Archives of Gastroenterology and Hepatology

ISSN: 2639-1813

Volume 3, Issue 2, 2020, PP: 64-83

Demographics and Predictive Clinical-Laboratory Parameters of Systemic and Hepatic AA Amyloidosis – A Postmortem Clinicopathologic Study of 152 Rheumatoid Arthritis Patients

Miklós Bély[1], Ágnes Apáthy[2]

[1]Department of Pathology, Hospital of the Order of the Brothers of Saint John of God, Hungary.
[2]Department of Rheumatology, St. Margaret Clinic, Budapest, Hungary.

***Corresponding Author:** Miklós Bély M.D., Ph.D., D.Sc. Acad. Sci. Hung, Hospital of the Order of the Brothers of Saint John of God in Budapest, Hungary, Department of Pathology, H- 1027 Budapest, Frankel L. 17-19, Hungary.*

Abstract

*The **aim** of this study was to determine the demographics of **s**ystemic **AA a**myloidosis syndrome (**sAAa**), and **h**epatic **AA a**myloidosis (**hAAa**) in rheumatoid arthritis (**RA**), to assess the predictive clinical laboratory parameters.*

***Patients (autopsy population) and Methods:** The demographics and clinical laboratory parameters were analyzed based on **152** autopsy patients with **RA**. **RA** was confirmed clinically according to the criteria of the American College of Rheumatology (**ACR**). The patients were treated and died in one institute (National Institute of Rheumatology, Budapest, Hungary) between 1969 and 1992.*

*The **sAAa** and **hAAa** was specified histologically, based on evaluation of five organs (kidneys, heart, pancreas, lungs, and liver).*

Amyloid A deposition in different tissue structures of various organs was diagnosed histologically according to Romhányi, by a modified (more sensitive) Congo red staining.

*The correlations were determined by the Student (Welch) t-probe, comparing the age, sex, onset of **RA**, duration of disease, and classic laboratory parameters at the last hospitalization with or without **sAAa** or **hAAa**.*

***Results and Conclusions:** Amyloidosis may develop in both sexes and at any time of the disease.*

The diagnostic values of the discussed laboratory parameters are limited, and none are specific for amyloidosis

*The more or less significant differences between **RA** patients **with** and **without sAAa** show the impaired function of the kidneys or are connected to the basic disease only.*

*There is no significant difference in classic laboratory parameters between cohorts of **RA** patients **with sAAa** and **hAAa**.*

*For exact diagnosis of amyloidosis a biopsy is needed using an „appropriate staining procedure". Gingival or rectal biopsies are suggested. The increased erythrocyte sedimentation rate associated to laboratory parameters of impaired renal function arises the possibility of **hAAa**, and especially in hepatomegaly, a liver biopsy may be considered to confirm **hAAa**.*

Keywords: *Rheumatoid arthritis, demographics, clinical laboratory parameters, systemic AA amyloidosis, AA amyloidosis of the liver*

Abbreviations

RA – Rheumatoid Arthritis

ACA – American College of Rheumatology

AA – **a**myloid **A** protein

AAa – **AA a**myloidosis (**A**myloid **A** protein **a**myloidosis)

sAAa – systemic **AA a**myloidosis

hAAa – **h**hepatic **AA a**myloidosis

SAA – **S**erum **a**myloid **A**

SAP – **S**erum **a**myloid **P** component

ESR – Erythrocyte sedimentation rate

CRP – C reactive protein

A/G ratio – Albumin/**G**lobulin ratio

BUN– blood urea nitrogen

RBC – red blood cells

WBC – white blood cells

GOT– **G**lutamat-**O**xalacetat-**T**ransaminase (=AST – Aspartate transaminase)

GPT – **G**lutamat-**P**iruvat **T**ransaminase (=ALT – Alanine transaminase)

GGT (**gamma GT**) – **G**amma-**G**lutamyl **T**ranspeptidase

LDH – **L**actate **De**h**y**drogenase

DMARDs– Disease **M**odifying **A**ntirheumatic **D**rugs

TNF-α – **T**umour **N**ecrosis **F**actor alpha (**α**)

CoD – Cause of death

U – Uremia

Cl+ – clinically diagnosed

Cl- – clinically not diagnosed

ND – No Data available

NS - Not Significant

SD – Standard **D**eviation

Introduction

Amyloidosis syndromes are systemic or localized disorders characterized by the extracellular deposition of chemically heterogeneous fibrillar proteins. The amyloidosis syndromes are named according to these fibrillar proteins [1, 2]. After **a**myloid **A** protein (**AA**) deposits the disease is named **AA a**myloidosis (**A**myloid **A** protein **a**myloidosis – **AAa**). The precursor of amyloid A protein fibrils is the **s**erum **a**myloid **A** (**SAA**) polypeptide [1, 2]. **SAA** is produced by the liver, spread via the bloodstream, and is deposited in target organs throughout the body, leading toa **s**ystemic **AAa** syndrome (**sAAa**). All forms of systemic amyloidosis are connected to the circulation, and all forms of amyloidosis not connected to the circulation are isolated (localized, organ or tissue limited) [3, 4, 5]. **sAAa** is related to the cardiovascular system, and **h**epatic **AA a**myloidosis (**hAAa**) is connected with it [6].

The **aim** of this study was to determine the demographics of **sAAa** and **hAAa** in **RA**, to assess the predictive clinical laboratory parameters for severe or mild **sAAa** and **hAAa**.

Patients (Autopsy Population) and Methods

The demographics and clinical laboratory parameters were analyzed based on **152** autopsy patients with rheumatoid arthritis (**RA**). **RA** was confirmed clinically according to the criteria of the American College of Rheumatology (**ACR**) [7]. The patients were treated and died in one institute (National Institute of Rheumatology, Budapest, Hungary) between 1969 and 1992 in an era of steroid and disease-modifying antirheumatic drugs (**DMARDs**), before introduction of biological therapy (anti-**TNF-α** treatment, etc.).

The **sAAa** and **hAAa** was specified histologically, based on evaluation of five organs (kidneys, heart, pancreas, lungs, and liver).

Amyloid A deposition in different tissue structures of various organs was diagnosed histologically according to Romhányi [8] by a modified (more sensitive) Congo red staining [9]. The prevalence (existence) and severity (extent) of amyloid A deposition were evaluated microscopically by semi-quantitative visual estimation on a 0 to 3 plus scale, based on the number of involved bloody vessels or tissue structures/light microscopic field x40 objective lens of an Olympus BX51 polarizing microscope.

Footnote

Semi-objective score system of amyloid A deposition: "**0**" – no amyloid deposits, "**1**" –sporadic, minimal amyloid deposits in different tissue structures, "**2**" – less than five involved tissue structures, "**3**" – five or more involved tissue structures/light microscopic field x40 objective lens. **Remark**: in case of medium size arteries and veins this corresponds to the absolute number of involved medium size vessels of a tissue sample, e.g. "**0**" none, "**1**" only one,"**2**" less than five, "**3**" 5 or more than five medium size vessels/tissue sample with a x20 objective lens.

The correlations were determined by the Student (Welch) t-probe [10], comparing the age, sex, onset of **RA**, duration of disease, and classic laboratory parameters {Latex, Waaler-Roose values, ESR, CRP, serum albumin/globulin ratio, serum electrophoresis (albumin, alpha-1-globulin, alpha-2-globulin, beta-globulin, gamma-globulin), RBC, hemoglobin, WBC, blood urea nitrogen (BUN), serum creatinin, serum potassium and sodium values, urine specific gravity, proteinuria, urine sediment (RBC, WBC), serum bilirubin, LDH, GPT, GGT, blood sugar, and diastase values, systolic and diastolic blood pressure} at the last hospitalization with or without **sAAa** or **hAAa**, and with "**mild**" (**< 0.8** / patient) or "**severe**" amyloid A deposits (**0.8 ≤** / patient).

Footnote

GPT – **G**lutamate-**P**iruvate **T**ransaminase (=ALT – Alanine transaminase), or immunological parameters (**SAA**, serum amyloid P component – **SAP**, etc. were not analyzed between 1969 and 1992 in our Institute.

Results

sAAa complicated **RA** in **32** (21.05 %) of 152 patients. **sAAa** was histologically excluded in **120** (78.95 %) of 152 **RA** patients.

Seventeen (53.125 %) of 32 patients had "**slight** (**mild**)" amyloidosis (with average amyloid A deposits / patient **<0.8**), involving only a few tissue structures in the kidneys, heart, pancreas, and lung.

Fifteen (46.875 %) of 32 patients revealed "**marked** (**severe**)" amyloidosis (with average amyloid A deposits / patient **0.8 ≤**), massively involving many tissue structures of the kidneys, heart, pancreas, and lung.

Branches of blood vessels of different calibers and various tissue structures of the liver were involved in **26** (81.25 % of 32, 17.11 % of 152) cases; **hAAa** was histologically excluded in **6** (18.75 % of 32) patients with **sAAa**.

In **15** (46.875 %) of 26 **RA** patients with **hAAa** the average amount of amyloid A deposits of the liver was less than **<0.8**, and was regarded "**slight** (**mild**)", and in **11** (34.375 %) of 26 patients it was more than **0.8 ≤**, and was considered "**marked** (**severe**)".

In Table 1 are summarized the demographics, onset and duration of disease of the **total population** (n=**152**), **with mild** (n=**17**) and **severe** (n=**15**) **sAAa** (n=**32**), **without sAAa** (n=**120**), **with mild** (n=**15**) and **severe** (n=**11**) **hAAa** (n=**26**), and **without** (n=**6**) amyloid A deposits of the **liver**.

Table1. *Sex, mean age with SD, range, onset and disease duration (in years) of 152 RA patients with or without sAAa and hAAa*

(Extended data published by Miklós Bély and Ágnes Apáthy in: Archives of Gastroenterology and Hepatology, 2020,3.2: 1:26 [6])

Sex	Number of autopsies	Mean age in years at death ± SD	Range (in years)	Mean age at onset of disease ± SD	Disease duration (in years) mean ± SD
RA patients (total)	**152**	**65.81±13.06**	**16 – 88**	**51.43±17.20**	**14.30±10.61**
Female	108	65.54±11.85	16 – 87	50.99±15.89	14.54±10.78
Male	44	66.45±15.62	19 – 88	52.56±20.15	13.69±10.12
with sAAa	**32 of 152**	**63.25±15.64**	**19 – 88**	**48.17±18.41**	**16.00±9.51**
Female	27	65.48±10.54	32 – 83	49.28±15.48	16.20±10.08
Male	5	51.20±28.18	19 – 88	41.25±30.07	14.75±4.44
with mild sAAa	**17 of 32**	**69.06±10.55**	**50 – 88**	**55.59±14.28**	**13.47±9.12**
Female	16	67.88±9.72	50 – 83	54.13±13.43	13.75±9.33
Male	1	88.00±0.00	88	79.00±00.00	9.00±0.00
with severe sAAa	**15 of 32**	**56.67±17.73**	**19 – 82**	**37.67±18.49**	**19.58±8.88**
Female	11	62.00±10.71	32 – 73	40.67±15.14	20.56±9.88
Male	4	42.00±23.86	19 – 82	28.67±23.92	16.62±3.40

without sAAa	**120 of 152**	**66.49±12.19**	**16 – 87**	**52.37±16.71**	**13.81±10.86**
Female	81	65.56±12.25	16 – 87	51.62±15.99	13.93±10.97
Male	39	68.41±11.83	20 – 87	53.97±18.05	13.56±10.61
with hAAa	**26 of 32**	**61.31±9.34**	**19 – 88**	**47.83±19.83**	**14.39±9.34**
Female	21	63.71±10.65	32 – 83	49.21±16.57	14.32±2.00
Male	5	51.20±28.18	19 – 88	41.25±30.07	14.75±4.44
with mild hAAa	**15 of 26**	**66.80±10.34**	**50 – 88**	**55.50±15.04**	**11.50±8.14**
Female	14	65.29±8.96	50 – 83	53.69±14.06	11.69±8.42
Male	1	88.00±0.0	88	79.00±0.0	9.0±0.0
with severe hAAa	**11 of 26**	**53.82±19.79**	**19 – 82**	**35.89±20.47**	**18.89±9.31**
Female	7	60.57±12.84	32 – 73	39.50±17.42	20.00±10.98
Male	4	42.00±23.86	19 – 82	28.67±23.92	16.67±3.40
without hAAa	6 of 32	71.67±7.34	**66 – 82**	**49.50±11.34**	**22.17±7.40**
Female	6	71.67±7.34	66 – 82	49.50±11.34	22.17±7.40
Male	0	–	–	–	–

Glossary to Table 1

RA – Rheumatoid **A**rthritis
sAAa – systemic **AA a**myloidosis
hAAa – hepatic **AA a**myloidosis (amyloid A deposits in the liver)
SD – Standard **D**eviation

There was **no significant difference** in **survival time** (63.25 ys versus 66.49 ys; p< 0.290 – NS), **onset** (48.17 ys versus 52.37 ys; p< 0.284 – NS), and **duration** of **disease** (16.00 ys versus 13.81 ys; p< 0.303 – NS), between **RA** patient cohorts **with sAAa** (n=32) and **without sAAa** (n=120), **with sAAa** (n=32) and **with hAAa** (n=26) or **with hAAa** (n=26) and **without hAAa** (n=6), neither **females** nor **males** (except the age of patients at death **with hAAa** and **without hAAa**; the patients **with hAAa** died earlier, than the patients **without hAAa** (61.31 ys versus 71.67 ys; **p <0.040**) (Tables 1 and 2).

Comparing **age, sex, onset of RA**, and **duration of disease** at the time of death **RA** started earlier in patients **with severe sAAa** (n=15) in comparison to the patients **with mild sAAa** (n=17) (37.67 ys versus 55.59 ys; p<0.095 – NS) or compared to the patients **without sAAa** (n=120) (37.67 ys versus 52.37 ys; p<0.065 – NS).

Disease duration **with severe** systemic amyloid A deposition was significantly longer in comparison to the patients **with mildsAAa** (19.58 ys versus 13.47 ys; p< **0.014**) or compared to patients **without sAAa** (19.58 ys versus 13.81 ys; p< **0.025**).

The patients **with severe sAAa** (n=15) died significantly earlier in comparison to the patients **with mildsAAa** (n=17) (56.67 ys versus 69.06 ys; p< **0.032**) or compared to the patients **without sAAa** (n=120) (56.67 ys versus 66.49 ys; p<0.061 – NS) (Tables 1 and 2).

Comparing **age, sex, onset of RA**, and **duration of disease** at the time of death **RA** started earlier in patients **with severe hAAa** (n=11) in comparison to the patients **with mild hAAa** (n=15) (35.89 ys versus 55.50 ys; p<0.804 – NS) or compared to the patients **without hAAa** (n=6) (35.89 ys versus 49.50 ys; p<0.148 – NS).

Disease duration **with severe hAAa** was significantly longer in comparison to the patients **with mild hAAa** (18.89 ys versus 11.50 ys; p< **0.035**) or compared to patients **without hAAa** (11.89 ys versus 22.17 ys; p<0.496 – NS).

The patients **with severe hAAa** (n=11) died significantly earlier in comparison to the patients **with mild hAAa** (n=15) (53.82 ys versus 66.80 ys; p<0.079 – NS) or compared to the patients **without hAAa** (n=6) (53.82 ys versus 71.67 ys; p< **0.024**) (Tables 1 and 2).

Demographics, onset and duration of disease of female and male **RA** patients **with** (n=32) and **without sAAa** (n=120), are summarized in Figures 1.a-e – 2.a-e

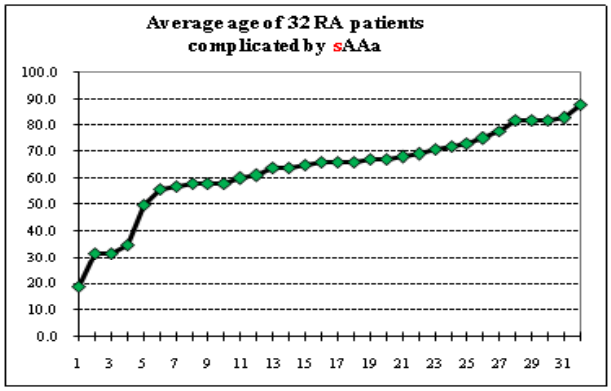

Figure1.1a

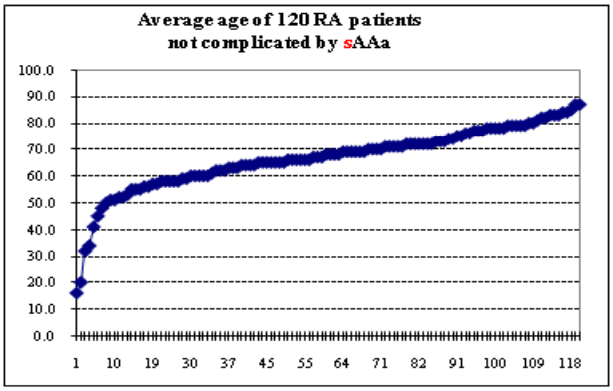

Figure1.2a

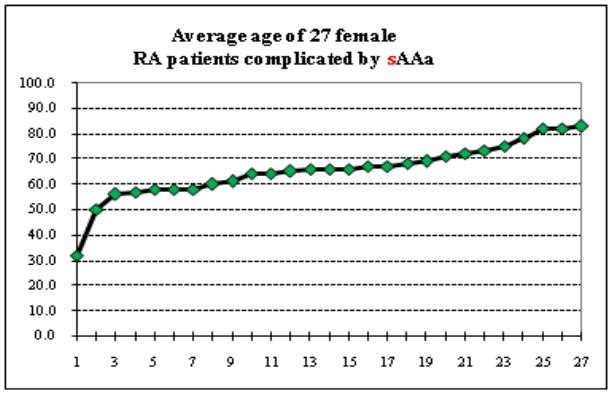

Figure1.1b

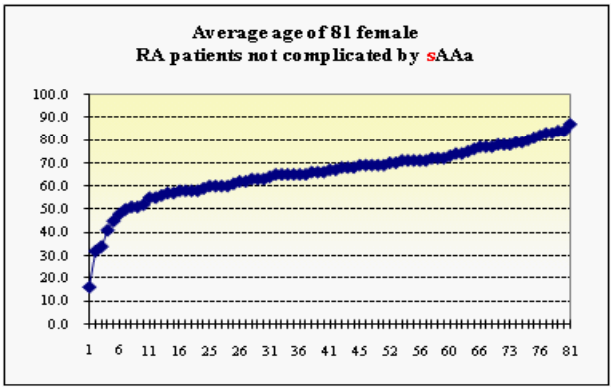

Figure1.2b

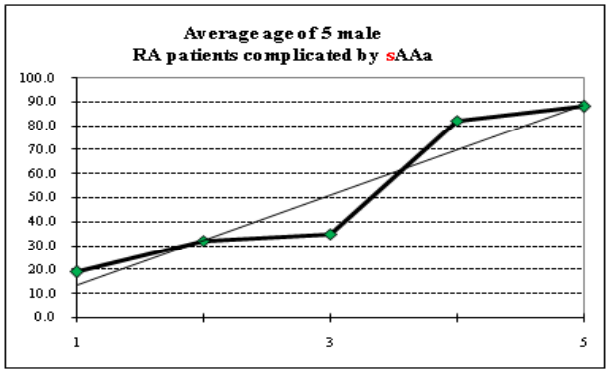

Figure1.1c

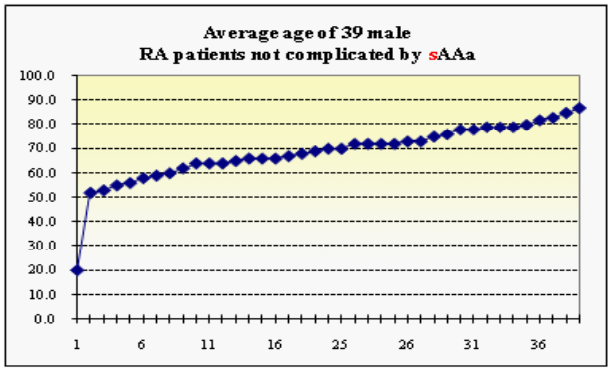

Figure1.2c

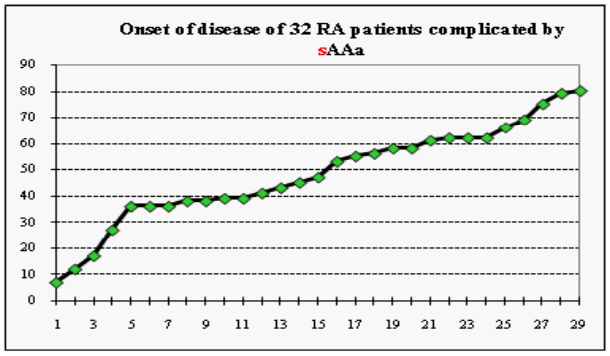

Figure1.1d

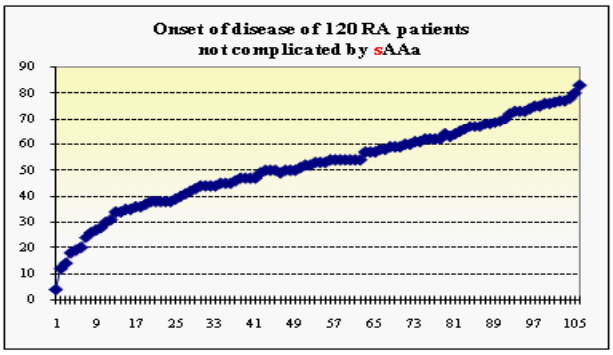

Figure1.2d

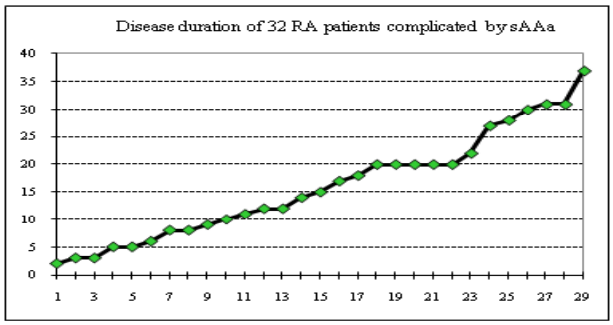

Figure1.1e

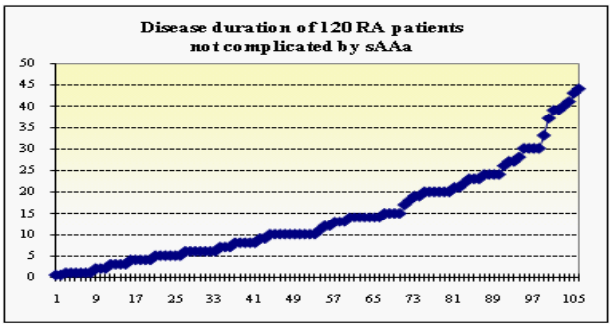

Figure1.2e

Figure1.1a-e – 1.2a-e. *There was no significant difference in survival time, onset and duration of disease between RA patients with sAAa (n=32) and without sAAa (n=120), neither females nor males*

Demographics, onset and duration of disease of **RA** patients **with mild** (n=17 of 32) and **severe sAAa** (n=15 of 32) are summarized in Figures 1.3a-c – 1.4a-c.

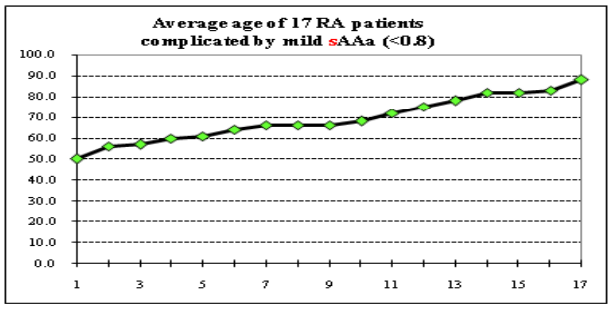

Figure1.3a

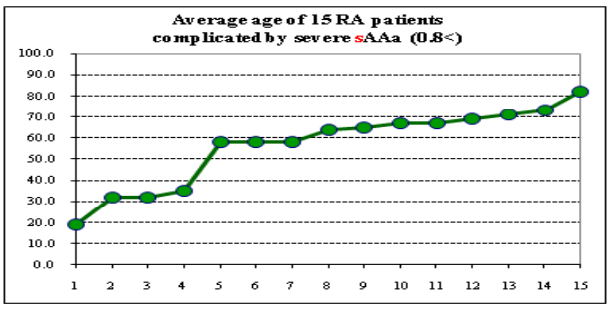

Figure1.4a

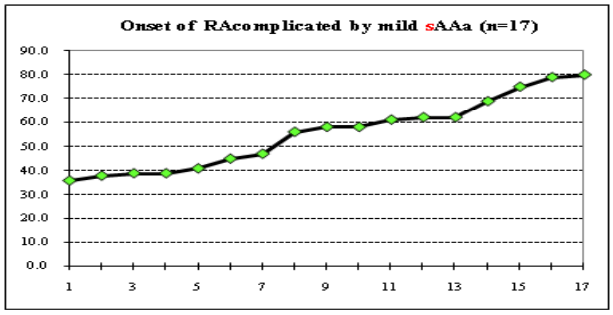

Figure1.3b

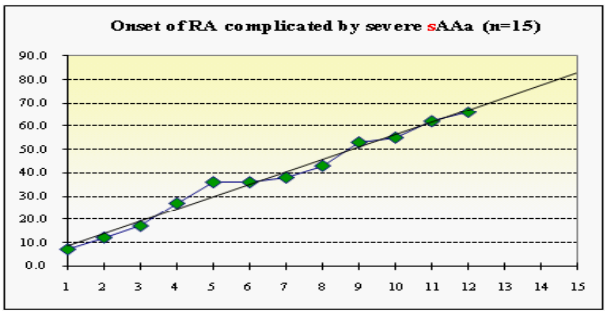

Figure1.4b

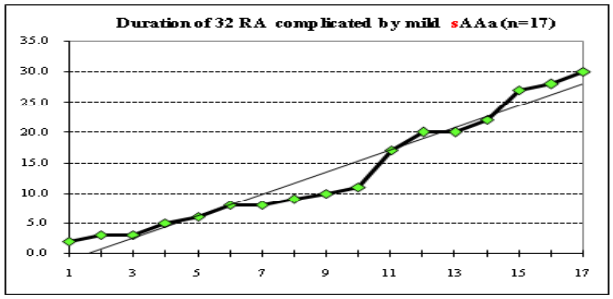

Figure1.3c

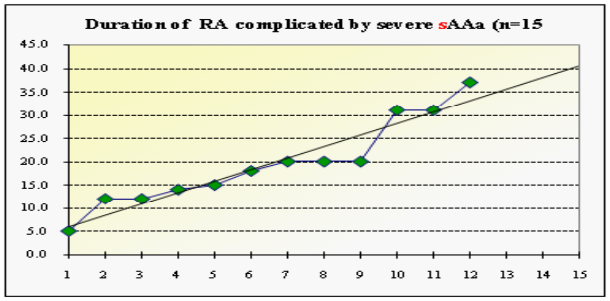

Figure1.4c

Figure 1.3a-c – 1.4a-c. *RA started earlier (p <0.095 - NS) in patients with severe sAAa (n=15), duration of RA was significantly longer (p<0.014), and the patients died significantly earlier (p<0.032), compared to the patients with mild sAAa (n=17)*

Demographics, onset and duration of disease of **RA** patients **with hAAa** (n=26 of 32) and **without hAAa** (n=6 of 32) are summarized in Figures 1.5a-c – 1.6a-c

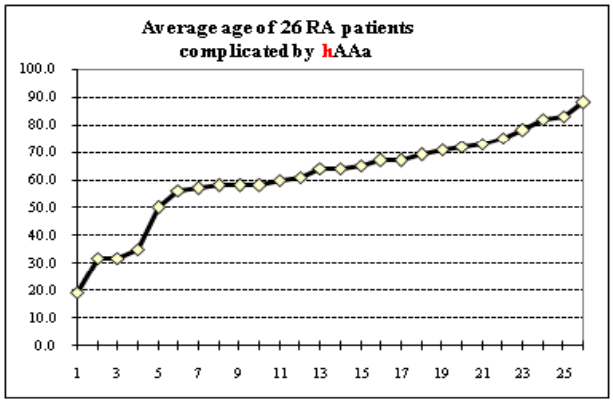

Figure1.5a

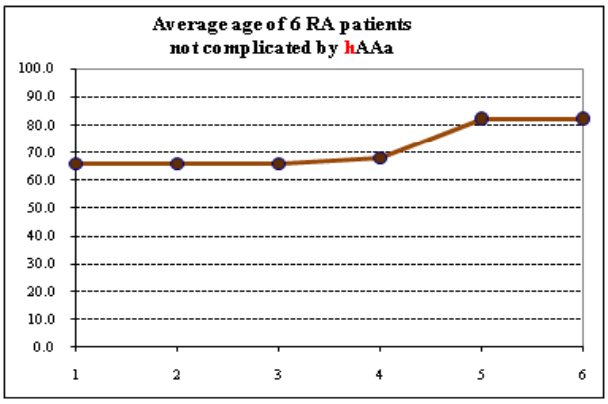

Figure1.6a

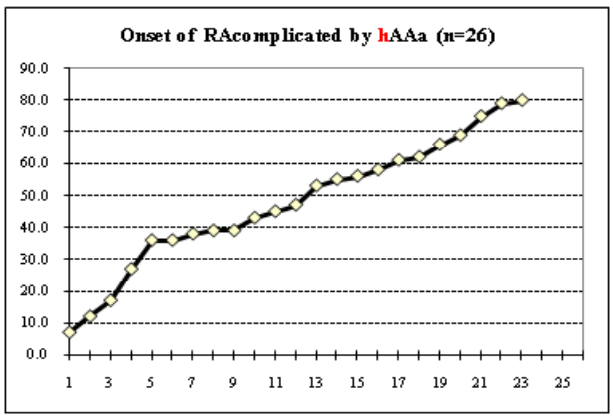

Figure1.5b

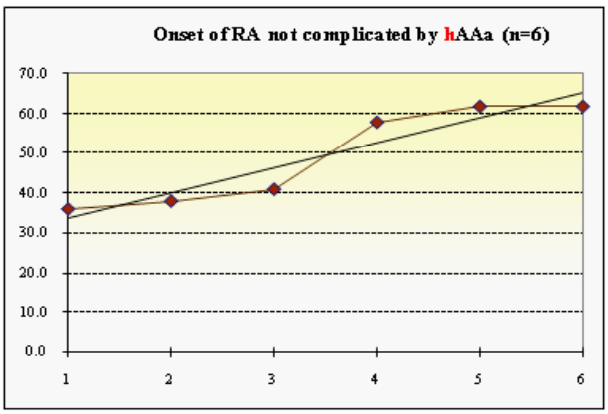

Figure1.6b

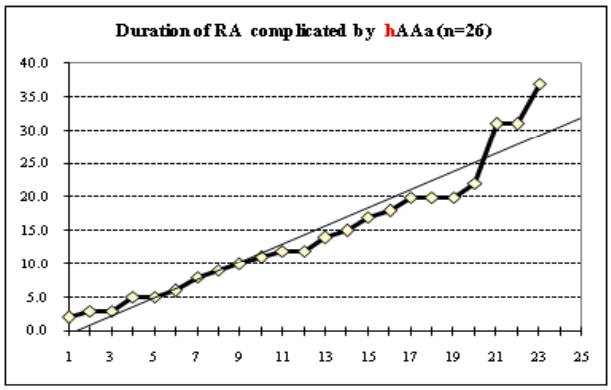

Figure1.5c

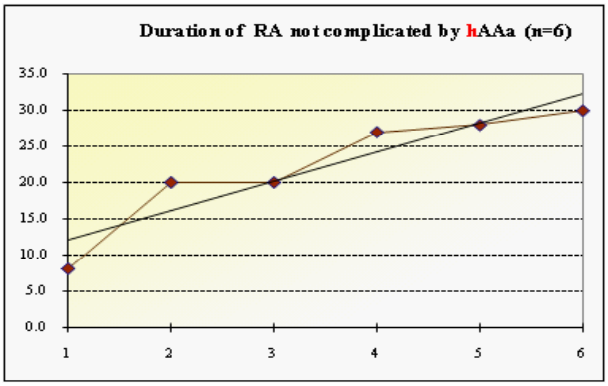

Figure1.6c

Figure 1.5a-e – 1.6a-e. *Survival of the patients with hAAa was lower (n=26, 61.31 ys) compared to the patients without hAAa (n=6, 71.67 ys); the difference was significant (p< 0.040).*

There was no significant difference between patient cohorts with hAAa and without hAAa in onset (p<0.804 – NS) and duration (p<0.075 – NS) of RA.

Demographics, onset and duration of disease of **RA** patients **with mild** (n=15 of 26) and **severe hAAa** (n=11 of 26) are summarized in Figures 1.7a-c – 1.8a-c

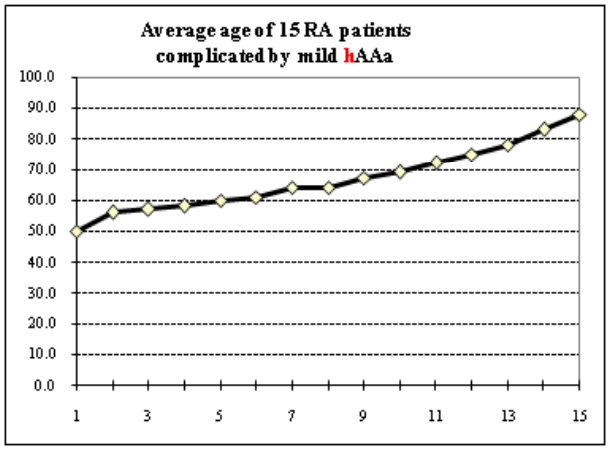

Figure1.7a

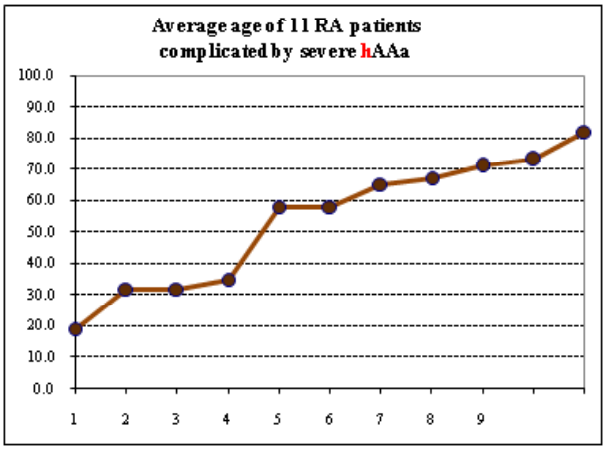

Figure1.8a

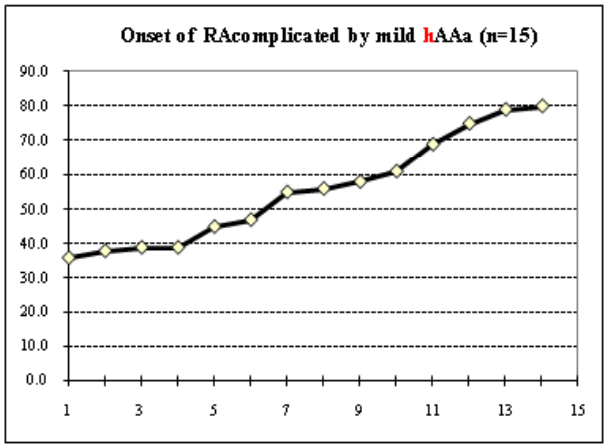

Figure1.7b

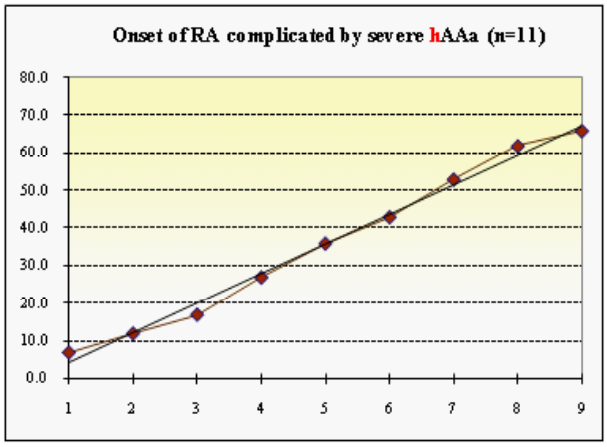

Figure1.8b

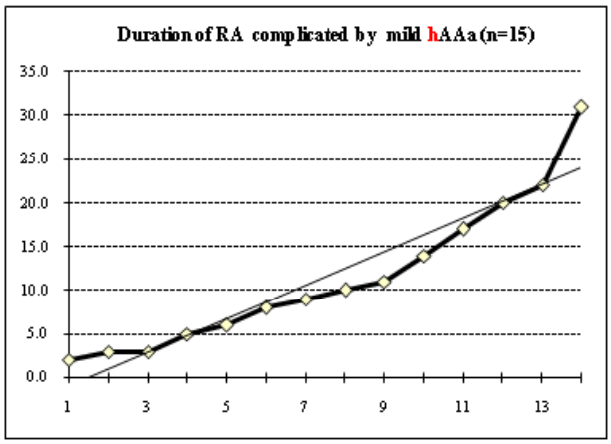

Figure1.7c

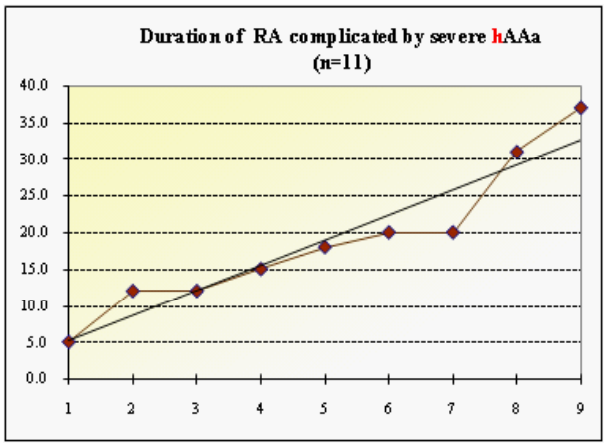

Figure1.8-c

Figure 1.7a-c – 1.8a-c. *RA started earlier (p<0.804 – NS) in patients with severe hAAa (n=11), duration of RA was significantly longer (p<0.035), and the patients died earlier (p<0.079 – NS), compared to the patients with mild hAAa (n=15)*

In Table 2 are summarized the statistical correlations ("p" values of significance) between female and male **RA** patients **with** and **without sAAa** or **hAAa**

Table2. *The statistical correlations ("p" values of significance) between female and male RA patients with or without sAAa and hAAa*

(Extended data published by Miklós Bély and Ágnes Apáthy in: Archives of Gastroenterology and Hepatology, 2020,3.2: 1:26 [6])

RA patients (Liver) n=152	Age	Onset of disease	Disease duration
RA n=152 versus **with sAAa n=32** of 152	**p <0.399**	**p <0.397**	**p <0.407**
Female n=**108** of 152 versus n=**27** of 32	p <0.407	p <0.981	p <0.397
Male n=**44** of 152 versus n=**5** of 32	p <0.397	p <0.482	p <0.981
with sAAa n=32 vs. **without sAAa n=120 of 152**	**p <0.290**	**p <0.284**	**p <0.303**
Female n=**27** of 32 versus n=**81** of 120	p <0.976	p <0.532	p <0.359
Male n=**5** of 32 versus n=**39** of 120	p <0.291	p <0.520	p <0.721
with mild sAAa n=17 vs. **with severe sAAa n=15 of 32**	**p <0.032**	**p <0.095**	**p <0.014**
Female n=**16** of 32 versus n=**11** of 32	p <0.179	p <0.129	**p <0.052**
Male n=**1** of 32 versus n=**4** of 32	-	-	-
with mild sAAa n=17 vs. **without sAAa n=120 of 152**	**p <0.380**	**p <0.894**	**p <0.423**
Female n=**16** of 32 versus n=**81** of 120	p <0.425	p <0.949	p <0.534
Male n=**1** of 32 versus n=**39** of 120	-	-	-
with severe sAAa n=15 vs. **without sAAa n=120 of 152**	**p <0.061**	**p <0.065**	**p <0.025**
Female n=**11** of 32 versus n=**81** of 120	p <0.348	p <0.105	p <0.083
Male n=**n** of 32 versus n=**39** of 120	p <0.150	p <0.357	p <0.271
with sAAa n=32 vs. **with hAAa n=26** of 32	**p <0.655**	**p <0.950**	**p <0.552**
Female n=**27** of 32 versus n=**21** of 26	p <0.578	p <0.989	p <0.552
Male n=**5** of 32 versus n=**5** of 26	p <1.000	p <1.000	p <1.000
with hAAa n=26 vs. **without hAAa n=6 of 32**	**p <0.040**	**p <0.804**	**p <0.075**
Female n=**21** of 26 versus n=**6** of 6	p <0.076	p <0.965	p <0.081
Male n=**5** of 26 versus n=**0** of 6	-	-	-
with mild hAAa n=15 vs. **with severe hAAa n=11 of 26**	**p <0.079**	**p <0.084**	**p <0.035**
Female n=**14** of 15 versus n=**7** of 11	p <0.438	p <0.170	p <0.146
Male n=**1** of 15 versus n=**4** of 11	-	-	-
with mild hAAa n=15 of 26 vs. **without hAAa n=6 of 32**	**p <0.278**	**p <0.379**	**p <0.024**
Female n=14 of 15 versus n=6 of 6	p <0.149	p <0.531	**p <0.028**
Male n=1 of 15 versus n=0 of 6	-	-	-
with severe hAAa n=11 of 26 vs. without hAAa n=6 of 32	**p <0.024**	**p <0.148**	**p <0.496**
Female n=7 of 11 versus n=6 of 6	p <0.104	p <0.311	p <0.723
Male n=4 of 11 versus n=0 of 6	-	-	-

Glossary to Table 2
RA – Rheumatoid **A**rthritis
sAAa – systemic **AA a**myloidosis (amyloid A deposits in the kidneys, heart, pancreas, and lungs)
hAAa – hepatic **AA a**myloidosis (amyloid A deposits in the liver)

The patients **with sAAa** (n=32) were **anemic**, with significantly lower levels of **hemoglobin** (7.311 g/l vs 8.637 g/l; p<**0.011**), and showed **decreased renal function**: they had **higher levels** of blood urea

nitrogen (**BUN**) (17.57 mmol/l vs 9.491 mmol/l; p <**0.021**), serum **creatinine** (182.44 µ mol/l vs 96.373 µ mol/l; p <**0.008**), and lower levels of serum **sodium** (138.27 mmol/l vs 140.62 mmol/l; p <**0.027**), and were accompanied by **proteinuria** (1.821 "+" vs 0.806 "+"; p<**0.003**) compared to the patients **without sAAa** (n=120).

Urine specific gravity (1011.37 g/ml vs 1015.65 g/ml; p<**0.011**), and serum **bilirubin** values (7.435 µmol/l vs 11.041 µmol/l; p<**0.001**) were **lower** in patients **with sAAa** compared to the patients **without sAAa**.

Massive (severe) sAAa (n=15) was accompanied by significantly **lower levels** of **hemoglobin** (6.258 g/l vs 8.15 g/l; p<**0.019**), **higher levels** of serum **creatinine** (252.61 µ mol/l vs 127.31 µ mol/l; p <**0.042**), serum **potassium** (5.35 µmol/l vs 4.50 µmol/l; p<**0.011**), and was accompanied by **proteinuria** (2.833 "+" vs 1.06 "+"; p<**0.002**), compared to the patients **with mild sAAa** (n=17).

Massive (severe) sAAa (n=15) was accompanied by significantly **lower levels** of **hemoglobin** (6.258 g/l vs 8.637 g/l; p<**0.000043**), **higher levels** of blood urea nitrogen (**BUN**) (18.43 mmol/l vs 9.491 mmol/l; p <**0.003**), serum **creatinine** (252.61 µ mol/l vs 96.373 µ mol/l; p <**0.009**), and was accompanied by **proteinuria** (2.833 "+" vs 1.33 "+"; p<**0.000324**), compared to the patients **without sAAa** (n=120).

Urine specific gravity (1011.63 g/ml vs 1015.65 g/ml; NS – p<0.111), and serum **bilirubin** values (7.435 µmol/l vs 11.041 µmol/l; p<**0.001**) were **lower** in patients **with massive (severe)sAAa** compared to the patients **without sAAa**.

There was no significant difference in classic laboratory parameters between cohorts of **RA** patients **with sAAa** and **hAAa**.

The patients **with hAAa** (n=26) had significantly **higher levels** of serum **creatinine** (214.09 µ mol/l vs 82.22 µ mol/l; p <**0.003**), and were accompanied by **proteinuria** (2.09 "+" vs 0.83 "+"; p<**0.049**) compared to the patients **without hAAa** (n=6).

Massive (severe) hAAa (n=11) was accompanied by significantly **lower levels** of **hemoglobin** (5.87 g/l vs 8.12 g/l; p<**0.009**), **higher levels** of serum **potassium** (5.45 m mol/l vs 4.59 mmol/l; p <**0.031**), and was accompanied by **proteinuria** (3.28 "+" vs 1.27 "+"; p<**0.001**), compared to the patients **with mild hAAa** (n=15).

Massive (severe) hAAa (n=11) was accompanied by significantly **higher levels** of blood urea nitrogen (**BUN**) (19.37 mmol/l vs 12.18 mmol/l; p <**0.043**), serum **creatinine** (264.41 µ mol/l vs 82.22 µ mol/l; p <**0.014**), **serum potassium** (5.45 mmol/l vs 4.47 mmol/l; p<**0.031**), and was accompanied by **proteinuria** (3.28 "+" vs 0.83 "+"; p<**0.002**), compared to the patients **without hAAa** (n=6).

The relevant clinical laboratory parameters of **RA** patients **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa** are summarized in Table 3.1 and Figures 3.1-3.2.

Table3.1. *Significantly different or characteristic clinical laboratory parameters of RA patients with and without sAAa or hAAa*

Parameters ± SD	Normal range	With sAAa n=32 of 152	With mild sAAa n=17 of 32	With severe sAAa n=15 of 32	Without sAAa n=120 f 152
Hemoglobin – g/L	14,0-17,5 g/L	7.31 ± 2.19	8.15 ± 2.39	6.26 ± 1.30	8.64 ± 2.53
BUN – mmol/L	2.00-8.90 mmol/L	17.57 ± 16.56	16.8 ± 21.03	18.43 ± 7.90	9.49 ± 6.53
Creatinine – µmol/L	62-106 µmol/L	182.44 ± 144.27	127.31 ± 109.56	252.61 ± 152.44	96.37 ± 49.12
Serum Potassium – mmol/L	3,70-5,10 mmol/L	4.83 ± 0.81	4.50 ± 0.66	5.35 ± 0.74	4.57 ± 3.29
Serum Sodium – mmol/L	135-145 mmol/L	138.27 ± 4.54	138.94 ± 3.93	137.20 ± 5.19	140.62 ± 4.13
Urine specific gravity –g/L	1005-1030 g/L	1011.37 ± 5.69	1011.18± 5.87	1011.63 ± 5.41	1015.65 ± 6.70
Proteinuria – 0-4+	0+	1.82 ± 1.54	1.06 ± 1.21	2.83 ± 1.33	0.81 ± 0.92
Serum bilirubin – µmol/L	5,0-17,1 µmol/L	7.44±3.82	8.13 ± 4.15	6.63±3.22	11.04 ± 6.25

Parameters ± SD	Normal range	With hAAa n=26 of 32	With mild hAAa n=15 of 26	With severe hAA n=11 of 26	Without hAAa n=6 of 32
Hemoglobin – g/L	14,0-17,5 g/L	7.15 ± 2.12	8.12 ± 2.14	5.87 ± 1.22	8.87 ± 2.36
BUN – mmol/L	2.00-8.90 mmol/L	19.11 ± 18.29	18.92 ± 23.56	19.37 ± 6.34	12.18 ± 5.06
Creatinine – μ mol/L	62-106 μmol/L	214.09 ± 150.54	168.80 ± 119.86	264.41 ± 164.53	82.22 ± 41.81
Serum Potassium – mmol/L	3,70-5,10 mmol/L	4.94 ± 0.83	4.59 ± 0.69	5.45 ± 0.78	4.47 ± 0.61
Serum Sodium – mmol/L	135-145 mmol/L	137.90 ± 4.53	138.67 ± 3.33	136.75 ± 5.70	139.50 ± 4.35
Urine specific gravity –g/L	1005-1030 g/L	1011.38 ± 5.27	1011.27± 5.5	1011.60 ± 4.84	1011.33 ± 7.54
Proteinuria – 0-4+	0+	2.09 ± 1.54	1.27 ± 1.25	3.28 ± 1.08	0.83 ± 1.03
Serum bilirubin – μmol/L	5,0-17,1 μmol/L	7.30±3.73	7.59 ± 3.79	6.93±3.62	7.90 ± 4.10

Glossary to Table 3.1

sAAa – systemic **AA a**myloidosis (amyloid A deposits in the kidneys, heart, pancreas and lungs)

hAAa – hepatic **AA a**myloidosis (amyloid A deposits in the liver)

SD – Standard **D**eviation

Figure3.1. Significantly different or characteristic clinical laboratory parameters of RA patients with mild or severe sAAa and without sAAa

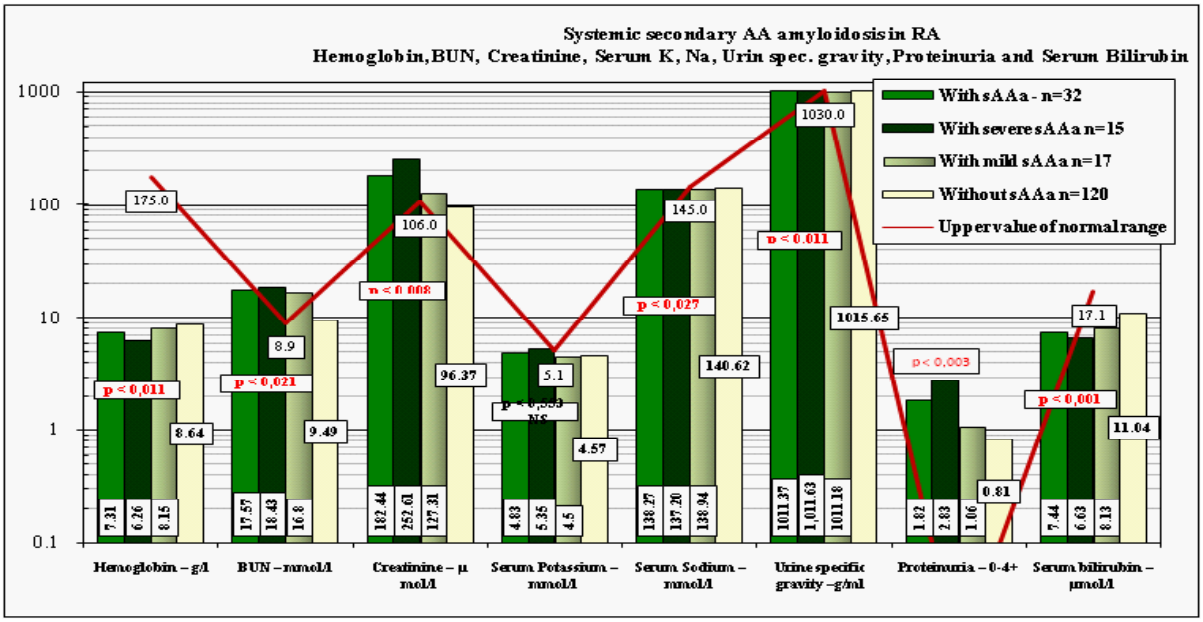

Figure 3.1*. *There was a significant relationship between RA patients with mild or severe sAAa and without sAAa: the RA patients with sAAa had significantly lower levels of hemoglobin (7.311 g/l vs 8.637 g/l; p<0.011), and showed decreased renal function: they had higher levels of blood urea nitrogen (BUN) (17.57 mmol/l vs 9.491 mmol/l; p <0.021), serum creatinine (182.44 μ mol/l vs 96.373 μ mol/l; p <0.008), and lower levels of serum sodium (138.27 mmol/l vs 140.62 mmol/l; p <0.027), serum bilirubin (7.435 μmol/l vs 11.041 μmol/l; p<0.001), and were accompanied by proteinurea (1.821 "+" vs 0.806 "+"; p<0.003) compared to the patients without sAAa.*

**Figure 3.1 indicates only the "p" values between sAAa and without sAAa.*

Figure3.2. Significantly different or characteristic clinical laboratory parameters of RA patients with mild or severe hAAa and without hAAa

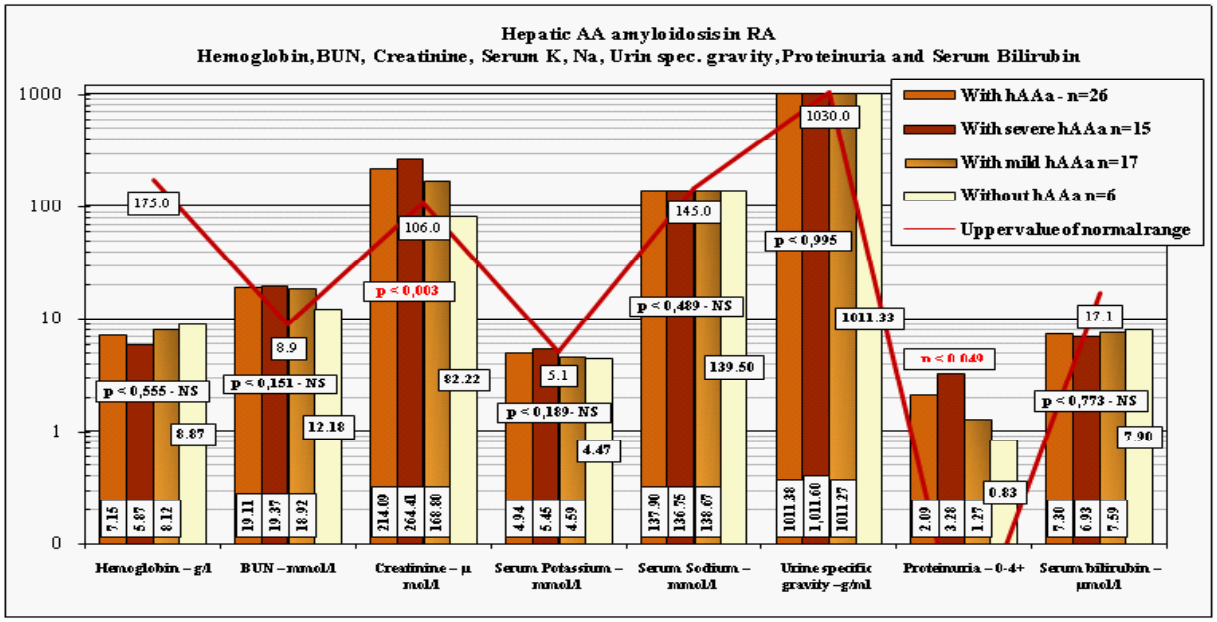

Figure 3.2. There was no significant difference in clinical laboratory parameters between RA patients with (n=26) and without hAAa (n=6), except serum creatinine level (214.09 μ mol/l vs 82.22 μ mol/l; p <0.003), and proteinurea (2.09 "+" vs 0.83 "+"; p<0.049).*

** Figure 3.2 indicates only the "p" values between hAAa and without hAAa.*

Table 3.2 summarizes the statistical correlations ("p" values of significance) of **RA** patient cohorts **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa**.

Table3.2. *The statistical correlations ("p" values of significance) between RA patients with and without sAAa or hAAa*

Relationship between laborytory parameters – p <0.05 (marked **red)**	RBC	Hemoglobin	BUN	Creatinine	Serum Potassium	Serum Sodium	Urine specific gravity	Proteinuria	Serum bilirubin
With sAAa (n=**32** of 152) versus **without sAAa** (n=**120** of 152)	**0,100**	**0,011**	**0,021**	**0,008**	**0,533**	**0,027**	**0,011**	**0,003**	**0,001**
With mild sAAa (n=**17** of 32) versus **with severe sAAa** (n=**15** of 32)	**0,808**	**0,019**	**0,802**	**0,042**	**0,011**	**0,400**	**0,875**	**0,002**	**0,328**
With mild sAAa (n=**17** of 32) versus **without sAAa** (n=**120**)	**0,176**	**0,492**	**0,212**	**0,336**	**0,859**	**0,146**	**0,046**	**0,446**	**0,040**
With severe sAAa (n=**15** of 32) versus **without sAAa** (n=**120**)	**0,265**	**0,000043**	**0,003**	**0,009**	**0,087**	**0,084**	**0,101**	**0,00032**	**0,001**
With sAAa (n=**32** of 152) versus **with hAAa** (n=**26** of 32)	**0,816**	**0,805**	**0,770**	**0,497**	**0,666**	**0,790**	**0,997**	**0,550**	**0,904**
With hAAa (n=**26** of 32) versus **without hAAa** (n=**6** of 32)	**0,637**	**0,555**	**0,151**	**0,003**	**0,189**	**0,489**	**0,995**	**0,049**	**0,773**
With mild hAAa (n=**17** of 26) versus **with severe sAAa** (n=**11** of 26)	**0,507**	**0,009**	**0,953**	**0,196**	**0,031**	**0,438**	**0,915**	**0,001**	**0,712**
With mild hAAa (n=**17** of 26) versus **without hAAa** (n=**6**)	**0,598**	**0,844**	**0,383**	**0,073**	**0,719**	**0,714**	**0,992**	**0,471**	**0,891**
With severe hAAa (n=11 of 26**) versus without hAAa** (n=**6**)	**0,947**	**0,124**	**0,043**	**0,014**	**0,031**	**0,362**	**0,967**	**0,002**	**0,675**

The hepatic amyloid A deposits (especial the massive **hAAa**) was accompanied by significantly **increased erythrocyte sedimentation rate** (81.227-101.0 mm/h versus 48.667 mm/h; p<**0.003**), and higher levels of **alpha2-globulin** (15.194-15.714 % versus 12.90 %; p < **0.048**) in comparison of **RA** patients **without hAAa** (Tables 4.1-4.2).

The inflammatory laboratory parameters of **RA** patient cohorts **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa** are summarized in Table 4.1.

Table4.1. *Significantly different or characteristic inflammatory laboratory parameters of RA patients with and without sAAa or hAAa*

Parameters ± SD	Normal range	With sAAa - n=32	With severe sAAa n=15	With mild sAAa n=17	Without sAAa n=120
ESR – mm/h	≤ 15-20 mm/h	74.25 ± 34.279	86.167 ± 40.214	65.313 ± 25.634	79.143 ± 36.095
CRP – mg/L	0.0-5.0 mg/L	334.55 ± 434.79	291.56 ± 316.22	364.61 ± 498.52	517.00 ± 1002.55
Albumine – g/L	35-50 g/L	28.556 ± 7.79	29.700 ± 9.133	27.125 ± 5.349	31.222 ± 6.259
Globuline – g/L	25-35 g/L	36.444 ± 5.871	37.300 ± 6.357	35.375 ± 4.998	35.185 ± 6.392
Albumin/Globulin ratio	<1	0.784 ± 1.327	0.730 ± 0.52	0.133 ± 0.192	0.98 ± 0.133
Albumine – %	59.8-72.4%	42.783 ± 6.723	41.678 ± 7.299	43.447 ± 6.259	45.701 ± 6.019
Alpha 1 globulin – %	1.0-3.2 %	5.333 ± 1.491	4.944 ± 1.681	5.567 ± 1.310	5.452 ± 1.639
Alpha 2 globulin – %	7,4-12.6 %	14.621 ± 3.707	15.378 ± 5.215	14.167 ± 2.261	13.035 ± 3.017
Beta globulin – %	7.5-12.9%	13.804 ± 2.357	13.289 ± 2.495	14.113 ± 2.214	13.496 ± 2.426
Gamma globulin – %	8.0-15.8%	23.438 ± 5.823	24.378 ± 5.293	22.873 ± 6.049	21.594 ± 5.542
Parameters ± SD	Normal range	With hAAa - n=26	With severe hAAa n=11	With mild hAAa n=15	Without hAAa n=6
ESR – mm/h	≤ 15-20 mm/h	81.227 ± 34.931	101.000 ± 34.590	67.538 ± 27.903	48.667 ± 13.287
CRP – mg/L	0.0-5.0 mg/L	362.667 ± 468.73	333.714 ± 342.033	381.091 ± 533.102	208.00 ± 177.449
Albumine – g/L	35-50 g/L	28.857 ± 8.676	26.429 ± 8.415	31.286 ± 8.241	27.500 ± 2.872
Globuline – g/L	25-35 g/L	37.071 ± 6.364	36.143 ± 5.844	38.000 ± 6.719	34.250 ± 2.681
Albumin/Globulin ratio	<1	0.78 ± 1.363	0.731 ± 1.441	0.823 ± 1.227	0.803 ± 1.071
Albumine – %	59.8-72.4%	42.033 ± 6.961	41.329 ± 8.166	42.482 ± 6.029	45.033 ± 5.352
Alpha 1 globulin – %	1.0-3.2 %	5.528 ± 1.508	4.643 ± 1.715	6.091 ± 1.018	4.750 ± 1.270
Alpha 2 globulin – %	7,4-12.6 %	15.194 ± 4.08	15.714 ± 5.772	14.864 ± 2.399	12.900 ± 1.036
Beta globulin – %	7.5-12.9%	13.767 ± 2.40	13.186 ± 2.810	14.136 ± 2.013	13.917 ± 2.219
Gamma globulin – %	8.0-15.8%	23.450 ± 6.125	24.900 ± 5.872	22.527 ± 6.103	23.400 ± 4.807

Table 4.2 summarizes the statistical correlations ("p" values of significance) of **RA** patient cohorts **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa**.

Table4.2. *The "p" values of significance of inflammatory parameters between RA patients with and without sAAa or hAAa*

Relationship between laborytory parameters – p <0.05 (marked **red**)	ESR	CRPn	Albumin	Globulin	Albumin %	Alfa 1 globulin %	Alfa 2 globulin %	Beta globulin %	Gamma globulin %
With sAAa (n=**32** of 152) versus **without sAAa** (n=**120** of 152)	0,520	0,247	0,211	0,457	0,069	0,745	0,069	0,588	0,186
With mild sAAa (n=**17** of 32) versus **with severe sAAa** (n=**15** of 32)	0,149	0,694	0,492	0,507	0,574	0,383	0,547	0,450	0,550
With mild sAAa (n=**17** of 32) versus **without sAAa** (n=**120**)	0,079	0,428	0,093	0,929	0,227	0,775	0,116	0,355	0,470
With severe sAAa (n=**15** of 32) versus **without sAAa** (n=**120**)	0,589	0,187	0,640	0,374	0,165	0,435	0,245	0,827	0,189
With sAAa (n=**32** of 152) versus **with hAAa** (n=**26** of 32)	0,492	0,850	0,922	0,784	0,735	0,688	0,651	0,961	0,995
With hAAa (n=**26** of 32) versus **without hAAa** (n=**6** of 32)	0,003	0,332	0,649	0,254	0,329	0,278	0,048	0,899	0,985
With mild hAAa (n=**17** of 26) versus **with severe sAAa** (n=**11** of 26)	0,038	0,831	0,332	0,619	0,770	0,095	0,741	0,486	0,455
With mild hAAa (n=**17** of 26) versus **without hAAa** (n=**6**)	0,077	0,396	0,341	0,265	0,422	0,073	0,043	0,856	0,768
With severe hAAa (n=11 of 26**) versus without hAAa** (n=**6**)	0,003	0,486	0,786	0,522	0,387	0,908	0,283	0,639	0,650

Characteristic liver enzymes showed no significant difference between **RA** patients **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa** (Tables 5.1-5.2).

Table 5.1 summarizes the level of liver enzymes of **RA** patients **with mild** or **severe sAAa** and **without sAAa**, furthermore **with mild** or **severe hAAa** and **without hAAa**.

Table5.1. *Level of liver enzymes of RA patients with and without sAAa or hAAa*

Parameters ± SD	Normal range	With sAAa - n=32	With severe sAAa n=15	With mild sAAa n=17	Without sAAa n=120
LDH U/L	0-480 U/L	281.37 ± 207.25	282.40 ± 203.43	280.86 ± 209.13	338.69 ± 197.36
GPT U/L	5-41 U/L	59.38 ± 180.11	125.86 ± 283.02	20.61 ± 23.63	21.17 ± 17.80
GGT U/L	11-50 U/L	50.438 ± 42.642	45.20 ± 25.388	52.818 ± 48.309	63.955 ± 61.066
Parameters ± SD	Normal range	With hAAa - n=26	With severe hAAa n=11	With mild hAAa n=15	Without hAAa n=6
LDH U/L	0-480 U/L	278.20 ± 155.619	194.333 ± 28.335	314.143 ± 173.048	287.72 ± 283.474
GPT U/L	5-41 U/L	73.929 ± 207.78	11.20 ± 5.492	108.778 ± 252.468	18.66 ± 11.796
GGT U/L	11-50 U/L	51.909 ± 46.756	50.00 ± 30.822	52.625 ± 51.456	47.20 ± 31.53

Table5.2. *The "p" values of significance of liver enzymes between RA patients with and without sAAa or hAAa*

Relationship between liver enzymes – p <0.05	LDH	GPT	GGT
With sAAa (n=**32** of 152) versus **without sAAa** (n=**120** of 152)	0,386	0,381	0,355
With mild sAAa (n=**17** of 32) versus **with severe sAAa** (n=**15** of 32)	0,99	0,398	0,707
With mild sAAa (n=**17** of 32) versus **without sAAa** (n=**120**)	0,468	0,942	0,541
With severe sAAa (n=**15** of 32) versus **without sAAa** (n=**120**)	0,622	0,4	0,263
With sAAa (n=**32** of 152) versus **with hAAa** (n=**26** of 32)	0,967	0,841	0,937
With hAAa (n=**26** of 32) versus **without hAAa** (n=**6** of 32)	0,952	0,357	0,832
With mild hAAa (n=**17** of 26) versus **with severe sAAa** (n=**11** of 26)	0,148	0,306	0,932
With mild hAAa (n=**17** of 26) versus **without hAAa** (n=**6**)	0,873	0,343	0,832
With severe hAAa (n=11 of 26**) versus without hAAa** (n=**6**)	0,548	0,298	0,922

Figures 3-5 illustrate Amyloid A deposition in the kidneys representing different stages of **sAAa**.

The original magnification (O) of electron microphotographs (elements of montage) correspond to the 60x90 mm negatives

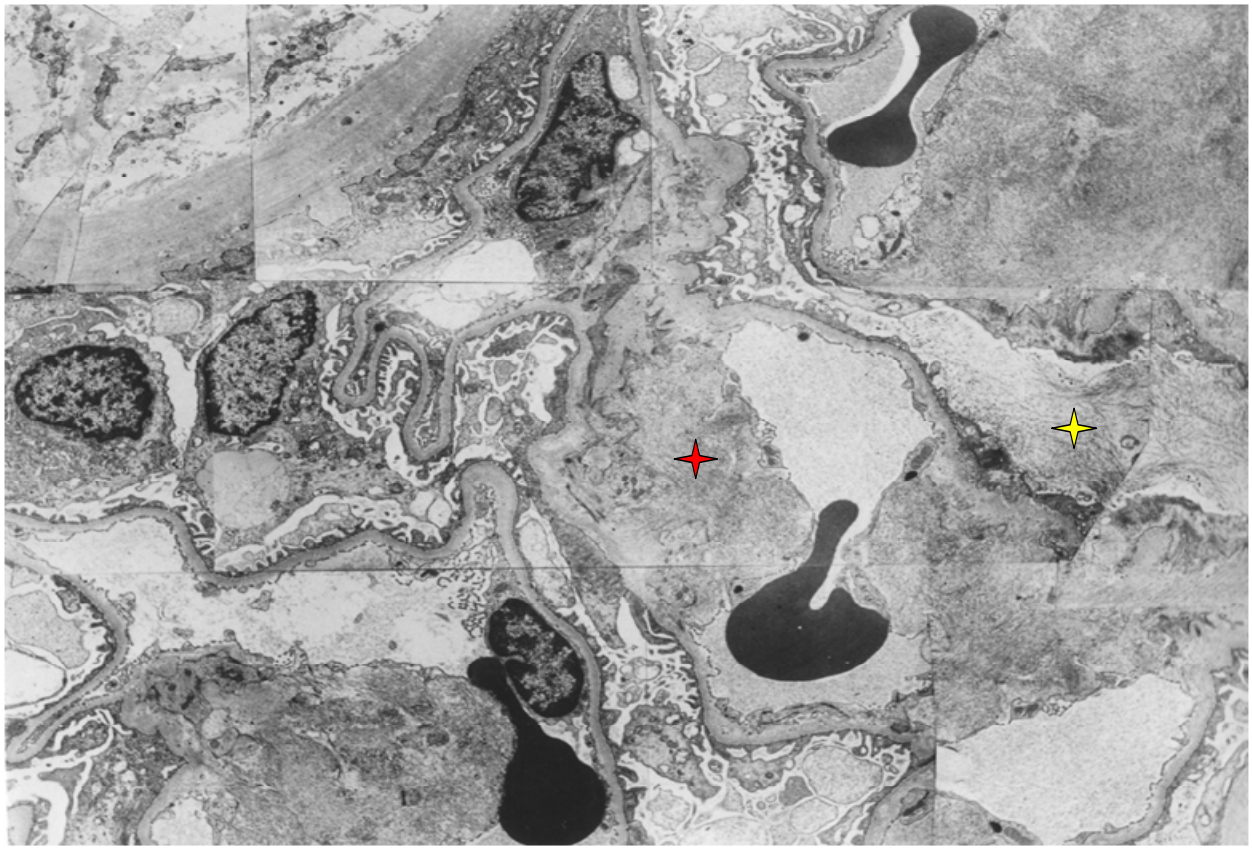

Figure3. *Electron micrograph (montage)*
Rheumatoid arthritis, kidney, early stage of amyloid A deposition
Moderate subendothelial (red star), and mesangial (yellow star) amyloid deposits, damaged endothelial cells. Early, advanced and late stages (see next Figure 4 and Figure 5) of amyloid deposits side by side in different glomeruli in the same kidney demonstrate the progressive accumulation of amyloid deposits,
O: x2600.
CL – capillary lumen

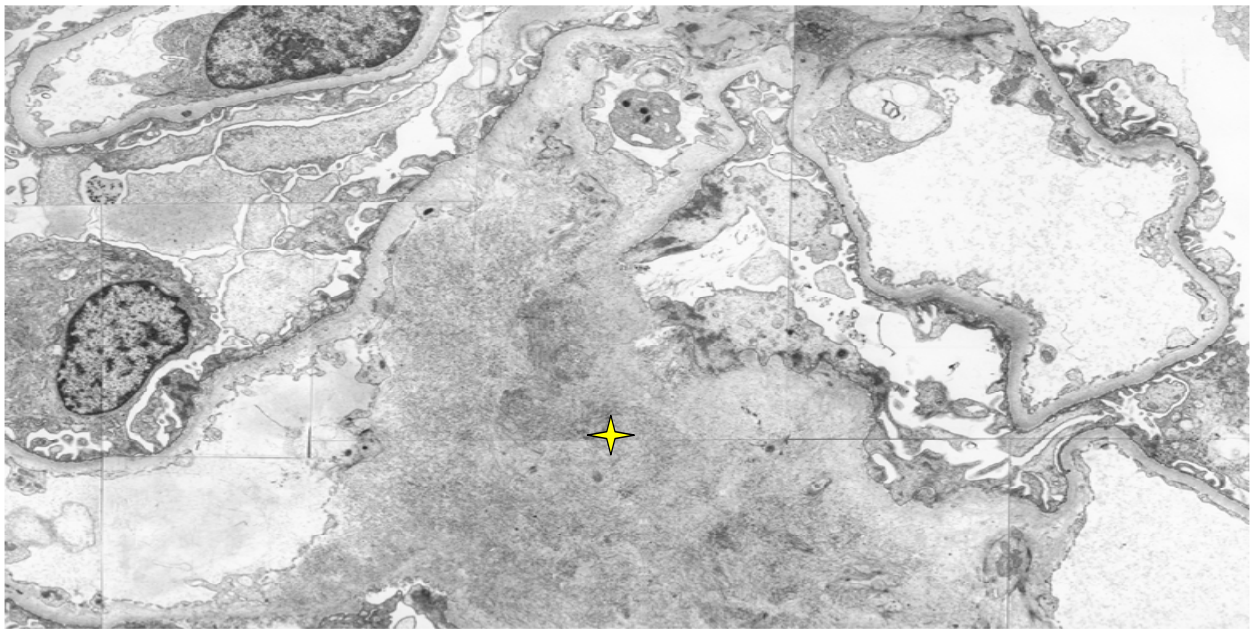

Figure4. *Electron micrograph (montage)*
Rheumatoid arthritis, kidney, advanced stage of amyloid A deposition
Moderate subendothelial and massive mesangial amyloid deposits of different density (yellow star), damaged endothelial cells, and podocytes, side by side in the same glomerulus demonstrate the progressive accumulation of amyloid deposits,
O: x2600.

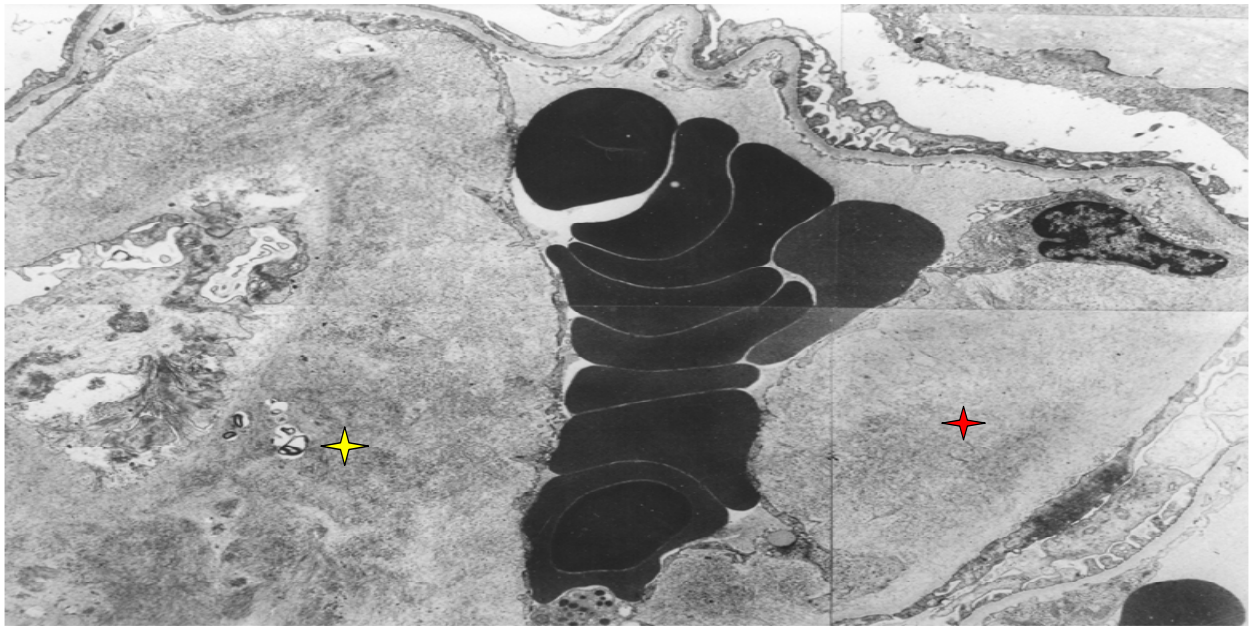

Figure5. *Electron micrograph (montage)*
Rheumatoid arthritis, kidney, late stage of amyloid A deposition, with massive subendothelial (red star) and mesangial (yellow star) amyloid deposits
The capillary is lined by damaged endothelial cells and the podocytes are degenerated.
Early, advanced and late stages of amyloid deposits side by side in different glomeruli in the same kidney demonstrate the progressive accumulation of amyloid deposits,
O: x2600.

Figures 6 and 7 demonstrate the extent of amyloid A deposits in the liver by traditional HE and Congo red staining, viewed by light microscopy and under polarized light respectively.

Original magnifications correspond to the 24x36 mm transparency slide; the correct height: width ratio is 3:2. The printed size may be different; therefore it is necessary to indicate the original magnifications.

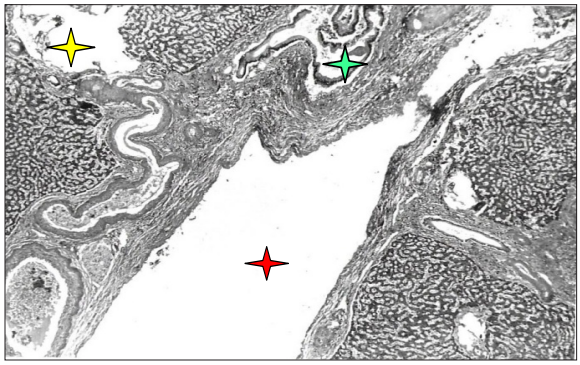

Figure6a

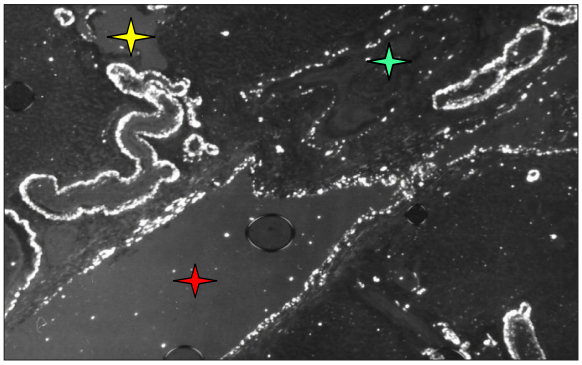

Figure7a

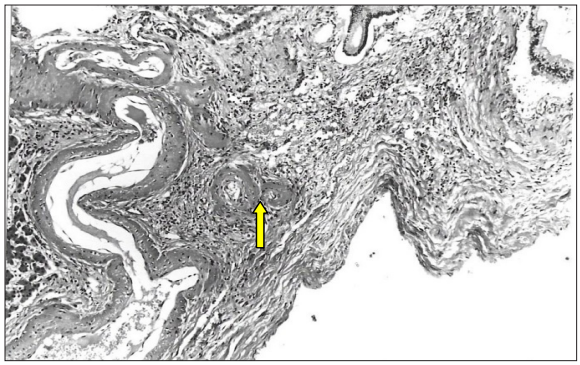

Figure6b

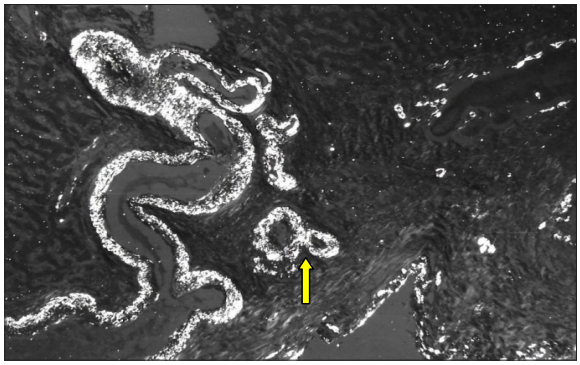

Figure7b

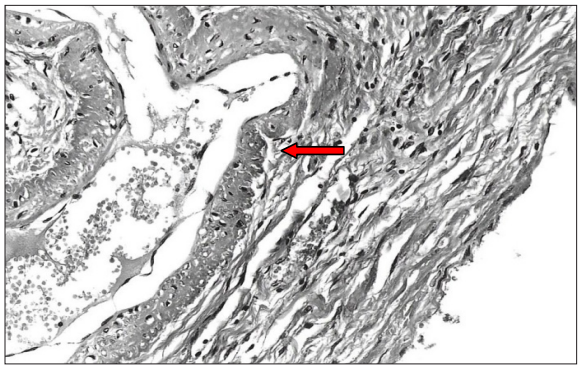

Figure6c

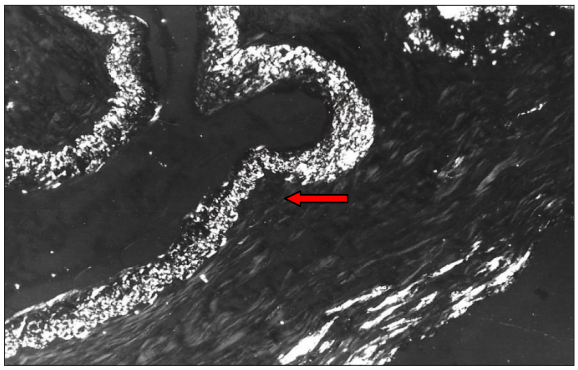

Figure7c

Figure6a-c. *Rheumatoid arthritis, liver, advanced stage of hAAa*

Arteriole, small artery (arrowhead yellow), medium size artery (arrowhead red) and small (star yellow) and medium size (star red) veins, amyloid A deposits are within the walls of blood vessels

Collecting biliary duct with subendothelial amyloid A deposits in basal lamina

(a) HE traditional staining and ligh microscopy, x50, (b) same as (6a) x125, (c) same as (6a) x200

Figure7a-c. *Rheumatoid arthritis, liver, advanced stage of hAAa*

Amyloids A deposits within the wall of blood vessels, and in basal lamina of collecting biliary duct are birefringent

(a) same as (6a) Congo red staining, without alcoholic differentiation, covered with gum Arabic, viewed under polarized light, x50, (b) same as (6b) x125, (c) same as (6c) x200

Discussion

In our patient cohorts there was **no significant difference** in **survival time**, **onset**, and **duration** of **disease**, between **RA** patient cohorts **with sAAa** and **without sAAa**, **with sAAa** and **with hAAa**, neither in **females** nor in **males**. ***Amyloidosis may develop in both sexes and at any time of the disease*** [6].

Diagnostic values of discussed laboratory parameters are limited. No blood test is specific for amyloidosis [11].

In some cases the values of clinical-laboratory parameters mentioned in the pertinent literature remained in the normal range, for example serum albumin (35-50 g/l), total serum bilirubin (5.0-17.1 μmol/l), serum potassium (3.70-5.10 mmol/L), sodium (135-145 mmol/lL, urine specific gravity between 1000 and 1030), etc., which reduce the clinical significance of these parameters, and only their graduall impairment may suggest a trend.

There was no significant difference in classic laboratory parameters between cohorts of **RA** patients **with sAAa** and **hAAa**, including the inflammatory laboratory parameters, and the characteristic liver enzymes.

The more or less significant differences between **RA** patients **with** and **without sAAa** are connected with **decreased renal function**; the **higher** levels of blood urea nitrogen (**BUN**) (17.57 mmol/L vs 9.49 mmol/L, p < **0.021**), serum **creatinine** (182.44 μmol/L vs 96.37 μmol/L vs, p < **0.008**), serum **potassium** (4.83 mmol/L vs 4.57 mmol/L, p < 0.533 – NS), and **proteinuria** (1.82 "+" vs 0.81 "+", p<**0.003**), with **lower** levels serum **sodium** (138.27 mmol/L vs 140.62 mmol/L, p <**0.027**), and **urine specific gravity**, namely the less concentrated urine (1011.37 g/L vs 1015.65 g/L, p <**0.001**) show the impaired function of the kidneys only, like other causes of nephrotic syndrome. ***The mentioned clinical-laboratory parameters are not specific for* sAAa**, and ***they imply more or less only renal impairment***.

Progression of renal amyloidosis is associated with severe proteinuria or nephrotic syndrome [12]. Indeed, several authors call attention the importance of proteinuria in the early literature [13, 14, 15].

The rate of **proteinuria** in our autopsy population proved to be of prognostic value, and may indicate the severity of **sAAa**; the rate of proteinuria correlated with the severity of **sAAa** (**severe** vs **mild sAAa**: 2.83 "+" vs 1.06 "+"; p<**0.002**; **severe sAAa** vs **without sAAa**: 2.83 "+" vs 0.81 "+"; p<**0.00032**), based on the increasing levels of significance. The elevated serum **creatinine** level may also indicate progression of renal amyloidosis (**severe** vs **mild sAAa**: 252.61 μmol/L vs 127.31 μmol/L, p<**0.042**; **severe** vs **without sAAa**: 252.61 μmol/L vs 96.37 μmol/L, p<**0.009**).

The differences were marked in **hemoglobin** levels (**severe** vs **mild sAAa**: 6.26 g/L vs 8.15 g/L, p <**0.019**; **severe** vs **without sAAa**: 6.26 g/L vs 8.64 g/L, p <**0.000043**), and in serum **bilirubin** values as well (**severe** vs **mild sAAa**: 6.63 μmol/L vs 8.13 μmol/L, p<0.328; **severe** vs **without sAAa**: 6.63 μmol/L vs 11.04 μmol/L, p<**0.001**).

The low levels of **hemoglobin** (7.31 g/L vs 8.64 g/L, p <**0.01**), and the subsequent serum **bilirubin** values (7.44 μmol/L vs 11.04 μmol/L vs, p <**0.001**) may underline the existence of **sAAa**, but likewise are not specific for amyloidosis.

Anemia may be regarded as a kind of non-hemolytic anemia only [16] accompanied by lower levels of serum **bilirubin** (degradation product of hemoglobin).

Increased **ESR** (81.227 mm/h vs 48.667 mm/h, p <**0.003**), and higher levels of **alpha2-globulin** (15.19 % vs 12.90 %, p <**0.048**), may indicate the existence of **hAAa**, in comparison to **RA** patients **without hAAa**.

Erythrocyte sedimentation rate may increase in any inflammation. High values of **alpha2-globulin** are related to the basic inflammatory processes of the disease, and not to amyloid A deposition [17].

More recent laboratory parameters such as serum amyloid A (**SAA**) level were not determined between 1969 and 1992. **SAA** and **CRP** are regarded now the most sensitive indicators for assessing inflammatory activity [18]. **SAA** proteins are produced predominantly by the liver [19]. **SAA** is implicated in several chronic inflammatory diseases, such as rheumatoid arthritis, amyloidosis, atherosclerosis etc. [20]. Prolonged elevation of **SAA** is not specific for amyloidosis, and does not necessarily indicate tissue deposition of amyloid A [17].

Radioactive imaging, using radiolabeled amyloid molecules, i.e. serum amyloid P component (**SAP**)

scintigraphy [21] or positron emission tomography (**PET**) [22] has not yet fullfiled the hopes [23].

For exact diagnosis of amyloidosis biopsy is needed using an „appropriate staining procedure" [24]. We suggest gingival or rectal biopsies in any suspected cases, stained with Congo red according to Romhányi, using an appropriate polarizing microscope with high brightness. In case of increased **ESR**, the possibility of **hAAa** arises, and especially in hepatomegaly, liver biopsy may be considered to confirm **hAAa**.

Conclusions

Amyloidosis may develop in both sexes and at any time of the disease.

The diagnostic values of the discussed laboratory parameters are limited, and none are specific for amyloidosis

The more or less significant differences between **RA** patients **with** and **without sAAa** show the impaired function of the kidneys or are connected to the basic disease only.

There is no significant difference in classic laboratory parameters between cohorts of **RA** patients **with sAAa** and **hAAa**.

For exact diagnosis of amyloidosis a biopsy is needed using an „appropriate staining procedure". Gingival or rectal biopsies are suggested [3]. In case of increased **ESR**, the possibility of **hAAa** arises, and especially in hepatomegaly a liver biopsy may be considered to confirm **hAAa**.

References

[1] **Sipe JD, Benson MD, Buxbaum JN, Ikeda Shu-ichi, Merlini G, Saraiva MJM, Westermark P**: "Amyloid fibril proteins and amyloidosis: chemical identification and clinical classification International Society of Amyloidosis 2016 Nomenclature Guidelines" *Amyloid*, 2016; 23.4: 209–213. https://doi.org/10.1080/13506129.2016.1257986

[2] **Benson MD, Buxbaum JN, Eisenberg DS, Merlini G, Saraiva MJM, Sekijima Y, Sipe JD, Westermark P**: "Amyloid nomenclature 2018: recommendations by the International Society of Amyloidosis (ISA) nomenclature committee" *Amyloid*, 2019; 26:1-5. https://doi.org/10.1080/13506129.2018.1549825

[3] **Bély M**: „Sekundäre Amyloidose bei chronischer Polyarthritis" *Zentralblatt für allgemeine Pathologie und pathologische Anatomie*, 1990, 136:337-357.

[4] **Bély M, Apáthy Á**: "Histochemical and immunohistichemical differential diagnosis of amyloidosis - a brief illustrated essay and personal experience with Romhányi's method" *Amyloid*, 2000; 7:212-217

[5] **Bély M**: "Identification of Amyloid Deposits by Histochemical Methods of Romhányi" *Amyloid* (Guest Editor: M Bély), 2001; 8.2:177-182.

[6] **Bély M, Apáthy Á**: "AA amyloidosis of the liver in rheumatoid arthritis – A postmortem clinicopathologic study of 152 autopsy patients" *Archives of Gastroenterology and Hepatology*, 2020; 3.2: 1-26.

[7] **Arnett FC, Edworthy SM, Bloch DA, McShane DJ, Fries JF, Cooper NS, Healey LA, Kaplan SR, Liang MH, Luthra HS, Medsger TA, Mitchell DM, Neustadt DH, Pinals RS, Schaller JG, Sharp JT, Wilder RL, Hunder GG**: "The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis". *Arthritis and Rheumatism*, 1988; 31: 315-324. DOI: 10.1002/art.1780310302

[8] **Romhányi G**: "Selective differentiation between amyloid and connective tissue structures based on the collagen specific topo-optical staining reaction with Congo red". *Virchows Archive*, 1971; 354:209-222.

[9] **Bély M, Makovitzky J**: "Sensitivity and Specificity of Congo red Staining According to Romhányi - Comparison with Puchtler's or Bennhold's Methods". *Acta Histochemica*, 2006; 108:175-180.

[10] **Lentner C**: "Statistical methods" In *Geigy scientific tables*, 8th revised and enlarged ed: Ciba-Geigy Limited, Basle, Switzerland, Editor: Lentner C, Compiled by: Diem K, Seldrup J, 1982, volume 2, p: 227.

[11] **Kuroda T, Wada Y, Nakano M**: "Diagnosis and treatment of AA amyloidosis with rheumatoid

arthritis: state of the art" Chapter 9 In *Amyloidosis*. (Ed: Feng D) Educational Technology Research and Development, 1999; 47:43–62. http://dx.doi.org/10.5772/54332

[12] Sasatomi Y, Kiyoshi Y, Uesugi N, Hisano S, Takebayashi S: "Prognosis of Renal Amyloidosis: A Clinicopathological Study Using Cluster Analysis". *Nephron*, 2001; 87:42-49. https://doi.org/10.1159/000045883

[13] Husby G. "Amyloidosis and rheumatoid arthritis". *Clinical and Experimental Rheumatology*, 1985; 3: 173-180.

[14] Gertz MA and Kyle RA. „Secondary systemic amyloidosis: response ans survival in 64 patients". *Medicine*, 1991; 70: 246-254.

[15] Schneider F. „Die AA-Amyloidose bei entzündlich-rheumatischen Erkrankungen". *Zeitschrift für Rheumatologie*, 1992; 51: 177-182.

[16] **Shcherbinina MB**: "Low blood bilirubin level: possible diagnostic and prognostic importance". *Klinicheskaia Meditsina* (Mosk) [Article in Russian], 2007; 85 (10):10-4. PMID: 18154172

[17] Dhawan R. AA, Ahmed MM, Mubashir E, Buxbaum JN, Kaur RJ. „(inflammatory) amyloidosis workup". *Medscape reference* Updated: Dec 06, 2015 Availablee from: http://emedicine.medscape.com/article/335559-workup

[18] **Yamada T.** "Serum amyloid A (SAA): a concise review of biology, assay methods and clinical usefulness". *Clinical Chemistry and Laboratory Medicine*, 1999; 37: 381-388.

[19] **Uhlar CM and Whitehead AS.** "Serum amyloid A, the major vertebrate acute-phase reactant". *European Journal of Biochemistry*, 1999; 265.2: 501–23. PMID: 10504381, doi: 10.1046/j.1432-1327.1999.00657.x

[20] **Zhang N et al.** "Serum amyloid A-luciferase transgenic mice: response to sepsis, acute arthritis, and contact hypersensitivity and the effects of proteasome inhibition". *Journal of Immunology*, 2005; 174.12: 8125–34. PMID: 15944321, doi:10.4049/jimmunol.174.12.8125.

[21] **Hawkins PN.** "10 years experience with serum amyloid P component scintigrapy". Book of Anstracts, The IXth International Symposium on Amyloidosis, Budapest Hungary, July 15-21, 2001. *Amyloid: The Journal of Protein Folding Disorders* (Guest Editor: M Bély), 8(Suppl 2): 166-168.

[22] Spitz J and Freudenberg LS. „The impact of nuclear medicine procedures on the diagnosis and follow-up of amyloidosis". Book of Anstracts, The IXth International Symposium on Amyloidosis, Budapest Hungary, July 15-21, 2001. *Amyloid: The Journal of Protein Folding Disorders*. (Guest Editor: M Bély), 8(Suppl 2): 170-174.

[23] **Linke RP.** "Diagnosis of the various amyloid diseases and new developments". Book of Anstracts, The IXth International Symposium on Amyloidosis, Budapest Hungary, July 15-21, 2001. *Amyloid: The Journal of Protein Folding Disorders* (Guest Editor: M Bély), 2001; 8(Suppl 2): 153-155.

[24] **Cohen AS.** "Amyloidosis associated with rheumatoid arthritis". *The Medical Clinics of North Amerika*, 1968; 52: 643-653.

Citation: **Miklós Bély, Ágnes Apáthy.** *Demographics and Predictive Clinical-Laboratory Parameters of Systemic and Hepatic AA Amyloidosis – A Postmortem Clinicopathologic Study of 152 Rheumatoid Arthritis Patients. Archives of Gastroenterology and Hepatology. 2020; 3(2): 64-83.*