Semiquantitative assessment of the regional loads of Lewy body and tau neuropathology

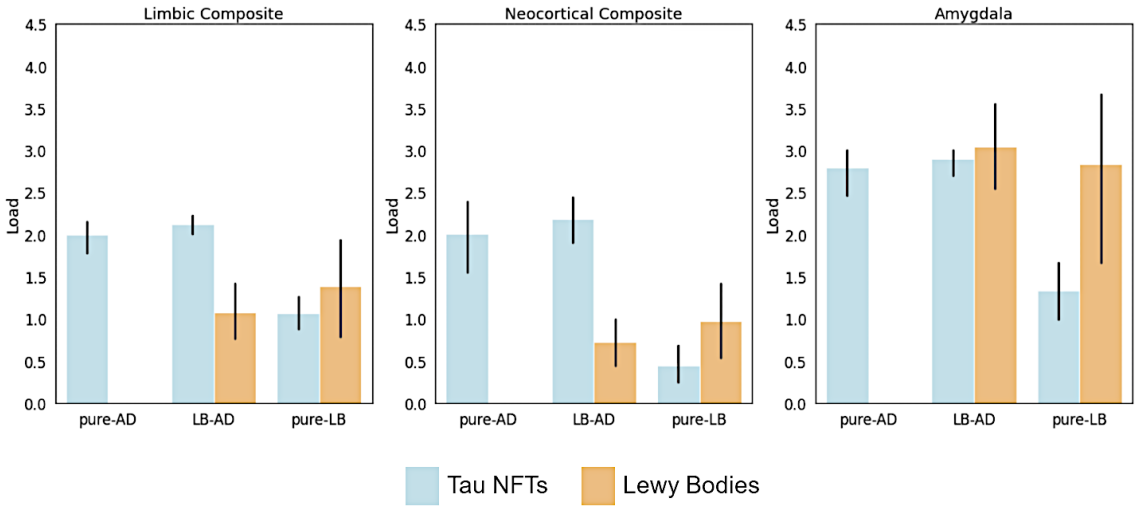
For a subset of patients (n=45/59; n=41 within our pathological groups; pure-AD=15; AD-LB=20; pure-LB=6) semiquantitative information about the regional loads of Lewy bodies (LBs) and tau neurofibrillary tangles (NFTs) in different brain regions was available. These were reported on a scale from 0 to 4 in the case of LBs (0, none; 1 = <1 LB x10 field; 2 = 1-3 LBs; 3 = 4-10; LB; 4 = >10) and from 0 to 3 for NFTs (0 = no NFTs, 1 = 1-5 NFT/1mm2, 2 = 6-20, 3 = > 20). The available data was used to calculate the average loads of LBs and tau NFTs in limbic and neocortical composite regions. The brain areas sampled for microscopic assessments are reported in Supplementary Table S1, together with their classification into the limbic or neocortical composite and the number of patients with data available for each region (right column). Regions with data for less than 30 patients in our pathological groups were excluded from subsequent analysis (marked in red).

Region	Neuropathology Code	Classification	Patients with data available (n)
Amygdala	L23AMYG	Limbic	41
Entorhinal	L23ENTX	Limbic	41
Hippocampus, CA1	L5CA1	Limbic	41
Hippocampus, Dentate Gyrus	L5DG	Limbic	41
Parahippocampal Gyrus	L5PHG	Limbic	41
Superior and Middle Temporal	L2STG	Neocortical	41
Middle frontal	L1MFG	Neocortical	41
Anterior Cingulate	L19CING	Limbic	41
Precentral Gyrus Motor Cortex	L21MX	-----	25
Inferior Parietal	L3IPL	Neocortical	41
Occipital	L4OL	Neocortical	41
Olfactory Cortex	L6OLFX	----	29
Caudate Putamen	L6PUTC	----	41
Globus Pallidus	L17GP	----	41
Thalamus	L8THAL	----	41
Pontine Base	L11PONS	----	41
Midbrain	L9SN	----	41
Nucleus Basalis Meynert	L17NBM	Limbic	39
Locus Caeruleus	L11LC	----	41
Medulla Oblongata	L12MED	----	40
Cebebellum Dentate Nucleus	L14CBM	----	41
Spinal Cord	L13SC	----	22

Supplementary Table S1: Regions for which semiquantitative information about the regional loads of NFTs and LBs was available. *Marked in red:* Regions with data for less than 30 patients.

Differences in the regional loads of LBs and NFTs between pathological groups

To test whether the differences between groups in the main analyses were related with the severity of pathology, we compared LB and NFT load between neuropathological groups. Results are presented in Suppl. Fig. 1. In statistical analysis, we did not find any significant differences in LB load between the pure-LB and the AD-LB groups (limbic composite: d=-0.35, p=0.394; neocortical composite: d=-0.36, p=0.382; amygdala: d=0.16, p=0.706). Similarly, for tau NFT burden, we did not observe any significant differences between the pure-AD and the AD-LB group (limbic composite: d=0.38, p=0.203; neocortical composite: d=0.20, p=0.505; amygdala: d=0.17, p=0.561).



Suppl. Fig. S1: Average regional loads of tau neurofibrillary tangles (NFTs) and Lewy bodies in limbic and neocortical composite regions, as well as in the amygdala separately.

Associations between regional LB loads and cognition

To better understand the role of LBs shaping the differences in cognition between the pathological groups, we evaluated continuous associations of the regional load of LBs in the amygdala and in limbic and neocortical composites with the reported memory performance (ADNI_MEM),

executive performance (ADNI_EXEC) and cognitive profile (Δ (MEM - EXEC)) variables. The correlations were evaluated using Spearman's correlation analysis (with and without controlling for the NFT load) for the whole cohort, and for the AD-LB group alone. Results are shown in Suppl. Table 2. While most of these correlations were not statistically significant, some interesting trends could be observed. Across the whole cohort, the LB load in all regions was negatively correlated with ADNI_MEM, especially when controlling for regional tau NFT load (limbic composite: ρ= -0.34, p=0.03; neocortical composite: ρ= -0.22, p=0.15; amygdala: ρ= -0.22, p=0.16). These associations were not observed for the AD-LB group alone, where the only remarkable finding was a correlation between Δ (MEM-EXEC) and amygdala LBs at trend-level statistical significance (ρ= 0.40, p=0.09), suggesting that these might help to shape a more executive phenotype within the AD-LB group.

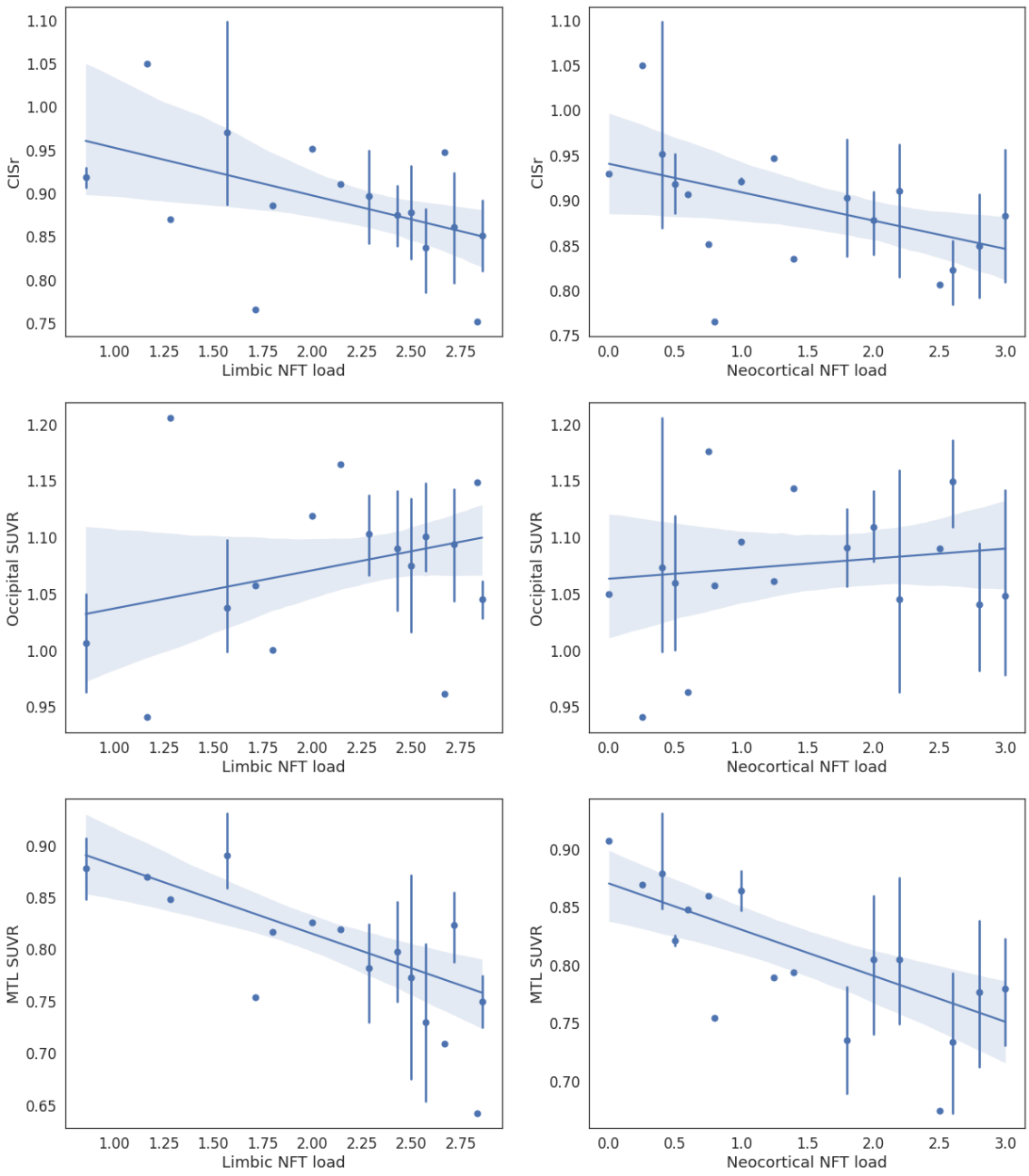
		Cognitive Score					
		ADNI_MEM		**ADNI_EF**		**Δ (MEM-EXEC)**	
		ρ (p)	**Partial ρ*, (p)**	**ρ (p)**	**Partial ρ*, (p)**	**ρ (p)**	**Partial ρ*, (p)**
		Results for the whole cohort					
Region Lewy Body Load	**Limbic**	-0.19 (0.20)	-0.34 (0.03)	-0.09 (0.56)	-0.15 (0.35)	-0.10 (0.54)	-0.15 (0.37)
	Neocortical	-0.09 (0.56)	-0.22 (0.15)	-0.13 (0.43)	-0.18 (0.26)	0.01 (0.97)	-0.02 (0.90)
	Amygdala	-0.21 (0.18)	-0.22 (0.16)	-0.10 (0.53)	-0.11 (0.52)	-0.10 (0.53)	-0.12 (0.47)
		Results for the AD-LB group					
Region Lewy Body Load	**Limbic**	0.15 (0.51)	0.08 (0.72)	0.22 (0.36)	0.15 (0.54)	-0.10 (0.70)	-0.06 (0.80)
	Neocortical	0.10 (0.67)	0.18 (0.45)	-0.01 (0.96)	0.04 (0.86)	0.05(0.85)	-0.05 (0.84)
	Amygdala	0.23 (0.33)	0.25 (0.28)	-0.02 (0.93)	-0.13 (0.59)	0.24 (0.30)	0.40 (0.09)

Suppl. Table S2: Associations between cognitive scores (ADNI_MEM, ADNI_EF, Δ (MEM-EXEC)) and neuropathologically assessed regional loads of LBs across different regions of interest

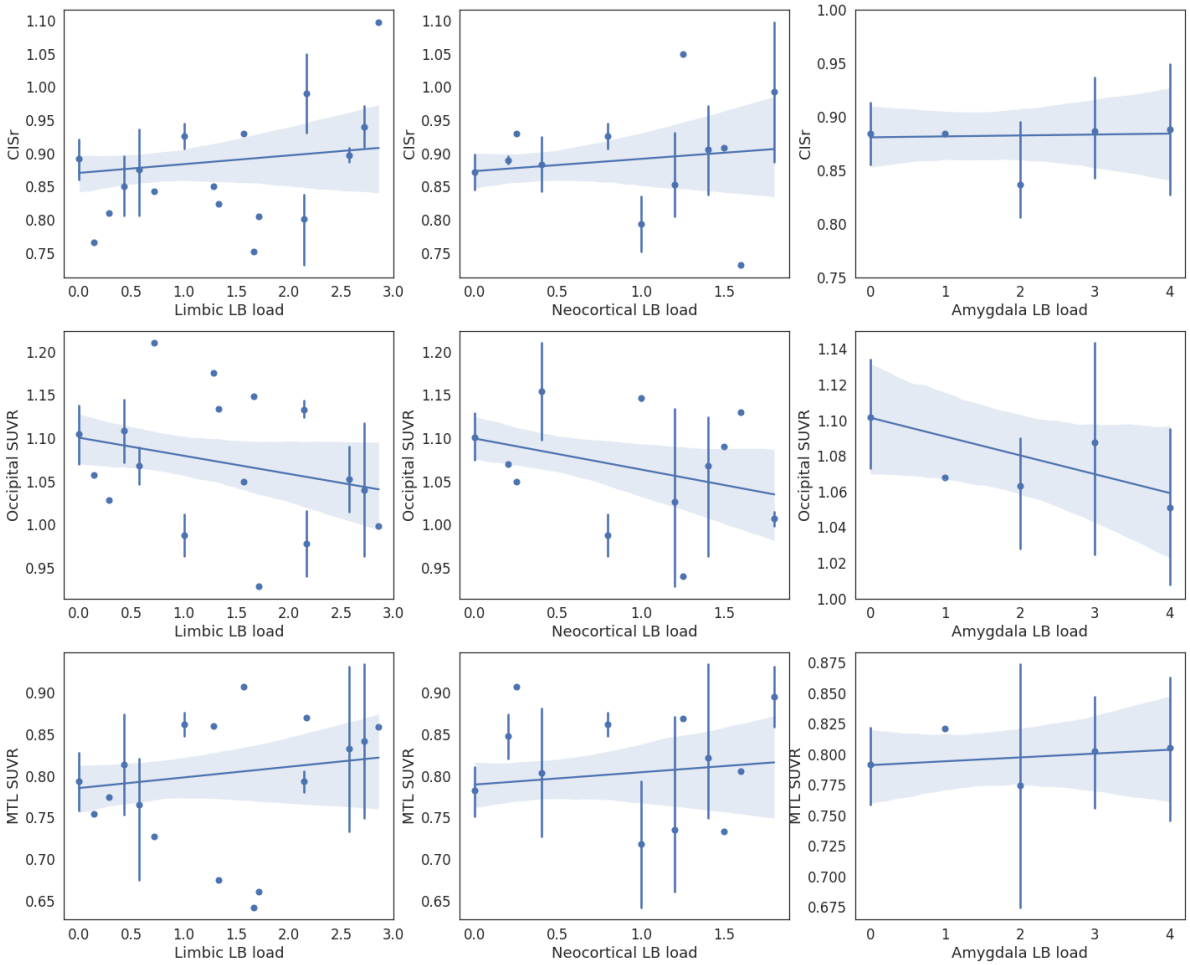
in the whole cohort and separately in the AD-LB group. * indicates partial Spearman correlation using the regional tau NTF load as a covariate

Associations of the regional loads of NFTs and LBs with FDG-PET

Following up on the observed associations of Braak stage and SNnl with the CISr, occipital SUVR and MTL SUVR (see Fig. 4 of the main manuscript), we tested whether similar associations were also found when using regional loads of tau NTFs and LBs as neuropathologic markers. Similar to Braak tau stages, both limbic and neocortical NFTs were significantly associated with a lower CISr (ρ=-0.341, p=0.027; ρ=-0.31, p=0.047) and lower MTL SUVR (ρ=-0.40 p=0.009; ρ=-0.44 p=0.003), but not with occipital metabolism (ρ=0.14, p=0.364; ρ=0.07, p=0.639; Supplementary Figure S2). However, in contrast to SNnl, limbic, neocortical and amygdala regional LB loads were not significantly associated with the CISr (ρ=0.04, p=0.782; ρ=0.11 p=0.510; ρ=0.02, p=0.905) or MTL (ρ=0.14, p=0.398, ρ=0.19, p=0.241; ρ=0.09, 0.564), although trend-level negative correlations were found between regional LB load and lower occipital SUVR (ρ=-0.30, p=0.060, ρ=-0.27, p=0.084; ρ=-0.26, p=0.108) (Supplementary Figure S3).



Suppl. Fig. S2: Associations between limbic (left) and neocortical (right) tau NFT loads and different FDG-PET ROI features: CISr (top), occipital SUVR (center) and MTL SUVR (bottom).



Suppl. Fig. S3: Associations between limbic (left), neocortical (center) and amygdala LB loads and different FDG-PET ROI features: CISr (top), occipital SUVR (center) and MTL SUVR (bottom).