renal histopathologist (PB) and assigned an 'Index of Chronic damage' category of mild, moderate or severe according to the previously described Modified Oxford Score.[19]

Assessment of Hematologic Response

Details and doses of chemotherapy regimens were collected. All patients had serial FLC concentration prospectively monitored on blood samples scheduled monthly during periods of

chemotherapy treatment, and every 1-3 months during subsequent follow up. Healthy polyclonal serum FLC concentrations increase progressively through advancing stages of chronic kidney disease (CKD)[20] which hinders the monitoring of monoclonal light chain disorders. In this study, the value of the FLC monoclonal component was estimated by subtracting the concentration of the uninvolved light chain from that of the amyloidogenic light chain to obtain the FLC difference (dFLC), a strategy previously validated in multiple myeloma and AL amyloidosis.[7, 21]

The FLC response to chemotherapy was determined according to previously validated 'consensus criteria'[12] and additionally, by the percentage of the baseline dFLC that remained at the time of analysis (percentage method), also validated in AL amyloidosis.[7] The consensus criteria define 'evaluable' patients as those with a pre-treatment (baseline) dFLC of >50 mg/L, and thus excluded 10/84 (12%) patients in the cohort, whereas the calculation of the percentage baseline dFLC remaining after chemotherapy can be applied to patients with low level pre-treatment amyloidogenic light chain concentration. A very good partial response (VGPR) was defined according to the consensus criteria as an absolute dFLC of <40mg/L, and by the percentage method as a ≥90% reduction of pre-treatment dFLC remaining after chemotherapy, as previously described.[7] When assessing dFLC response, all patients without an FLC assay at the relevant timepoint were excluded from analysis.

Patient Outcomes

Overall survival was defined as the time from baseline evaluation at the NAC to patient death and was evaluated in all 84 patients. Renal survival was defined as the time from baseline evaluation at the NAC to requirement for renal replacement therapy (RRT). For the analyses of renal survival, patients who were already established on RRT (n=16) at the time of their

baseline evaluation were excluded, and those who died without requiring RRT were censored at the time of death. For analyses of time to the composite endpoint of death or dialysis, patients who were on RRT at baseline were excluded and an event was recorded as the first of either death or dialysis. Patient follow up was censored on 1st October 2015.

Statistical Analysis

Survival analysis was performed separately for each of three possible endpoints: patient survival, renal survival, and survival to composite endpoint of dialysis or death. We determined Kaplan-Meier curves, and performed the log rank test to compare the overall survival curves for different subgroups. Cox proportional hazards regression analysis was used to investigate the factors independently associated with a particular endpoint. A test based on Schoenfeld residuals was used to test the proportional hazards assumption underlying the log rank and the Cox regression analyses. Analyses were performed using GraphPad Prism v5.03, IBM SPSS Statistics 23 and Stata 14 software. A significance level of 0.05 was used for all hypothesis tests.

Disclosures

None

References

1. Palladini G, Campana C, Klersy C, *et al.* Serum N-terminal pro-brain natriuretic peptide is a sensitive marker of myocardial dysfunction in AL amyloidosis. *Circulation* 2003; **107**: 2440-2445.

2. Pepys MB. Amyloidosis. *Annu Rev Med* 2006; **57**: 223-241.

3. Palladini G, Dispenzieri A, Gertz MA, *et al.* New criteria for response to treatment in immunoglobulin light chain amyloidosis based on free light chain measurement and cardiac biomarkers: impact on survival outcomes. *J Clin Oncol* 2012; **30**: 4541-4549.

4. Weiss BM, Wong SW, Comenzo RL. Beyond the plasma cell: emerging therapies for immunoglobulin light chain amyloidosis. *Blood* 2016; **127**: 2275-2280.

5. Kyle RA, Greipp PR, O'Fallon WM. Primary systemic amyloidosis: multivariate analysis for prognostic factors in 168 cases. *Blood* 1986; **68**: 220-224.

6. Wechalekar AD, Schonland SO, Kastritis E, *et al.* A European collaborative study of treatment outcomes in 346 patients with cardiac stage III AL amyloidosis. *Blood* 2013; **121**: 3420-3427.

7. Pinney JH, Lachmann HJ, Bansi L, *et al.* Outcome in Renal AL Amyloidosis following Chemotherapy. *Journal of Clinical Oncology* 2011; **29**: 674-681.

8. Palladini G, Hegenbart U, Milani P, *et al.* A staging system for renal outcome and early markers of renal response to chemotherapy in AL amyloidosis. *Blood* 2014; **124**: 2325-2332.

9. Mahmood S, Palladini G, Sanchorawala V, *et al.* Update on treatment of light chain amyloidosis. *Haematologica* 2014; **99**: 209-221.

10. Lachmann HJ, Gallimore R, Gillmore JD, *et al.* Outcome in systemic AL amyloidosis in relation to changes in concentration of circulating free immunoglobulin light chains following chemotherapy. *Br J Haematol* 2003; **122**: 78-84.

11. Palladini G, Sachchithanantham S, Milani P, *et al.* A European collaborative study of cyclophosphamide, bortezomib, and dexamethasone in upfront treatment of systemic AL amyloidosis. *Blood* 2015; **126**: 612-615.

12. Comenzo RL, Reece D, Palladini G, *et al.* Consensus guidelines for the conduct and reporting of clinical trials in systemic light-chain amyloidosis. *Leukemia* 2012; **26**: 2317-2325.

13. Palladini G, Dispenzieri A, Gertz M, *et al.* Validation of the Critieria of Response to Treatment in AL Amylodiosis. American Society of Haematology2010.

14. Gertz MA, Comenzo R, Falk RH, *et al.* Definition of organ involvement and treatment response in immunoglobulin light chain amyloidosis (AL): A consensus opinion from the 10th International Symposium on Amyloid and Amyloidosis. *Am J Hematol* 2005; **79**: 319-328.

15. Hawkins PN, Lavender JP, Pepys MB. Evaluation of systemic amyloidosis by scintigraphy with ^{123}I-labeled serum amyloid P component. *N Engl J Med* 1990; **323**: 508-513.

16. Karamitsos TD, Piechnik SK, Banypersad SM, *et al.* Noncontrast T1 mapping for the diagnosis of cardiac amyloidosis. *JACC Cardiovasc Imaging* 2013; **6**: 488-497.

17. Fontana M, Chung R, Hawkins PN, *et al.* Cardiovascular magnetic resonance for amyloidosis. *Heart failure reviews* 2015; **20**: 133-144.

18. Tennent GA, Cafferty KD, Pepys MB, *et al.* Congo red overlay immunohistochemistry aids classification of amyloid deposits. In: Kyle RA, Gertz MA,

editors. Amyloid and Amyloidosis 1998. Pearl River, New York: Parthenon Publishing; 1999. p. 160-162.

19. Working Group of the International Ig ANN, the Renal Pathology S, Cattran DC, *et al.* The Oxford classification of IgA nephropathy: rationale, clinicopathological correlations, and classification. *Kidney Int* 2009; **76**: 534-545.

20. Hutchison CA, Harding S, Hewins P, *et al.* Quantitative assessment of serum and urinary polyclonal free light chains in patients with chronic kidney disease. *Clin J Am Soc Nephrol* 2008; **3**: 1684-1690.

21. Dispenzieri A, Zhang L, Katzmann JA, *et al.* Appraisal of immunoglobulin free light chain as a marker of response. *Blood* 2008; **111**: 4908-4915.

Acknowledgements

We thank our many physician colleagues for referring and caring for the patients as well as the staff at the National Amyloidosis Centre. Core support for the Wolfson Drug Discovery Unit, Centre for Amyloidosis and Acute Phase Proteins is provided by NHS England, the UK National Institute for Health Research Biomedical Research Centre and Unit Funding Scheme.

Author Contributions

TR and JDG conceived the manuscript. HJL, MF, SS, SM, CW, JP, DF, TL, TY and ADW were responsible for care and follow up of patients during the study. PB and AP contributed to results and statistical analyses. PNH and JDG reviewed the final manuscript.

Table 1. Baseline demographics of all patients

Demographic or clinical characteristic		**No. of Patients (N =84)**	**%**
Male sex		49	58
Female sex		35	42
Age years	Median	68	
	Range	40-86	
Patients with isolated renal involvement		45	54
Patients presenting on RRT		16	19
Index of Chronic Damage on renal histology	Mild	4/49	8
	Moderate	11/49	22
	Severe	34/49	69
eGFR (ml/min/1.73 m^2)	Median	10	
	Range	10-19	
Amyloid load by SAP scintigraphy	Small	27	32
	Moderate	19	23
	Large	38	45
Serum albumin (g/L)	Median	31	
	Range	14-48	
24hr urinary protein loss (g)	Median	6.2	
	Range	0.1-29.7	
NT-proBNP (pmol/L)	Median	550	
	Range	15-8270	
Troponin T (ng/L)	Median	120	
	Range	10-1870	
Amyloidogenic light chain (n)	Lambda	55	65
	Kappa	29	35
Haemoglobin (g/dL)	Median	11.3	
	Range	7.8-17	
Serum creatinine (µmol/L)	Median	375	
	Range	228-979	
Bilirubin (µmol/L)	Median	5	
	Range	1-59	
Alkaline Phosphatase (u/L)	Median	108	
	Range	43-1703	
Supine systolic blood pressure	Median	137	
	Range	79-184	
Standing systolic blood pressure	Median	129	
	Range	63-180	
Bone Marrow Plasmacytosis (%)	Median	7	
	Range	0-30	
Bence Jones Protein (n)	Present	44	
	Absent	40	
λ sFLC in AL (lambda) patients (mg/L)	Median	241	
	Range	14-5820	
K sFLC in AL (kappa) patients (mg/L)	Median	431	
	Range	56-10300	

Table 2. Independent risk factors associated with dialysis

Variables	**Estimated Hazard Ratio**	**95% Confidence Interval**	**p value**
dFLC ≥90% at 3 months	**0.24**	0.106 - 0.547	**0.001**
Log NT-proBNP	1.35	0.773 – 2.369	0.289
eGFR	0.97	0.878 – 1.068	0.564
Proteinuria	1.01	0.961 – 1.054	0.618

Figure Legends

Figure 1. Consort Diagram showing selection of patients from the prospective UK AL chemotherapy study (ALchemy) for analyses. Patients in shaded boxes were excluded from analyses of renal survival.

Figure 2. Patient survival calculated by Kaplan Meier analysis in all evaluable patients with renal AL amyloidosis and eGFR <20 ml/min/1.73 m^2 at presentation. Survival among those with 'renal isolated' amyloidosis was significantly longer (median 49.2 months) than in those with both cardiac and renal (cardio-renal) involvement (median 8.4 months) ($p<0.001$). Number at risk at certain timepoints is shown in panel below graph.

Figure 3. Patient survival calculated by Kaplan Meier analysis in all evaluable patients with renal AL amyloidosis and eGFR <20 ml/min/1.73 m^2 at presentation. A) Patients were stratified according to degree of clonal response at 3 months into absolute dFLC <40 mg/L and ≥40 mg/L ($p=0.40$). B) Patients were stratified according to degree of clonal response at 3 months into dFLC response ≥90% and dFLC response <90% ($p=0.02$). C) Patients were stratified according to degree of clonal response at 3 months into dFLC response of <50% and 50-89% ($p=0.09$). Number at risk at certain timepoints is shown in panel below graph.

Figure 4. Renal survival calculated by Kaplan Meier analysis in all evaluable patients with renal AL amyloidosis and eGFR <20 ml/min/1.73 m^2 at presentation. A) Patients were stratified according to degree of clonal response at 3 months into absolute dFLC <40 mg/L and ≥40 mg/L ($p=0.18$). B) Patients were stratified according to degree of clonal response at 3 months into dFLC response ≥90% and dFLC response <90% ($p=0.003$). C) Patients were stratified according to speed of clonal response comparing those who achieved a dFLC ≥90%

within 3 months of baseline with those who achieved an equally good dFLC response, but only after 12 months ($p<0.003$). Number at risk at certain timepoints is shown in panel below graph.

Figure 5. Renal survival calculated by Kaplan Meir analysis in evaluable patients with 'renal isolated' AL amyloidosis and eGFR <20ml/min/1.73 m^2 at presentation. Patients were stratified according to degree of clonal response at 3 months into dFLC response ≥90% and dFLC response <90% ($p<0.007$). Number at risk at certain timepoints is shown in panel below graph.

Figure 6. Time to composite endpoint of death or dialysis calculated by Kaplan Meier analysis in all evaluable patients with renal AL amyloidosis and eGFR <20 ml/min/1.73 m^2 at presentation, stratified according to degree of clonal response at 3 months into dFLC response ≥90% and dFLC response <90% ($p<0.001$). Number at risk at certain timepoints is shown in panel below graph.