Supplementary material

Long-term Outcomes of Adult Pulmonary Langerhans Cell Histiocytosis: A Prospective Cohort

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Supplementary methods

Study design and patient evaluation during the study

The following items were recorded for the study: demographics, smoking status, cannabis consumption, clinical symptoms and signs, Langerhans cell histiocytosis (LCH) localizations, high-resolution computed tomography (HRCT) findings, lung function tests, oxygen saturation and/or blood gas analyses, long-term oxygen supplementation, Doppler echocardiography and right heart catheterization (RHC) results, systemic treatments received for LCH, the presence of malignancies, the performance of lung transplantation and survival status (deceased or alive).

For patients whose last visit was prior to the end of the study, the vital status (alive vs. deceased) was assessed through a telephone call to the patient or the relevant general practitioner. Additionally, if needed, a query was sent to the city hall (registry of births and deaths) of the town in which the patient was born.

At the diagnosis of pulmonary Langerhans cell histiocytosis (PLCH), a thorough history was collected and a comprehensive physical examination including ENT and stomatology was systematically performed to detect extra-pulmonary LCH involvement. Apart from routine blood analyses (complete blood count, blood chemistry analysis with total protein, electrolyte, creatinine, bilirubin, alanine aminotransferase, aspartic aminotransferase, alkaline phosphatase, γ-glutamyl transpeptidase, C-reactive protein and fibrinogen levels and protein electrophoresis), specific investigations to evaluate extrathoracic LCH involvement were performed in individual cases based on clinical or biological suspicion [1, 2].

The differential diagnoses considered vary according to the patient's age, smoking habits, gender, clinical presentation, and lung HRCT pattern (nodulocystic, nodular or cystic) [3-5].

Briefly, in case of constitutional symptoms, infection is rigorously looked for (particularly mycobacteria, and pneumocystis *jirovec*i in case of lymphopenia). Bronchoscopy with BAL is performed to search for alternative diagnoses in case of predominant nodular, or cavitary nodules pattern at lung HRCT (infection, cavitary metastatic carcinoma, sarcoidosis or granulomatous polyangiitis…). In purely cystic presentation, LAM (either sporadic or associated with tuberous sclerosis) is the main alternative diagnosis in females. In both genders, the principal other diagnoses to consider are Birt-Hogg-Dubé syndrome, lymphoid interstitial pneumonia. Amyloidosis and light chain cystic lung disease are considered in an appropriate context. At the end of this process, in the absence of straightforward diagnosis, a surgical lung biopsy is performed, if the patients' respiratory function allows it.

LCH lung involvement was assessed by chest imaging, lung function measurement, oxygen saturation and/or blood gas analyses. The 6-minute walk test and Doppler echocardiography were indicated in patients with unexplained dyspnoea or isolated/disproportionate decrease in diffusing capacity for carbon monoxide (D_{LCO}) to detect pulmonary hypertension (PH), and if needed, PH was confirmed by cardiac catheterization [1, 6].

Follow-up visits systematically comprised assessment of smoking status, a comprehensive physical examination, chest radiography and lung function, including D_{LCO} measurement. Serial lung HRCT was indicated in case of changes in clinical, chest radiography or functional status during follow-up [1, 6]. For patients with unexplained dyspnoea or an isolated/disproportionate decrease in D_{LCO}, a Doppler echocardiography was also performed to detect PH [1, 6]. During the study, the median number of lung HRCT examinations performed was 3, (IQR 2; 5), corresponding to 0.8 HRCT examinations/patient/year of follow-up. One hundred forty-six (71%) patients had at least one Doppler echocardiography examination. For

these patients, the median number of Doppler echocardiography examinations performed was 2 (IQR 1; 4), corresponding to one Doppler echocardiography examination/patient every two years during the study period.

Supplementary tables

Supplementary Table S1. Univariable analyses of the associations of baseline characteristics of PLCH patients with survival

	Number of deaths*	Value	HR	95% CI	p-Value
Age	12		1.09	1.05; 1.14	<0.0001
Sex	12	M	1.00		
		F	0.50	0.16; 1.58	0.24
Smoking exposure, pack-years	11		1.05	1.02; 1.07	0.001
Isolated PLCH	12		0.95	0.26; 3.52	0.94
Chronic respiratory failure	12	No	1.00		
		Yes	10	2.16; 46.37	0.003
TLC, % predicted, n = 152	11		0.96	0.92; 1.01	0.088
RV, % predicted, n = 150	11		1.00	0.98; 1.02	0.96
RV/TLC, % predicted, n = 150	11		1.01	0.99; 1.03	0.30
FVC, % predicted, n = 158	10		0.97	0.94; 1.00	0.056
FEV_1, % predicted, n = 167	11		0.97	0.94; 0.99	0.020
FEV_1/FVC %, n = 158	10		0.94	0.89; 0.98	0.008
D_{LCO}, % predicted, n = 127	6		0.90	0.84; 0.96	0.002
Restriction	11	No	1.00		
		Yes	2.67	0.66; 10.83	0.17
Obstruction	10	No			
		Yes	1.39	0.29; 6.56	0.68
Air trapping	11	No			
		Yes	1.80	0.55; 5.90	0.33
HRCT nodular score†, n = 196	11	Low (0-6)	1.00		
		Intermediate (7-12)	0.94	0.24; 3.65	0.93
		High (13-18)	0.86	0.11; 7.02	0.89
HRCT cystic score‡, n = 196	11	Low (0-6)	1.00		
		Intermediate (7-12)	0.61	0.07; 5.10	0.65
		High/very high (13-24)	4.05	1.14; 14.35	0.03

*Number of deaths that occurred according to the characteristic considered. †Maximal value of the nodular score = 18. ‡Maximal value of the cystic score = 24.

PLCH: pulmonary Langerhans cell histiocytosis; HR: hazard ratio; CI: confidence interval; M = male; F: female; TLC: total lung capacity; RV: residual volume; FVC: forced vital capacity;

FEV_1: forced expiratory volume in 1 second; D_{LCO}: diffusing capacity for carbon monoxide; HRCT: high resolution computed tomography.

Supplementary Table S2. Univariable analyses of the associations of time-dependent characteristics of PLCH patients with survival

Characteristics (all evaluated at time t)	**Number of deaths**	**Values**	**HR**	**95% CI**	**p-Value**
Smoking status at time t	12	Non-smoker Smoker	1.00 2.5	0.79; 7.89	0.12
Previous use of systemic treatment	12	No Yes	1.00 5.64	1.79; 17.79	0.003
Previous use of cladribine	12	No Yes	1.00 5.69	1.25; 26.04	0.025
FEV_1 (n = 167)	11		0.95	0.91; 0.98	0.003
Age	12		1.09	1.05; 1.13	<0.0001

PLCH: pulmonary Langerhans cell histiocytosis; HR: hazard ratio; CI: confidence interval; FEV_1: forced expiratory volume in 1 second.

Supplementary Table S3. Characteristics of the 14 PLCH patients with CRF and/or PH

Patient	Age at diagnosis of PLCH, years	Sex	Smoking status at diagnosis	Smoking status at CRF	Long-term oxygen	Systemic treatment	PH characteristics	Status Alive/Dead
1	58	M	Former	Former	Yes	Steroids	Confirmed (mPAP=41 mmHg)	Dead
5	33	M	Current	Current	Yes	None	Confirmed (mPAP=28 mmHg)	Dead
6	47	M	Current	Current	No	Cladribine	Confirmed (mPAP=27 mmHg)	Dead
7	48	M	Current	Current	No	None	Probable (TRV=3.31 m/s; sPAP=53 mmHg)	Dead
8*	38	F	Former	Former	Yes	Steroids	Confirmed (mPAP=25 mmHg)	Dead
9	72	F	Current	Current	Yes	Cladribine†	Confirmed (mPAP=27 mmHg)	Dead
10	49	F	Current	No CRF	No	None	Probable (TRV=2.95 m/s; sPAP=40 mmHg)	Dead
20	19	M	Current	Current	Yes	Steroids	Confirmed (mPAP=34 mmHg)	Alive
21	48	F	Current	Current	Yes	Steroids	Confirmed (mPAP=26 mmHg)	Alive
22	51	F	Current	No CRF	No	None	Probable (TRV=3.04 m/s; sPAP=47 mmHg)	Alive
23	43	M	Current	Current	No	None	None	Alive
24	59	F	Current	Current	No	None	None	Alive
25	33	M	Current	Former	Yes	Cladribine	None	Alive
26	53	F	Current	Current	No	Cladribine	None	Alive

*This patient died one year after lung transplantation.

†This patient was previously treated with corticosteroids.

PLCH: pulmonary Langerhans cell histiocytosis; CRF: chronic respiratory failure; PH: pulmonary hypertension; M: male; F: female; mPAP: mean pulmonary arterial pressure; TRV: tricuspid regurgitation velocity; sPAP: systolic pulmonary arterial pressure.

Supplementary Table S4. Type and time of occurrence of the 27 malignancies observed in 23 PLCH patients during the study

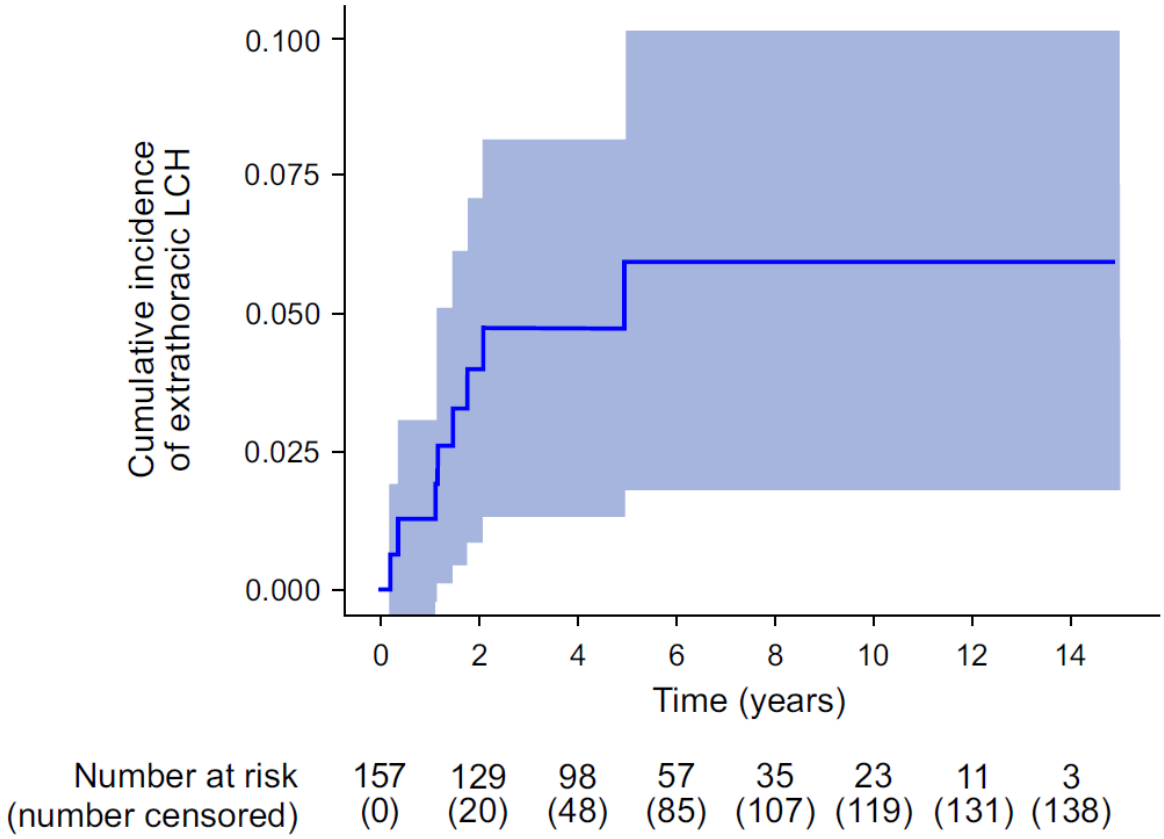
	Before PLCH (n=11)	**Concurrent with PLCH (n=6)**	**After PLCH (n=10)**
Time, median, (IQR), years	-3.13 (-6.2; -2.1)	Concurrent	4.1 (3.1; 5.0)
Solid neoplasms			
Anus	1		
Prostate	1		
Colon	1		
Stomach	1		
Lung	1	3*	7
Thyroid	1		
Spindle cell skin carcinoma	1		
Breast			3†
Haematological malignancies			
Marginal zone lymphoma	1		
Myelodysplasia	1		
Myeloma	1		
Skin lymphoma	1		
CMML		3	

*Eight patients with lung carcinomas had histological confirmation of PLCH either before or at the time of lung surgery for their tumour. The remaining two patients (one patient with metastatic lung carcinoma confirmed on liver biopsy, and one patient with lymphangitic carcinomatosis and respiratory failure) had a typical nodulocystic pattern on lung HRCT at the time of PLCH diagnosis.

†One patient with breast carcinoma had histological confirmation of PLCH and the remaining two patients presented a typical nodulocystic pattern on lung HRCT at the time of PLCH diagnosis.

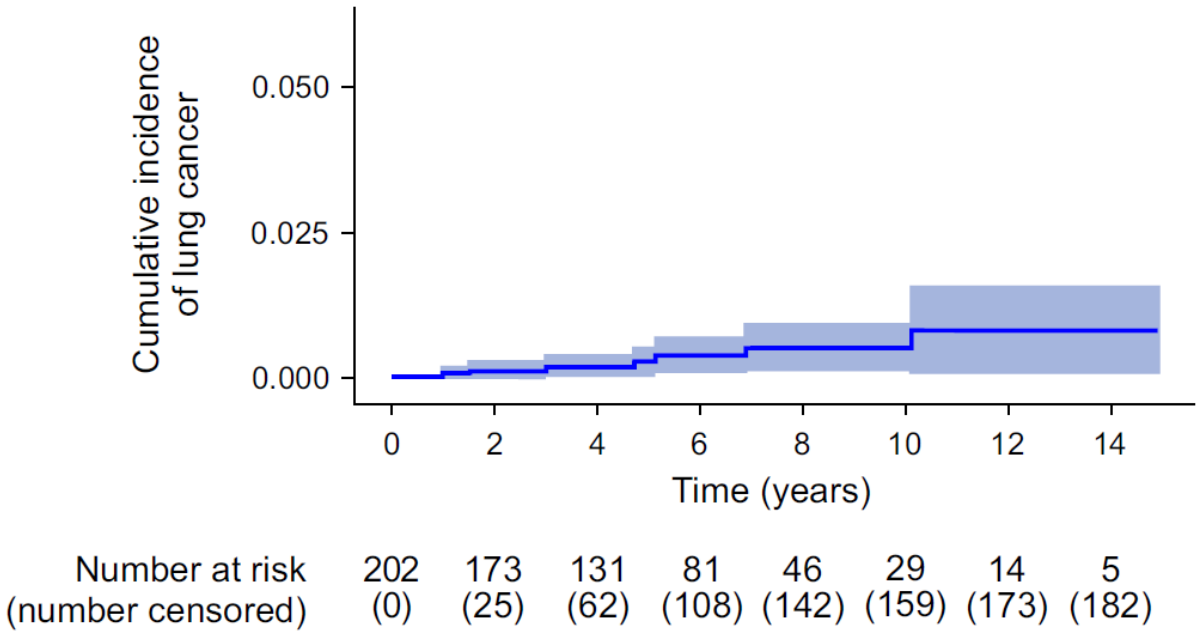
PLCH: pulmonary Langerhans cell histiocytosis; IQR: interquartile range; CMML: chronic myelomonocytic leukaemia.

Supplementary figures



Supplementary Figure S1. Cumulative incidence during follow-up of extra-pulmonary LCH localizations among the 157 patients with isolated PLCH at diagnosis. LCH involved the bone (n = 5), pituitary stalk with diabetes insipidus (n =3) or liver (n = 1). The patient with liver involvement, which occurred 2.1 years after diagnosis, also developed diabetes insipidus 4.3 years later. The shaded area represents the 95% confidence interval.

PLCH: pulmonary Langerhans cell histiocytosis.



Supplementary Figure S2. Cumulative incidence of lung carcinoma occurring after the diagnosis of PLCH. The shaded area represents the 95% confidence interval.

PLCH: pulmonary Langerhans cell histiocytosis.

Supplementary references

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