ORIGINAL ARTICLE

History of extracardiac/cardiac events in cardiac amyloidosis: prevalence and time from initial onset to diagnosis

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Abstract

Aims Cardiac amyloidosis (CA) has a poor prognosis which is aggravated by diagnostic delay. Amyloidosis extracardiac and cardiac events (AECE and ACE) may help improve CA diagnosis and typing. The aim of this study was to compare AECE and ACE between different CA types and assess their relationship with survival.
Methods and results This retrospective cohort study conducted in France from June 2008 to May 2019, at the Henry Mondor Hospital. This cohort included 983 patients with CA. Mean age at inclusion was 73.1 ± 11.4 years, 726 (75.1%) were male and the mean body mass index was 24.5 ± 4.1 kg/m². Among them, 321 had immunoglobulin light chain (AL) amyloidosis, 434 had wild-type transthyretin (ATTRwt), and 212 had hereditary transthyretin (ATTRv). The first AECE and/or ACE occurred at a mean age of 63 ± 11 years for AL and ATTRv, and 70 ± 12 years for ATTRwt ($P < 0.01$). The median (Q1–Q3) delay between declaration of the first events and diagnosis varied from 11.1 (5.9; 34.8) months for AL to 92.2 (39.0; 174.7) months for ATTRwt ($P < 0.01$). The nature of the onset of AECE or ACE varied based on amyloidosis type, heart failure symptoms for AL (26%) and integumentary symptoms for ATTRv with cardiologic or mixed phenotype (39%) and ATTRwt (42%). In AL and ATTRwt, a short delay between the onset of the first AECE or ACE and diagnosis was associated with reduced survival rate (log-rank test P-value <0.01).
Conclusions This study highlights the impact of amyloidosis type and evolution on diagnostic delay and on prognosis. Physicians must be aware and vigilant in front of extracardiac and cardiac events to considerably improve early diagnosis of amyloidosis.

Introduction

Amyloidosis is a systemic disease caused by misfolded proteins that form amyloid fibrils that deposit in organs.[1,2] Prognosis is based on cardiac involvement defined by consensus guidelines.[3–5] There are three main types of cardiac amyloidosis (CA): immunoglobulin light chain (AL) amyloidosis, due to amyloidogenic monoclonal light chain production by a plasma cell clone; hereditary transthyretin (TTR) amyloidosis (ATTRv), caused by the deposition of mutated TTR; and wild-type (non-hereditary) TTR amyloidosis (ATTRwt).[1,6–8]

Extracellular deposition of amyloid in tissues affects their function indirectly due to altered anatomic structure and stiffness, and through direct cytotoxic effects. The potential for amyloid deposits to affect almost any organ system means that the clinical features of amyloidosis and their time of occurrence are diverse, explaining the heterogeneity of amyloidosis types[9] and delays in diagnosis.[10] AL diagnosis criteria includes heart failure associated with neuropathy, nephrotic syndrome, hepatomegaly, periorbital bleeding, and macroglossia.[1,2] Cardiac involvement in transthyretin amyloidosis (ATTR) is often preceded by tegumental infiltration and/or neuropathy.[9] Due to the complexity and rarity of amyloidosis, most studies focus on describing infiltration of a specific organ. Thus, it is difficult to obtain a global picture of the natural history of the disease.[9]

An increased awareness and knowledge of the natural history and clinical signs of amyloidosis type is necessary to improve early diagnosis and outcomes.[11,12]

The aim of this study was to describe the association of the onset of amyloidosis cardiac and/or extracardiac events (ACE and AECE, respectively) associated with the three main types of CA, with time frame to diagnosis and survival outcome.

Methods

Study population

This retrospective cohort study was conducted in France from June 2008 to May 2019 at the French Referral Center for cardiac Amyloidosis at the hospital Henri Mondor (Créteil, France). All subjects referred to the center with an AL, ATTRv with cardiomyopathy or mixed cardio-neuropathic profile and ATTRwt diagnostic were included (*Supporting information*, *Figure S1*). Diagnostic criteria of amyloidosis used were previously described.[13]

Data collection

During the consultation, physicians collected and dated AECE and ACE using a checklist. AECE and AEC rallied different types of events such as the appearance of symptoms, surgeries, various medical analyses, and so forth. All related to amyloidosis (*Table 1*). These events were grouped into nine classes of AECE and eight classes of ACE to performed further analyses (*Table 1*). Baseline characteristics of patients were also collected, including clinical and cardiovascular characteristics, history, risk factor, biology, and echocardiography characteristics (*Table S2*).

Statistical analyses

Analyses were performed using Statistical Analysis Software version 9.4 (SAS Institute, Inc.). Continuous variables are presented as mean ± standard deviation (SD), or median (interquartile range) if highly skewed, and categorical variables were summarized as counts (percentages). The ANOVA test, Wilcoxon rank-sum test, or KruskalWallis test (depending on whether data were normally distributed or not) were used to compare continuous variables, and the χ^2 or Fisher's exact test was used to compare categorical variables. *P* values less than 0.05 were considered significant.

Chord diagrams using the Circlize package V0.4.8 in R were initially used to describe links between the first AECE and/or ACE and amyloidosis type and were subsequently used to illustrate the link between delay of CA diagnosis (<2 and ≥ 2 years for AL and <2 years, 2–5 years, and ≥ 5 years for ATTR diagnosis following particularities of disease development[13]) and the initial onset of an amyloid disorder depending on the type of amyloidosis.

For the three types of amyloidosis, Kaplan–Meier survival analyses using log-rank test was performed to compare the survival of population subgroups for the following outcomes: heart transplantation or death depending on diagnostic delay.

The study was approved by the Henri Mondor institutional ethics committee (authorization number #1431858), and informed consent for participation in this research was obtained for all patients. Data were recorded electronically in the Henri Mondor Amyloidosis Network registry according to the authorization given by the French CNIL (*Commission National de l'Informatique et des Libertés*).

Results

Study population

A population of 967 patients with a diagnostic of CA were included, composed by 321 AL, 212 ATTRv with cardiomyopathy with or without neuropathy, and 434 ATTRwt (*Table S1*). The mean age at the time of recruitment was 72 ± 12 years. The youngest patients presented AL, and the oldest patients

Table 1 Details of events associated with amyloidosis collected by physicians during consultation and classes used for analyses

Amyloidosis extracardiac events (AECEs)			Amyloidosis cardiac events (ACEs)		
Class	Event	Date	Class	Event	Date
ENT (ears-nose-throat)	☐ Deafness (hearing aid + deafness)	.../.../...	Angina	☐ Coronary symptom	.../.../...
	☐ Voice disorder	.../.../...		☐ Chest pain	.../.../...
	☐ Hand nails abnormality	.../.../...		☐ High troponin leading to coronary exploration (CT scan or coronarography)	.../.../...
Integumentary symptoms	☐ Symptom or surgery of the carpal tunnel	.../.../...	Heart failure	☐ Dyspnea	.../.../...
	☐ Narrow lumbar canal	.../.../...		☐ Edema of the lower limbs	.../.../...
	☐ Dupuytren disease (first symptom + surgery)	.../.../...		☐ Cardiac decompensation	.../.../...
Symptomatic digestive or vascular dysautonomia	☐ Digestive dysautonomia	.../.../...	Rhythm disorders	☐ Atrial arrhythmia	.../.../...
	☐ Vascular dysautonomia	.../.../...		☐ Ablation (NAV, flutter, or atrial fibrillation)	.../.../...
	☐ Orthostatic hypotension	.../.../...		☐ Ventricular arrhythmia	.../.../...
Embolic accidents	☐ Stroke/TIA	.../.../...		☐ Cardioversion (medicated or electric)	.../.../...
	☐ Pulmonary embolism	.../.../...		☐ Rhythmic atrium disease	.../.../...
Skin and mucosal symptoms	☐ Periorbital bruise	.../.../...		☐ Other rhythm disorder (atrial or junctional tachycardia)	.../.../...
	☐ Macroglossia	.../.../...	Conduction disorders	☐ Pacemaker implant	.../.../...
	☐ Purpura	.../.../...		☐ Conductive disorders	.../.../...
	☐ Haemorrhage	.../.../...		☐ Syncope/Vertigo	.../.../...
Renal failure or nephrotic syndrome	☐ Renal failure or nephrotic syndrome	.../.../...		☐ Atrio ventricular block (1,2 or 3)	.../.../...
Neuropathy	☐ Neuropathic pain	.../.../...		☐ Sino atrial block 3	.../.../...
	☐ Sensory loss	.../.../...		☐ Chronotropic insufficiency	.../.../...
	☐ Motor loss	.../.../...		☐ Branch block	.../.../...
Others	☐ Lymphadenopathy	.../.../...		☐ Sinus bradycardia	.../.../...
	☐ Gammopathy	.../.../...		☐ Sinus dysfunction	.../.../...
	☐ Haemoptysis	.../.../...	Cardiac enlargement	☐ Cardiac enlargement	.../.../...
			Valvular involvement	☐ Aortic stenosis	.../.../...
				☐ Mitral or tricuspid regurgitation	.../.../...
			Cardiac Biomarkers increased or cardiac imaging abnormalities	☐ Cardiac scintigraphy	.../.../...
				☐ Cardiac MRI	.../.../...
				☐ Increased in NTproBNP	.../.../...
			Pericardium damage	☐ Pericardial effusion	.../.../...

presented ATTRwt. Majority was male (75.1%), and 54.4% had New York Heart Association Classes I or II. Among patients with ATTRv, 115 carried the ATTR V122I mutation and 35 the ATTR V30M mutation (*Table S2*).

Occurrence of first amyloidosis extracardiac and cardiac events

Amyloidosis extracardiac events were more common as first symptoms compared with ACE or their combination, independent of the underlying amyloidosis disorder (*Figure 1*). The rate of initial AECE as first history ranged from 46% in AL to 63% in patients with ATTRv with cardiopathy with or without neuropathy. Meanwhile, the rate of initial ACE was 40% for AL and 36% for ATTRwt. As depicted on the chord diagram (*Figure 2A* and *Table S3*), integumentary symptoms appeared to be more frequently associated with ATTRwt, poor general condition with AL, and ATTRv with cardiomyopathy, with or without neuropathy ($P < 0.01$), possibly due to

V30M mutation. Details of AECE and ACE frequency, whatever the time of onset, are described on *Table S4*.

Figure 2B,C depicts the initial AECE and ACE separately based on the underlying amyloidosis disorder. Integumentary symptoms and ears-nose-throat (ENT) symptoms were frequently observed in ATTRwt, while poor general condition, symptomatic digestive or vascular dysautonomia, renal failure or nephrotic syndrome, and skin and mucosal symptoms were frequently observed in AL. ATTRv with cardiomyopathy with or without neuropathy group presented a high proportion of integumentary symptoms and neuropathy. Heart failure was frequently observed in AL. Rhythm, conduction disorders, and cardiac enlargement were common in ATTRwt (details of initial AECE and ACE frequency on *Table S5*). The nature of the initial AECE or ACE depended on the type of amyloidosis. For AL patients, the most frequent initial presentation was heart failure (26%); meanwhile, for ATTRv with cardiomyopathy or mixed and ATTRwt, integumentary symptoms was the first sign history in 39% and 42% of cases, respectively (*Table S3*).

Figure 1 Representation of number of patients with initial amyloidosis extracardiac (light grey), cardiac (black), or both (dark grey) events depending on the type of amyloidosis.
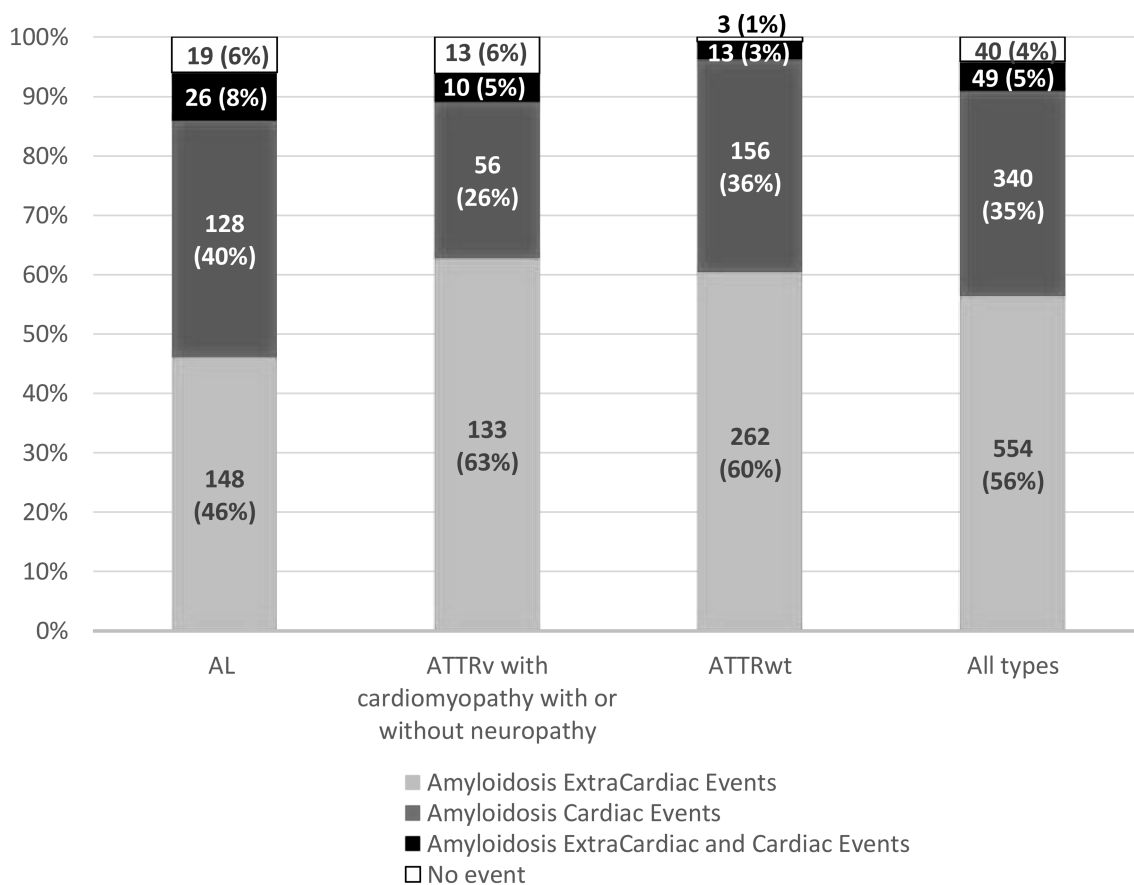
Figure 2 Chord diagrams: (A) chord diagram depicting the link and proportion between the initial amyloidosis extracardiac (AECE) (light grey), cardiac events (ACE) (black), or both (dark grey) and type of amyloidosis (AL in green, ATTRv with cardiomyopathy in orange, and ATTRwt in blue). Regarding AECE and ACE, there is a greater proportion of integumentary and ears-nose-throat symptoms in ATTRwt, neuropathy in ATTRv regardless of neural status, and heart failure and general poor condition in AL. (B) Depiction of the link and proportion between the type of amyloidosis and the initial amyloidosis extracardiac events. Regarding AECE, there is a greater proportion of integumentary and ENT symptoms in ATTRwt, neuropathy in ATTRv, poor general condition, skin and mucosal symptoms, and renal failure in AL. (C). Depiction of the link and proportion between the different types of amyloidosis and the initial ACE. Regarding cardiac events, there is a greater proportion of rhythm disorders in ATTRwt, biomarkers or imaging abnormalities in ATTRv, heart failure in AL.
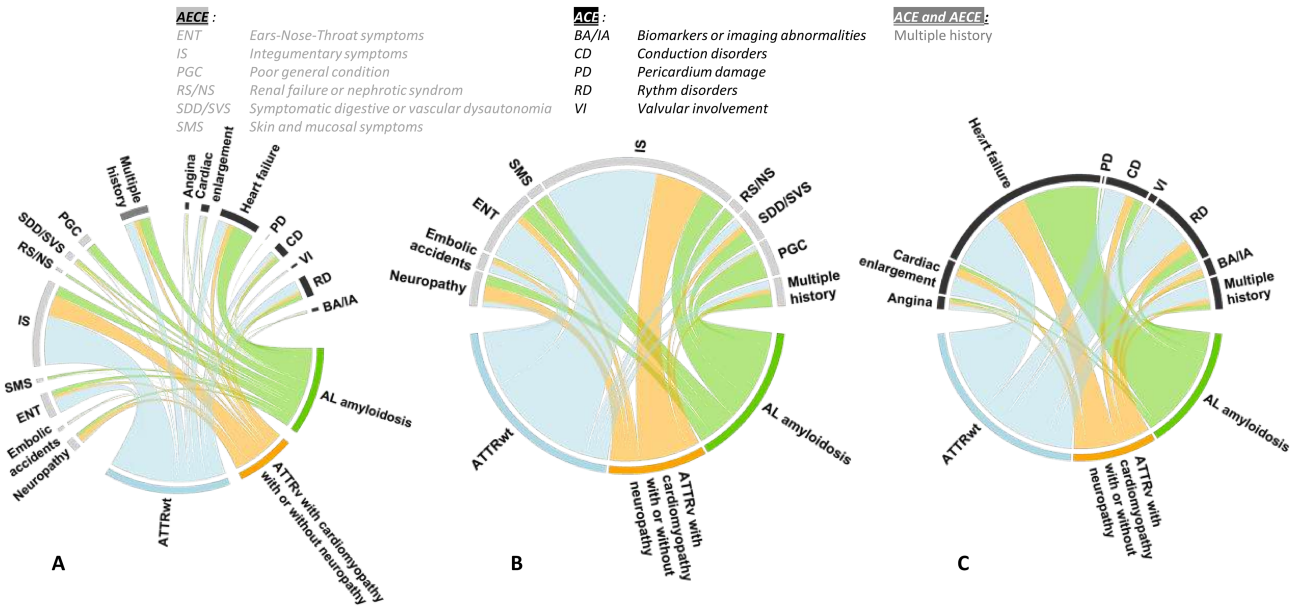
Delay between amyloidosis extracardiac/cardiac events onset and amyloidosis diagnosis

Patients with AECE were slightly younger than patients with ACE when first symptoms occurred, regardless of the type of amyloidosis: 63.6 vs. 64.2 years of age for AL and 68.7 vs. 76.2 for ATTRwt ($P < 0.01$). After the onset of the first symptoms, patients with AECE are diagnosed later than patients with ACE, particularly for ATTRwt [109 (47; 193) vs. 27 (8; 61) months ($P < 0.01$)]. The age of onset of the first AECE or ACE ranged from 62.7 years for the AL and ATTRv with cardiomyopathy or mixed to 70 years for the ATTRwt (*Table 2*).

As described in *Table 2*, the median time between the onset of integumentary symptoms and diagnosis of amyloidosis was the longest, regardless of the type of amyloidosis [121 (65–194) months], followed by ENT [61 (20–137) months], particularly in ATTRwt [140 (86–237) months]. The shortest delay between symptom onset and diagnosis was for cardiac biomarkers or imaging abnormality disorders, which often led to the diagnosis of CA and referral.

The chord diagram in *Figure 3* shows the association between diagnostic delay (in class) and AECE or ACE onset depending on amyloidosis types. Majority of AL was diagnosed before 2 years after AECE/ACE onset, especially if the initial event were cardiac. For ATTRv and ATTRwt, a high proportion

of patients had a diagnostic delay above than 5 years, particularly in case of first extracardiac event such as integumentary symptoms. However, when initial event was cardiac, diagnosis was generally established in less than 2 years as in case of AL.

Survival rate

As presented in *Figure 4*, survival rate changed depending on diagnostic delay. In patients with AL, a shorter delay between the appearance of the initial symptoms and diagnosis (<2 years) was associated with a shorter survival mean [22.7 months (10.5; 47.6) vs. 64.2 (10.2; 120.5) (log–rank $P = 0.034$)]. Diagnostic delay had no impact on the survival rate for patients with ATTRv with cardiomyopathy with or without neuropathy. Finally, in patients with ATTRwt, a longer diagnostic delay (>5 years) was associated with a longer survival rate [52.4 months (42; 84.1) vs. 24.18 months (18.8; 45.7) < 2 years (log-rank $P < 0.001$)]. Delays between diagnosis and death, or cardiac transplantation were described on *Table S6*. Survival rate changed also depending on the class of first symptoms (*Figure 5*). Heart failure is associated with a shorter survival mean (4.2 years vs. ≥20 years, log-rank $P < 0.001$) (*Figure 5*).

Table 2 Median (Q1; Q3) time (in month) between initial onset of extra-cardiac or cardiac event and diagnosis, depending on type of amyloidosis

	Overall N = 967	AL N = 321	ATTRv with cardiomyopathy or mixed N = 212	ATTRwt N = 434	P value
Age at diagnosis of amyloidosis, years, *mean ± SD*[a]	73.1 ± 11.4	65.2 ± 11.4	68.6 ± 10.8	80.1 ± 7	<0.01
Age at the time of the first AECE, years[a]	65.6 ± 11.7	63.6 ± 11.5	61.8 ± 10.6	68.7 ± 11.5	<0.01
Time from first AECE to diagnosis (in months), Median (Q1–Q3)	55.2 (9.4–131.5)	9 (5.9–29)	61 (26–125)	109 (47–193)	<0.01
Classes of time between the first AECE and the diagnosis of amyloidosis, *n*(%)					
<2 years	281 (37)	176 (71)	38 (22)	67 (19)	<0.01
Between 2 and 5 years	120 (16)	34 (14)	45 (27)	41 (12)	<0.01
>5 years	364 (48)	36 (15)	86 (51)	242 (69)	<0.01
Time between first AECE and diagnosis of amyloidosis (in months)					
Integumentary symptoms	121 (65–194)	46 (20–113)	94 (51–140)	140 (86–237)	<0.01
ENT symptoms	61 (17–137)	45 (8.0–129)	56 (36–135)	71 (18–155)	<0.01
Embolic accidents	50 (13–75)	27 (2.1–29)	58 (34–75)	60 (13–84)	<0.01
Neuropathy	20 (8.3–50)	9.4 (3.7–20)	35 (12–61)	26 (15–58)	<0.01
Multiple histories	16 (8.9–53)	11 (2.6–16)	28 (18–44)	69 (15–108)	<0.01
Skin and mucosal symptoms	8.6 (1.9–28)	8.2 (1.5–18)	24 (4.1–43)	245 (245–245)	<0.01
Symptomatic digestive or vascular dysautonomia	8.1 (3.7–31)	7.0 (3.7–12)	57 (22–171)	4.7 (3.3–8.8)	<0.01
Others[a]	6.1 (6.1–6.1)	6.1 (6.1–6.1)	N/A	N/A	
Poor general condition	6 (5.9–7.9)	5.9 (5.9–7.9)	5.9 (5.9–5.9)	5.9 (5.9–8.0)	<0.01
Renal failure or nephrotic syndrome	2.7 (0.4–12)	2.4 (0.4–10.0)	N/A	30 (30–30)	<0.01
Age at time of first ACE, years[a]	70.8 ± 11.7	64.2 ± 11.5	67.9 ± 9.4	76.2 ± 8.5	<0.01
Time between first cardiac history and diagnosis of amyloidosis (in months), median (Q1–Q3)	14.7 (4.7–39.9)	7.0 (3.0–14.9)	15.8 (3.9–37.2)	27 (8.4–61)	<0.01
Classes of time between first ACE and diagnosis of amyloidosis, *n*(%)					
<2 years	527 (60)	221 (82)	117 (64)	189 (45)	<0.01
Between 2 and 5 years	212 (24)	41 (15)	44 (24)	127 (30)	
>5 years	137 (16)	7 (2.6)	22 (12)	108 (25)	
Time between first ACE and diagnosis of amyloidosis (in month)					
Unspecified	60 (18–102)	N/A	18 (18–18)	102 (102–102)	<0.01
Conduction disorders	36 (9.4–68)	11 (7.0–40)	18 (3.9–39)	47 (28–93)	<0.01
Valvular involvement	30 (6.4–65)	0.6 (0.6–0.6)	55 (7.8–102)	30 (18–62)	<0.01
Rhythm disorders	28 (12–66)	16 (5.7–40)	26 (8.7–43)	33 (16–79)	<0.01
Angina	24 (10–86)	11 (3.0–31)	48 (24–86)	15 (8.3–99)	<0.01
Cardiac enlargement	16 (4.2–41)	6.1 (3.2–15)	11 (2.2–28)	28 (7.0–52)	<0.01
Multiple histories	16 (1.6–38)	5.9 (3.3–12)	15 (1.3–45)	23 (1.6–44)	<0.01
Pericardium damage	10 (6.0–26)	10 (6.0–26)	N/A	N/A	<0.01
Heart failure	9.8 (3.8–25)	6.1 (2.5–12)	16 (5.1–35)	18 (6.6–38)	<0.01
Cardiac biomarkers increased or cardiac imaging abnormalities (treated together)	0 (0–4.3)	3.3 (0.0–10)	0 (0–0)	0.6 (0–17)	<0.01
First ACE or AECE					
Age at the time of the first ACE or AECE, years	66.1 ± 12.1	62.7 ± 11.6	62.7 ± 11.1	70.0 ± 11.8	<0.01
Delay between the first ACE or AECE and the diagnosis (in months), median (Q1–Q3)	49.9 (11.4; 125.7)	11.1 (5.9; 34.8)	57.5 (23.5; 119.4)	92.2 (39.0; 174.7)	<0.01
Classes of time between the first ACE or AECE and the diagnosis, *n*(%)					
<2 years	337 (36)	207 (68)	51 (26)	79 (18)	<0.01
Between 2 and 5 years	177 (19)	54 (18)	54 (27)	69 (16)	
>5 years	418 (45)	41 (14)	94 (47)	283 (66)	

[a]Lymphadenopathy, gammopathy, haemoptysis.
Underlined are the four most frequent events, all types of amyloidosis combined.

Figure 3 Chord diagrams describing the link between time frame [(0–2) years in blue, (2–5) years in purple and (5 and +) years in pink] of cardiac amyloidosis diagnosis and the occurrence of the first amyloidosis extracardiac (light grey), cardiac (black), or both (dark grey) events depending on type of amyloidosis. The diagnosis of AL is performed mainly in the 2 years after the first symptoms, especially cardiac symptoms. The diagnosis of ATTRv and ATTRwt is mainly performed later than 5 years after the onset of symptoms, especially integumentary symptoms.
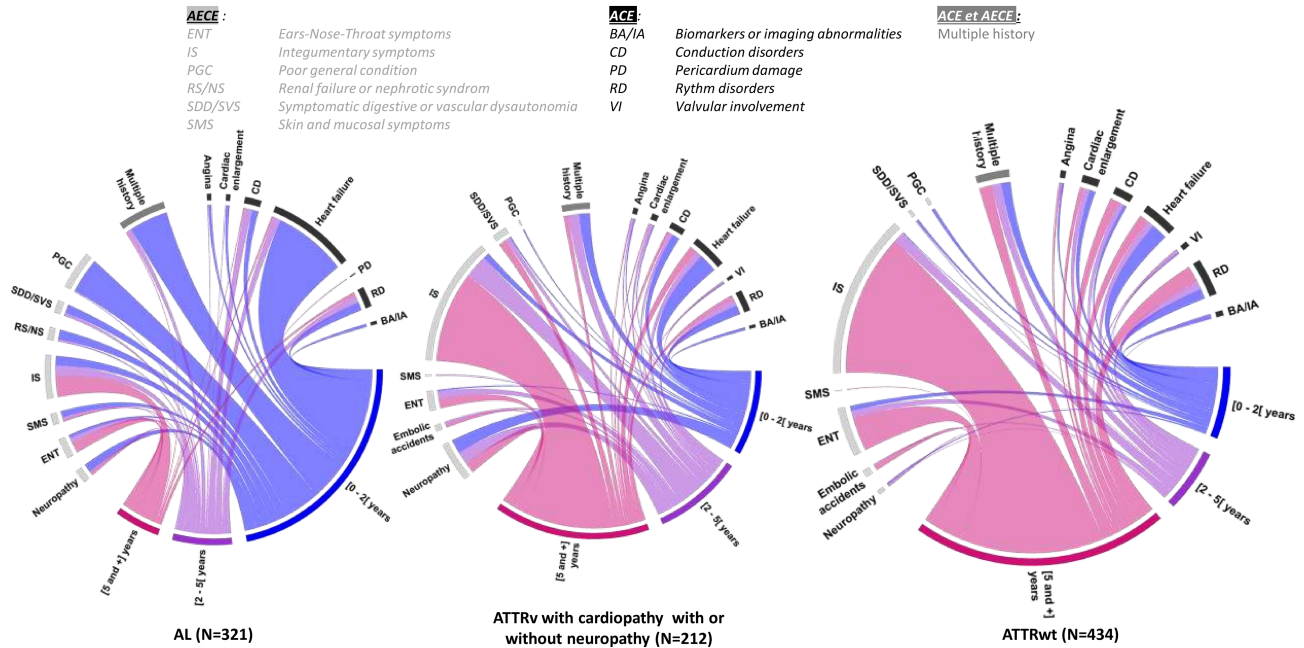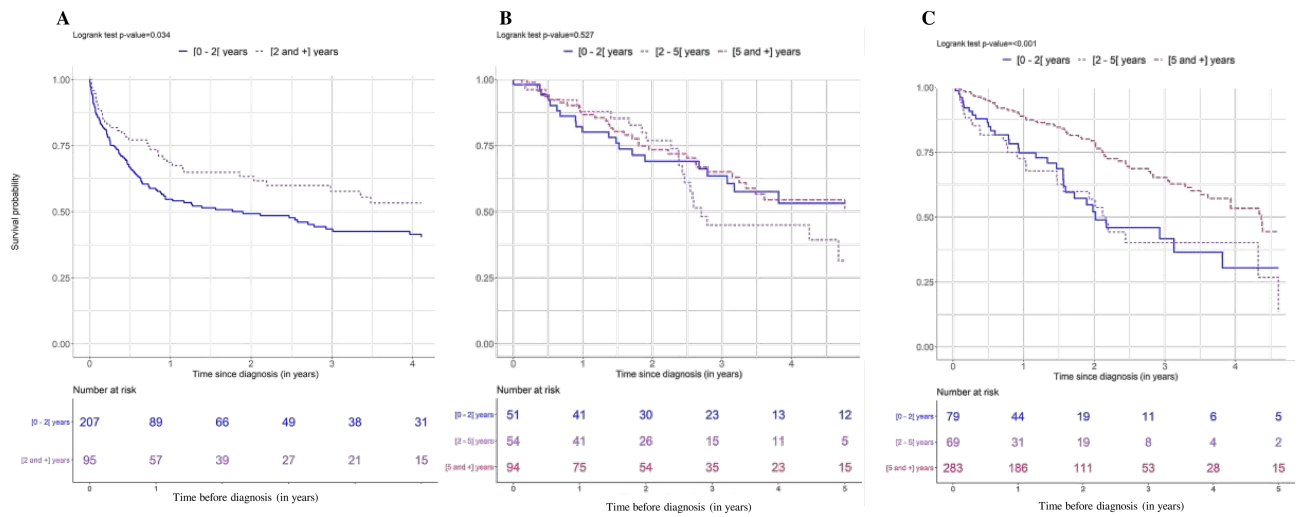

Figure 4 Kaplan–Meier curves describing the link between death or heart transplantation and delayed diagnosis [(0–2) years in blue and (2 and +) years in purple] (A), or [(0–2) years in blue, (2–5) years in purple, and (5 and +) years in pink (B) and (C)] for each type of amyloidosis: (A) AL; (B) ATTRv with cardiomyopathy with or without neuropathy; and (C) ATTRwt.


Discussion

To our knowledge, our study is the first comprehensive description of the heterogeneity of cardiac and extracardiac disorders according to underlying amyloidosis types using a large real-life cohort. Thus, patients with AL have a higher tendency to present poor general condition, heart failure symptoms, nephrotic syndrome or renal failure, and skin or mucosal symptoms, than patients with ATTR, especially if the diagnosis is made after 2 years.

Figure 5 Kaplan–Meier curves describing the link between death or heart transplant and first amyloidosis extracardiac or cardiac events (AECE or ACE) for the three main classes of symptoms: ears-nose-throat (ENT) symptoms (solid, dark grey line), heart failure (dotted, black line), and integumentary symptoms (dotted, light grey line).
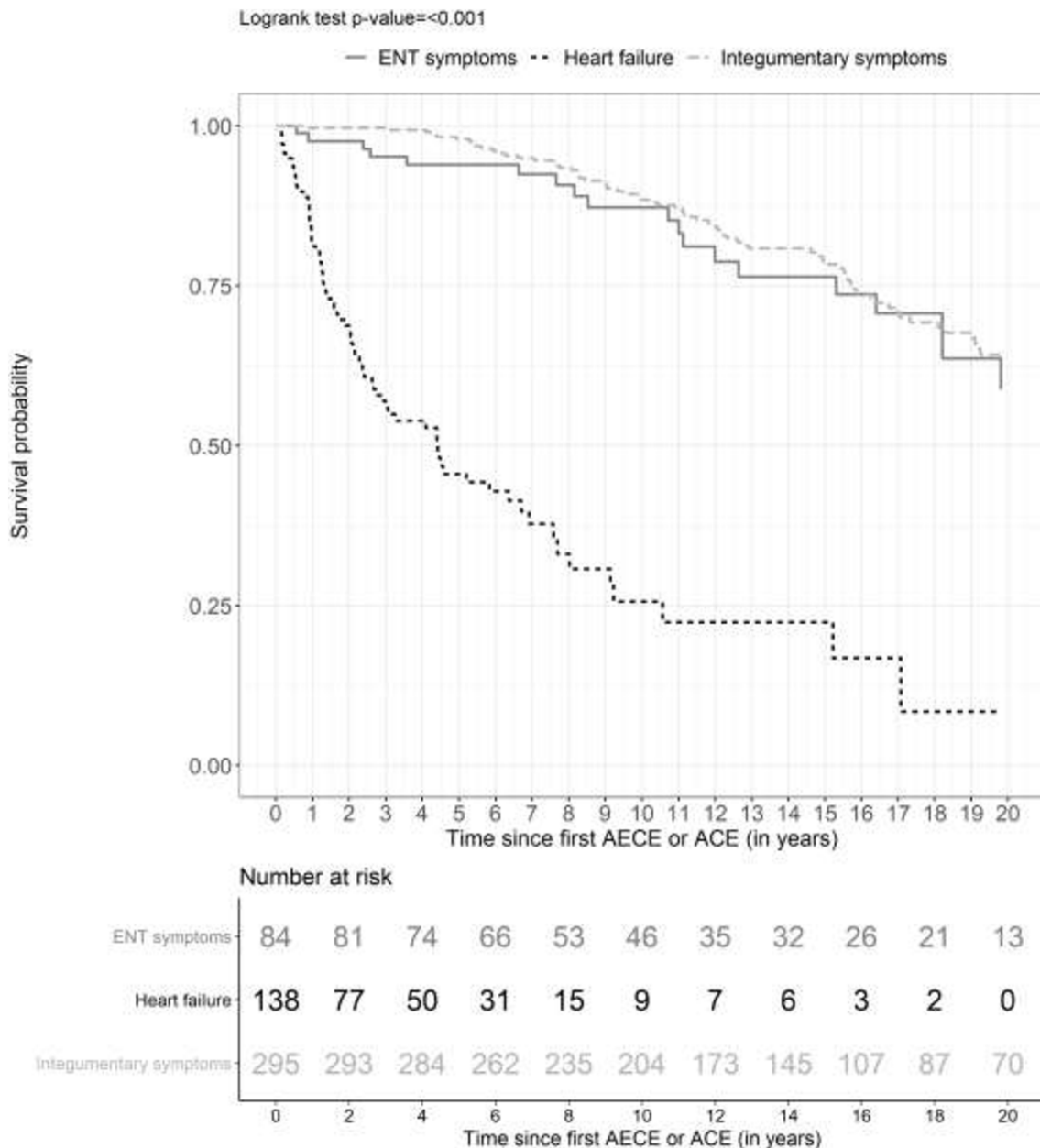
Additionally, although ATTRwt had more integumentary symptoms and hearing loss than ATTRv, diagnostic delay could be >5 years.

This study of natural history of amyloidosis highlights the impact of amyloidosis type and evolution on diagnostic delay and on prognosis. Physicians must be aware and vigilant in front of extracardiac and cardiac events in order to considerably improve early diagnosis of amyloidosis.

Amyloidosis extracardiac and cardiac events depending on amyloidosis type

The AL typically occurs in men or women in the sixth decade; however, it has a wide range of age at onset. The initial symptoms can be extracardiac or cardiac disorders, which typically occur shortly before diagnosis [median delay; 11.1 (5.9; 34.8) months (*Table 2*], suggesting that patients not diagnosed in

the two first years of disease died rapidly. AL is a systemic disease, affecting predominantly the heart and kidneys.[2,14,15] Extracardiac involvement can be seen in biphosphonate imaging in patients with AL even if the cardiac fixation of the bone tracer was not observed.[16] Compared with other amyloidosis types, the most common initial symptoms of AL patients were poor general condition and heart failure symptoms (*Figures 2 and 3*). Nephrotic syndrome or renal failure and skin or mucosal symptoms (such as periorbital ecchymosis and macroglossia) seem also more specific to this subtype, as these symptoms were rarely observed in ATTRv and were almost never observed in ATTRwt. In our study, renal failure or nephrotic syndrome as first manifestations were only associated with AL (8.2% vs. 0 and 0.3% for ATTRv and ATTRwt, respectively). This is in line with results reported by Rapezzi *et al.*[17] and in the recent study by Desport *et al.*, in which kidney involvement was a frequent early manifestation of AL, verified in two-thirds of patients at the time of diagnosis.[14] However, when considering only the initial symptoms, we did not find such a high prevalence in our cohort, which suggests a bias due to differences in recruitment between cardiac or nephrologic referral centres. Indeed, we also observed that AL was associated with a wide range of various manifestations, such as diarrhoea, severe orthostatic hypotension, dysphonia, dysphagia, and/or ageusia, which contributed to weight loss and altered general condition and contributing in misleading the diagnosis. Integumentary symptoms were observed in one fourth of patients with AL, occurring in most cases 2–5 years before AL diagnosis. Canal tunnel surgery (CTS) could contribute to earlier diagnosis of AL, if carpal tunnel biopsies were screened for kappa and lambda deposits.[11]

The ATTRv must be separated in two different phenotypes: ATTRv with neuropathy, excluded of the study, occurring mostly in young men and women of Portuguese descent (early onset, <55 years of age); and ATTRv with cardiomyopathy with or without neuropathy occurring in patients older than 55 years.[1] ATTRv with cardiomyopathy with or without neuropathy mimics ATTRwt regarding initial extracardiac or cardiac manifestations (*Figure 3*). Thus, it might be difficult to distinguish the two diseases in individual patients using only phenotype or past medical history. This finding emphasizes the need to perform genetic TTR testing in all patients with ATTR to diagnose ATTRv and to provide proper care of the patient's family through genetic counselling.[18]

The ATTRwt is observed particularly in old men (mean age of 80 years). In this setting, the initial symptoms include integumentary symptoms that occurred in 42% of cases and that were verified a substantial amount of time before diagnosis (mean diagnostic delay of 140 months). The association between CTS (included in 'integumentary symptoms' analysed here) and systemic amyloidosis has been widely described, and the main case series reports a high prevalence of CTS in ATTR, ranging from 15% to 68%.[17,19–22] In line with our

results, CTS preceded cardiac symptoms and ATTR diagnosis by a mean of 6.1 and 6.9 years, respectively.[11] These observations have major clinical implications for cardiologists and all physicians involved in integumentary symptom diagnosis and CTS, including the general practitioner.[8] Although CTS is a common manifestation of ATTR, it has not been well recognized by most surgeons or cardiologists in clinical practice.[11] In contrast with our previous work,[23] hearing loss was less frequently observed in ATTR[23]; however, in the present study, symptoms were reported by patients who had not undergone systematic audiometry measurement. Despite this, as expected, frequency of deafness was higher in ATTRwt patients than other amyloidosis. Therefore, we can conclude that patients are not necessarily conscious of their hearing loss or are unwilling to report it.

Our study showed that AECE and ACE differed among the three main types of amyloidosis regarding proportion and time of occurrence. All the information gathered provides a clearer picture of the natural history and phenotypes of CA that should facilitate early diagnosis by physicians.

Discrepancies in delay between initial symptoms and amyloidosis diagnosis

The first large study to document a delay in ATTR diagnosis was the THAOS registry,[22] reporting a delay in diagnosis >4 years since the onset of symptoms. In our study, the median delay between the initial AECE/ACE and diagnosis varied from 11 (6–35) months, for AL, to 92 (39–175) months, for ATTRwt. Bishop *et al.* reported a mean diagnostic delay of 22 months in 82 patients with ATTR with CA (34 months).[24] In comparison with the analyses performed by Bishop *et al.*, our results could be explained by the wide range of manifestations that were included in our study.

The diagnosis of ATTRwt is delayed because these patients have age-related symptoms that may lead physicians to consider other coexisting heart diseases that are common in the elderly population. The diagnosis of ATTRwt requires a high index of clinical suspicion, which can be increased by identifying diagnostic red flags.[25] Although integumentary symptoms represent the most frequent extracardiac event, the median time between their onset and diagnosis of amyloidosis is noteworthy: 121 (65–194) months varying from 46 (20–113) months for AL to 140 (86–237) months for ATTRwt. This highlights the difference in kinetics and toxicity of the process of amyloid fibril formation and/or deposition between AL and ATTRwt; thus, the shorter delay between initial onset of cardiac and extra-cardiac manifestations and the diagnosis is expected in AL vs. ATTR because of the toxic-infiltrative nature of its form. Similarly, it is not surprising that in ATTRwt, a longer time course between extracardiac manifestations and diagnosis with survival was observed. This is consistent with

the known natural history of these conditions and supports the need for rapid diagnosis in AL due to poor prognosis.

Relationship between heterogeneity and incidence of cardiac amyloidosis phenotypes and amyloid fibril formation kinetics

In our study, we were surprised to observe that a prolonged period between initial symptoms and diagnosis seems to be associated with increased survival for AL and ATTRwt. This could be explained by a heterogeneity of the speed of the amyloidogenesis process resulting in organs infiltration and dysfunction. Accordingly, several studies have shown that each type of amyloid protein precursor has a different affinity for target organs[26] and different multiplication kinetics.[27] Patients with a slower amyloidogenic process developed symptoms gradually letting them to be diagnosed after several years of progression. Patients with a fast amyloidogenic process developed severe symptoms and died rapidly after diagnosis or might be undiagnosed. This finding can have important implications in clinical practice, because patients with amyloidosis combining cardiac and extracardiac events in a short period of time (less than 2 years for AL or less than 5 years for ATTRwt) have a worse prognosis and, therefore, could benefit from more aggressive care vs. the others.

Study limitations

Given the large number of antecedents associated with CA, we had to group them to facilitate the analyses. The large number of antecedents allowed us to have a broad overview of the natural history of the disease. History was based solely on the number of months and years. This information was collected retrospectively in medical records; however, sometimes it was based on information provided by the patients, which could be biased due to age-related memory deficits. Thus, the retrospective nature of the data collection could create a bias in determination of manifestation occurred first. Erectile dysfunction and biceps rupture, or other ligament ruptures, were missing from the patients' record. We recognized that erectile dysfunction is an important red flag in young male patients with ATTRv; therefore, in our population composed of elderly men and women, the relationship between CA and erectile dysfunction was difficult to determine. Biceps rupture was not described at the time we began recording all the signs described here.[28]

This study showed that history of amyloid extracardiac and cardiac events occurring early in the development of CA had typical features depending on the type of amyloidosis. Measuring the time of onset for each event may help with diagnosis, typing, and determining the prognosis of amyloid-

osis, and an increase in physicians' awareness of these events may improve diagnosis.

Ethics statement

The authors should state that their study complies with the Declaration of Helsinki, that the locally appointed ethics committee has approved the research protocol, and that informed consent has been obtained from the subjects (or their legally authorized representative).

Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Flowchart of study subgroups population and exclusion criteria.
Table S1. Baseline characteristics of the study population: AL, ATTRv with cardiomyopathy with or without neuropathy and ATTRwt subgroups.
Table S2. Description of frequency of the first amyloidosis extra-cardiac (AECE) and cardiac events (ACE) depending on type of amyloidosis.

Table S3. Description of all grouped extra-cardiac and cardiac events depending on type of amyloidosis.

Table S4. Frequency of initial onset of extra-cardiac and/or cardiac event declared before diagnostic depending on type of amyloidosis.

Table S5. Description of number of deaths, and median time (in months) between diagnosis and death or cardiac transplantation and between initial onset of amyloid event and death depending on type of amyloidosis.

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