QUANTIFICATION OF NANOSCALE FREE VOLUMES IN HUMAN PLASMA CLOTS USING POSITRON ANNIHILATION LIFETIME SPECTROSCOPY

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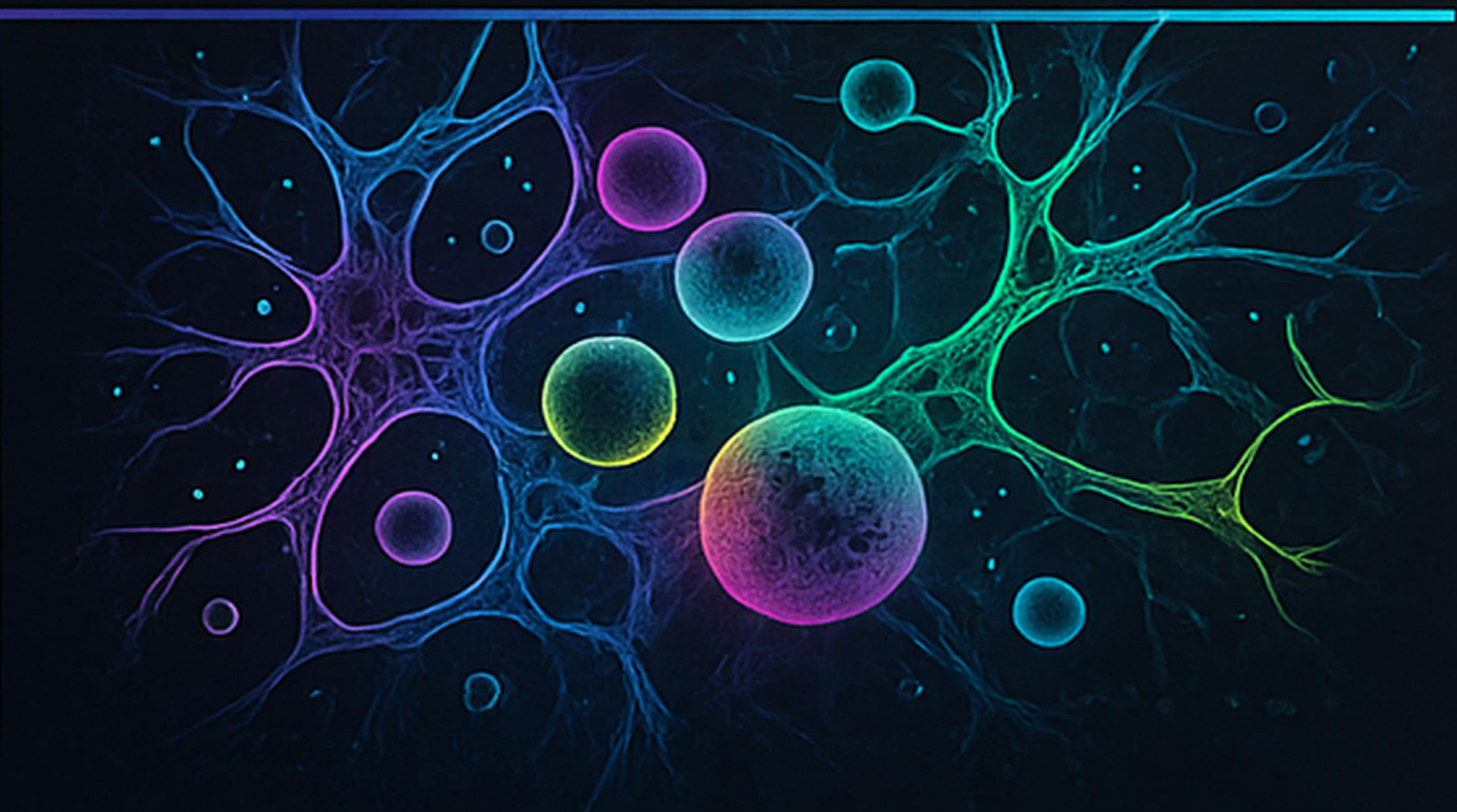
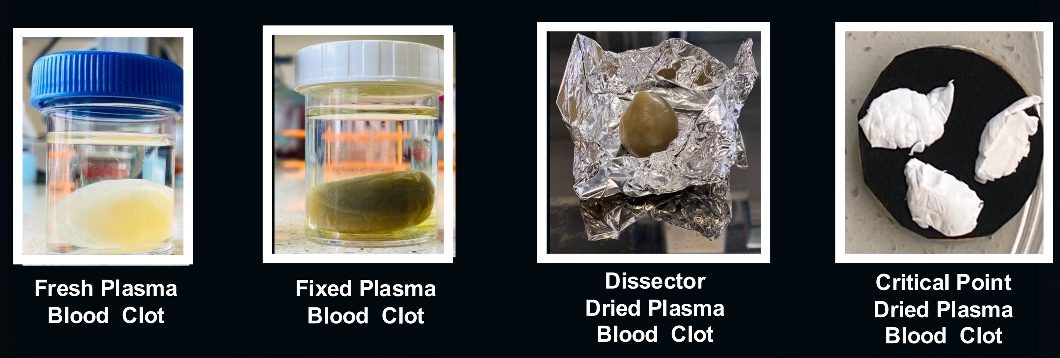
INTRODUCTION

- Blood clot microstructure, especially nanoscale free volumes, governs molecular transport, mechanical stability, and susceptibility to lysis [1–3].
- Positronium annihilation lifetime spectroscopy (PALS) is a non-destructive method that senses sub nanometer to nanometer voids by measuring the lifetime (τ_3) and intensity (I_3) of ortho-positronium formed in these spaces [4–9].
- The Tao–Eldrup model converts τ_3 to cavity radius (R) and then to mean free volume (V_f) and fractional free volume (f_v), which enables quantitative comparisons across temperatures and preparation states [4,8].
- This study applies PALS to the biomedical domain by quantifying nanoscale free volumes in human plasma clots and in patient pulmonary thrombi.
- Our objectives are to establish that o-Ps lifetimes can be measured reproducibly in these samples and to test for differences in τ_3, R, V_f, and f_v across preparation conditions and between plasma clots and thrombi [1–3,10–12].



MATERIALS



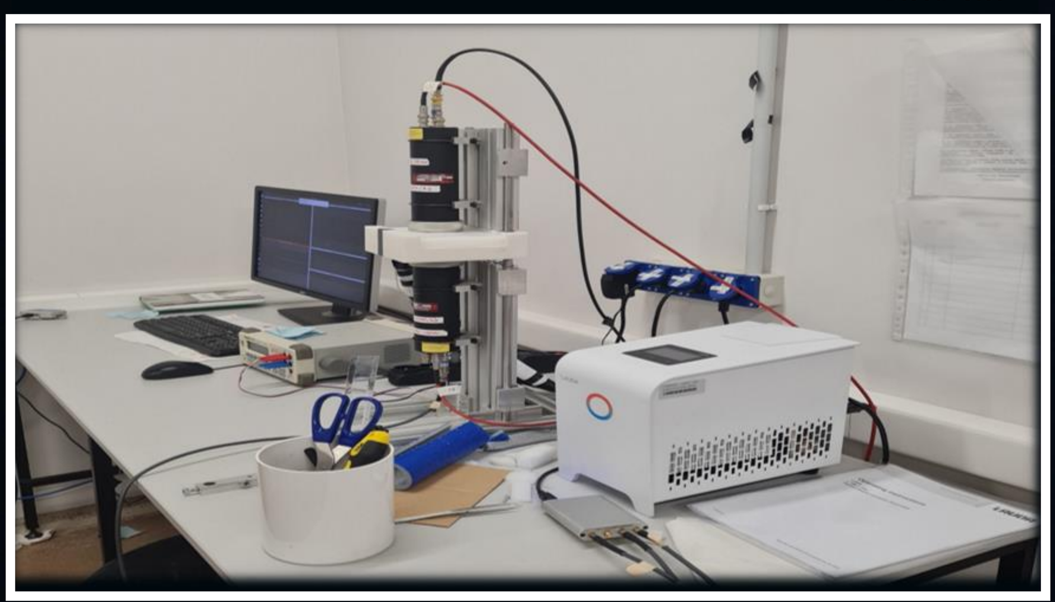


Fig 1. PALS system used in this work
(BaF_2 detectors, temperature control via LAUDA circulator).

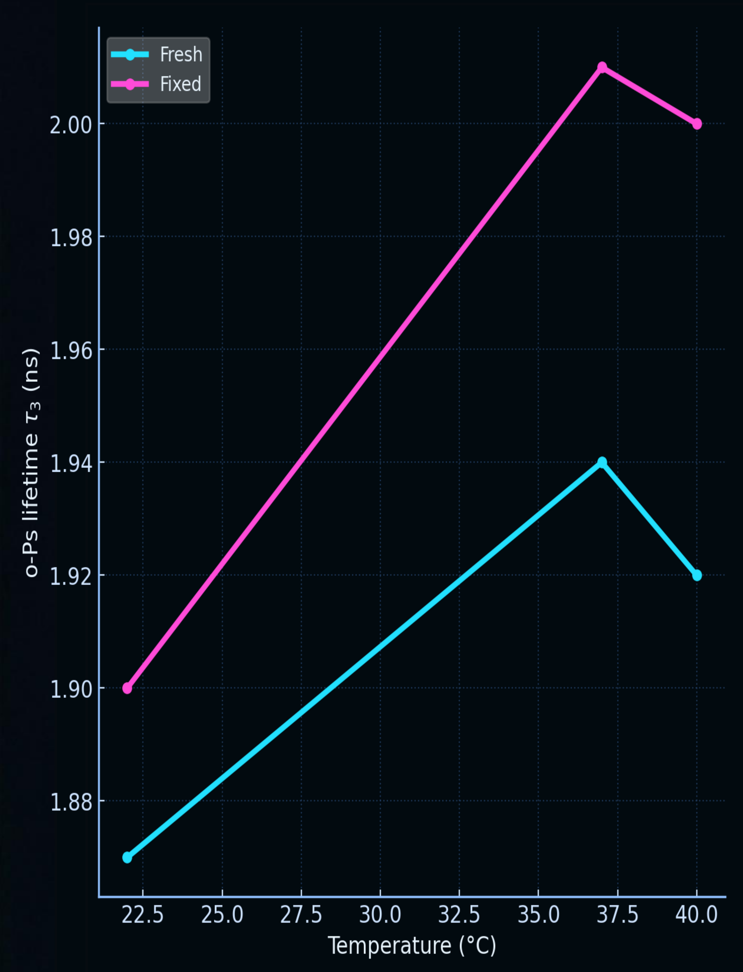


Fig. 3. Temperature effect on o-Ps lifetime (τ_3).

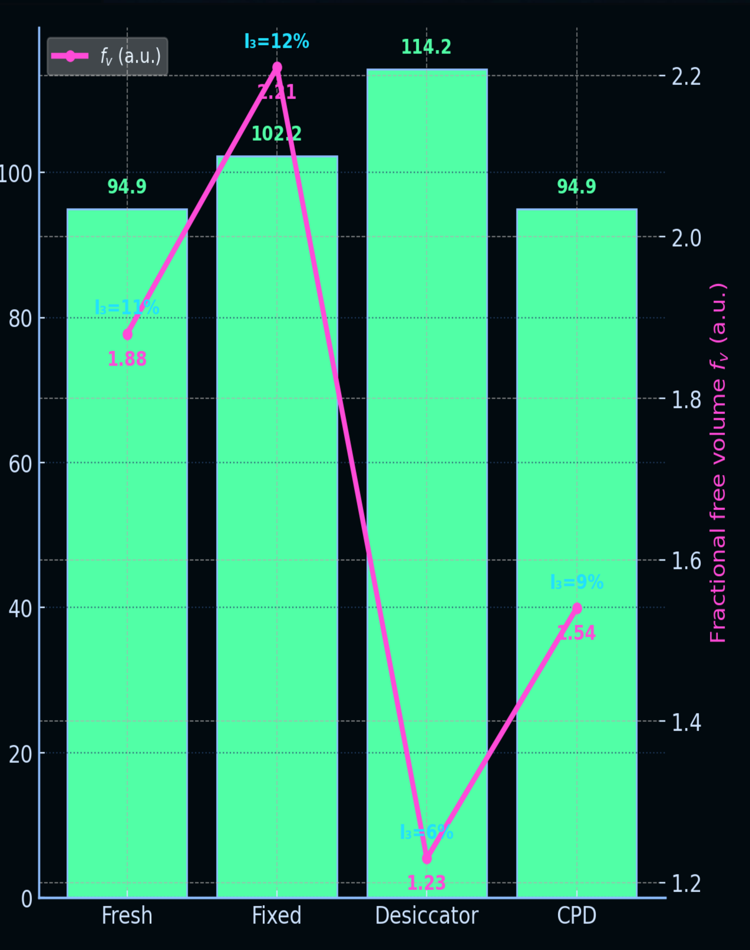


Fig. 4. Preparation States at 37 ºC :geometry vs effective free volume.

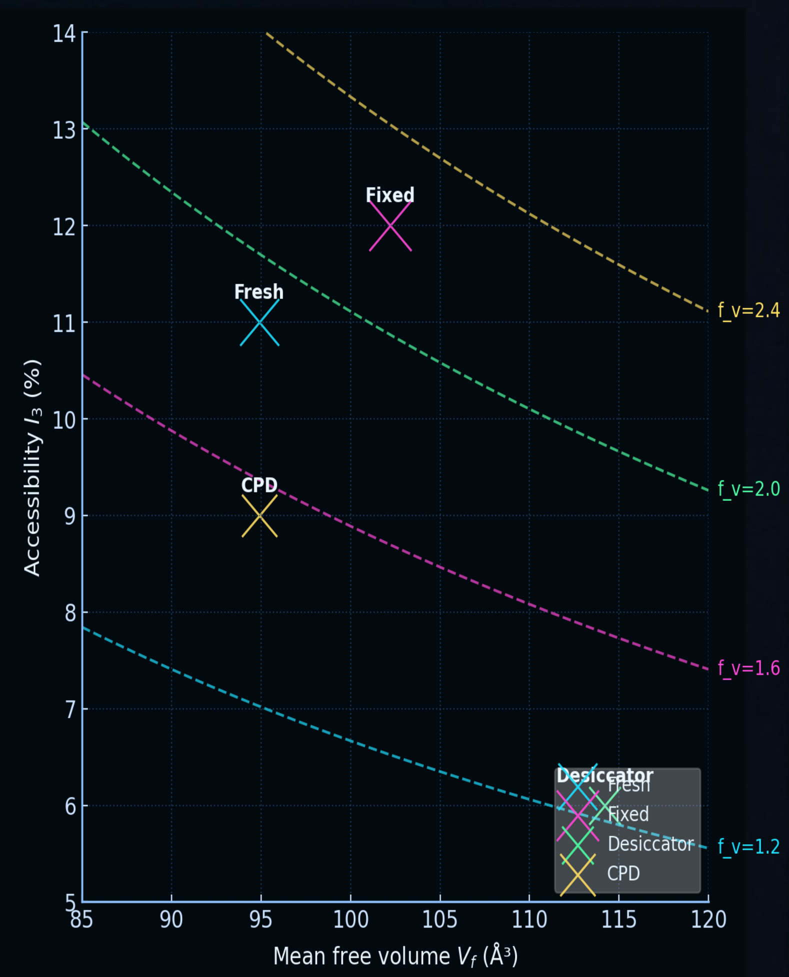


Fig. 5. Geometry vs accessibility map

Table 1. Free-volume characteristics vs temperature for fresh and fixed plasma clots (Å³-convention).

Sample	Temp (°C)	τ_3 (ns)	I_3 (%)	R (nm)	V_f (Å³)	f_v (a.u.)
Fresh	22	1.87	11.0	0.275	87.1	1.72
Fresh	37	1.94	11.0	0.282	93.9	1.86
Fresh	40	1.92	11.0	0.280	92.0	1.82
Fixed	22	1.90	3.8	0.280	92.0	0.63
Fixed	37	2.01	12.0	0.290	102.2	2.21
Fixed	40	2.00	14.0	0.286	98.0	2.47

Table 2. Free-volume characteristics of plasma clots at 37 °C across preparation states (Å³-convention).

Sample	Temp (°C)	τ_3 (ns)	I_3 (%)	R (nm)	V_f (Å³)	f_v (a.u.)
Fresh	37	1.95 ± 0.01	11.0 ± 0.07	0.283	94.9	1.88
Fixed	37	2.03 ± 0.02	12.0 ± 0.07	0.290	102.2	2.21
Desiccator	37	2.15 ± 0.04	6.0 ± 0.01	0.301	114.2	1.23
CPD	37	1.95 ± 0.01	9.0 ± 0.07	0.283	94.9	1.54

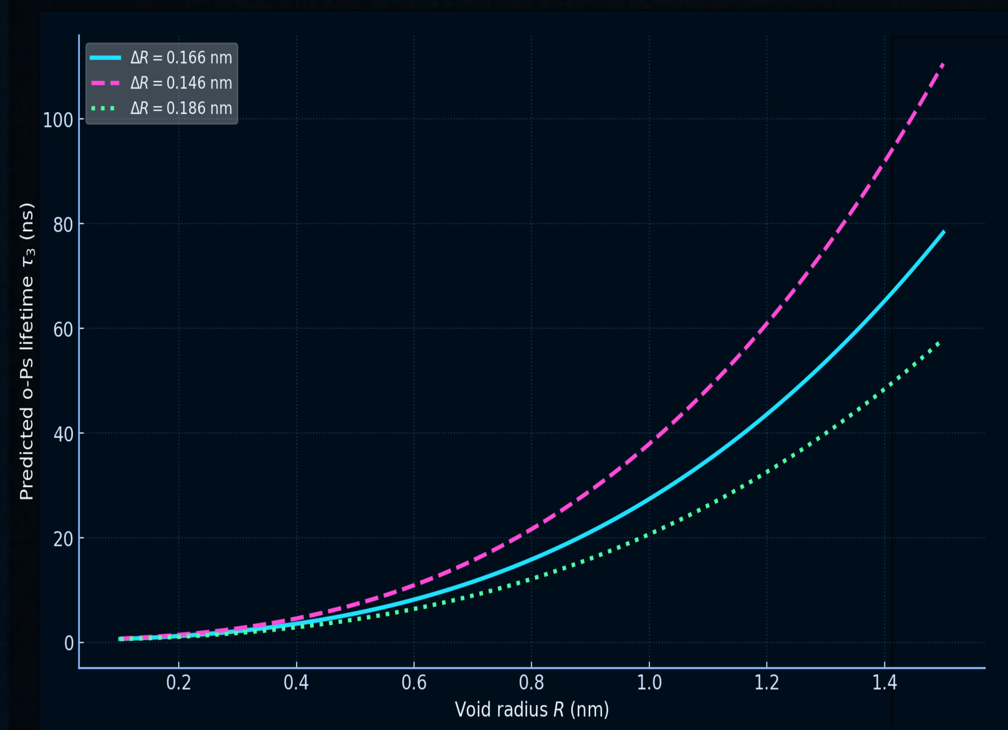


Fig. 2. Tao–Eldrup model prediction. Predicted o-Ps lifetime (τ_3) as a function of void radius (R) for different overlap layers ΔR (0.146, 0.166, 0.186 nm). Larger R gives longer τ_3; changing ΔR shifts the inferred R for a given τ_3.

Tao-Eldrup model (approximation)

$$\tau_3(\text{ns}) = 0.5\left[1-\frac{R}{R+\Delta R}+\frac{1}{2\pi}\sin\left(\frac{2\pi R}{R+\Delta R}\right)\right]^{-1}$$

$\Delta R = 1.656$ Å

Valid for $\tau_3 \approx$ 1–3 ns in our measurements.

Geometric free volume (sphere)

$$V_f = \frac{4}{3}\pi R^3$$

Units: R in Å, V_f in Å³.

Fractional free volume (Å³-convention)

$$f_v = C V_f I_3$$

C = 0.0018; I_3 in percent (%).

Note: 1 nm³ = 1000 Å³. If V_f is entered in nm³, f_v will be 1000× smaller; we standardize to Å³.

- τ_3 (o-Ps lifetime): tracks void size in the fibrin network.
- I_3 (o-Ps intensity): tracks accessibility of those voids (how often o-Ps forms).
- ΔR: empirical overlap layer at the wall; we use 1.656 Å.
- R = cavity radius
- V_f = (4/3)πR^3: geometric volume.
- f_v = C·V_f·I_3: fractional free volume (Å³-convention; C = 0.0018).

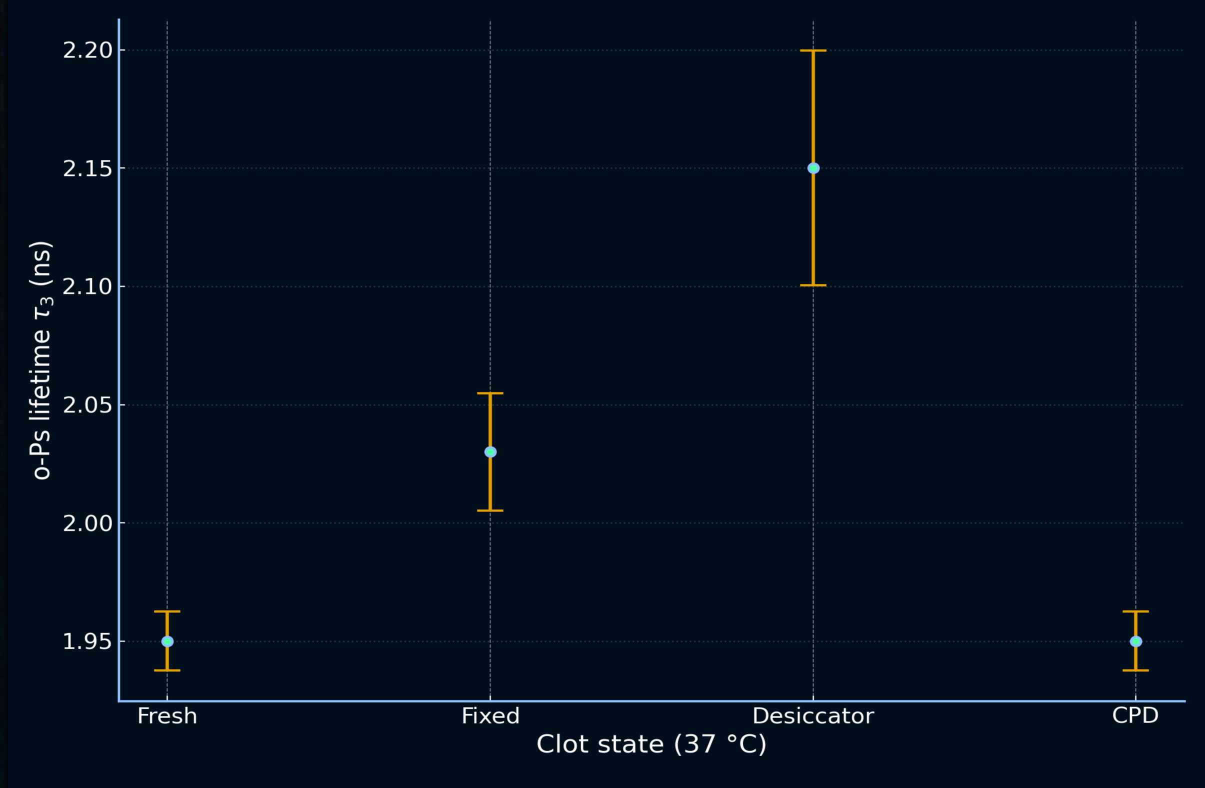


Fig. 6. o-Ps lifetime (τ_3) by Clot state at 37 ºC.

Key Findings

- Temperature modestly loosens fresh networks; in fixed samples, temperature mainly raises I_3, driving the largest gains in f_v. At 37 ºC, fixation best balances geometry and access; it yields the highest functional free volume f_v.
- Geometry alone isn't enough; bigger voids (R, V_f) raise f_v only if accessibility I_3 is high.
- Desiccation enlarges geometric voids but suppresses I_3 → lowest f_v (big pockets, poor access).
- CPD preserves fresh-like geometry but reduces I_3 → f_v below fresh.

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