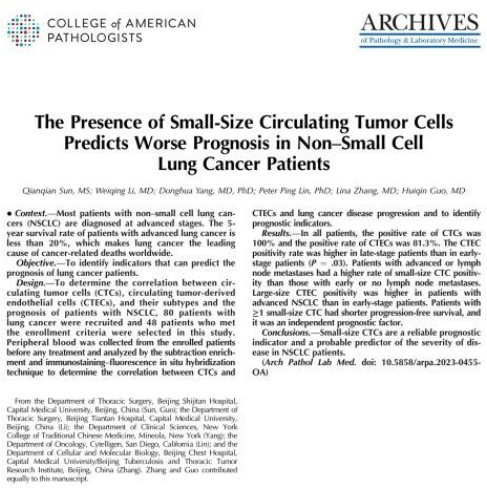
小细胞 CTC 与肺癌较差预后密切相关

大部分非小细胞肺癌 (NSCLC) 患者确诊时已是肿瘤晚期，5 年生存率的患者不足 20%，致使肺癌在全球范围内一直是肿瘤致死率最高的癌种。随着小细胞 CTC[1] 在肿瘤转移过程中的细胞间质化[2, 3]、乳腺癌肺转移[4]、肝癌及肺癌术后复发[5, 6] 中的作用被不断揭示，这类在使用细胞筛过滤法分离 CTC 过程中被大量丢失的小细胞 CTC[7] 的重要临床意义近年来已受到人们日益广泛的密切关注。最近，首都医科大学肿瘤医学院 (北京世纪坛医院)、首医北京天坛医院、首医北京胸科医院等单位与赛特生物应用 SE-i•FISH 技术，深入研究了 NSCLC 的小细胞 CTC、循环肿瘤血管内皮细胞 CTEC 与肿瘤进展及预后的相关性，相关成果刚刚在美国病理学家学会 (CAP) 官方期刊上得到发表 (Sun et al. 2024 Arch Pathol Lab Med doi 10.5858/arpa.2023-0455-OA)。



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The Presence of Small-Size Circulating Tumor Cells Predicts Worse Prognosis in Non–Small Cell Lung Cancer Patients

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• *Context.*—Most patients with non–small cell lung cancers (NSCLC) are diagnosed at advanced stages. The 5-year survival rate of patients with advanced lung cancer is less than 20%, which makes lung cancer the leading cause of cancer-related deaths worldwide.

Objective.—To identify indicators that can predict the prognosis of lung cancer patients.

Design.—To determine the correlation between circulating tumor cells (CTCs), circulating tumor-derived endothelial cells (CTECs), and their subtypes and the prognosis of patients with NSCLC, 80 patients with lung cancer were recruited and 48 patients who met the enrollment criteria were selected in this study. Peripheral blood was collected from the enrolled patients before any treatment and analyzed by the subtraction enrichment and immunostaining–fluorescence in situ hybridization technique to determine the correlation between CTCs and CTECs and lung cancer disease progression and to identify prognostic indicators.

Results.—In all patients, the positive rate of CTCs was 100% and the positive rate of CTECs was 81.3%. The CTEC positivity rate was higher in late-stage patients than in early-stage patients ($P = .03$). Patients with advanced or lymph node metastases had a higher rate of small-size CTC positivity than those with early or no lymph node metastases. Large-size CTEC positivity was higher in patients with advanced NSCLC than in early-stage patients. Patients with ≥1 small-size CTC had shorter progression-free survival, and it was an independent prognostic factor.

Conclusions.—Small-size CTCs are a reliable prognostic indicator and a probable predictor of the severity of disease in NSCLC patients.

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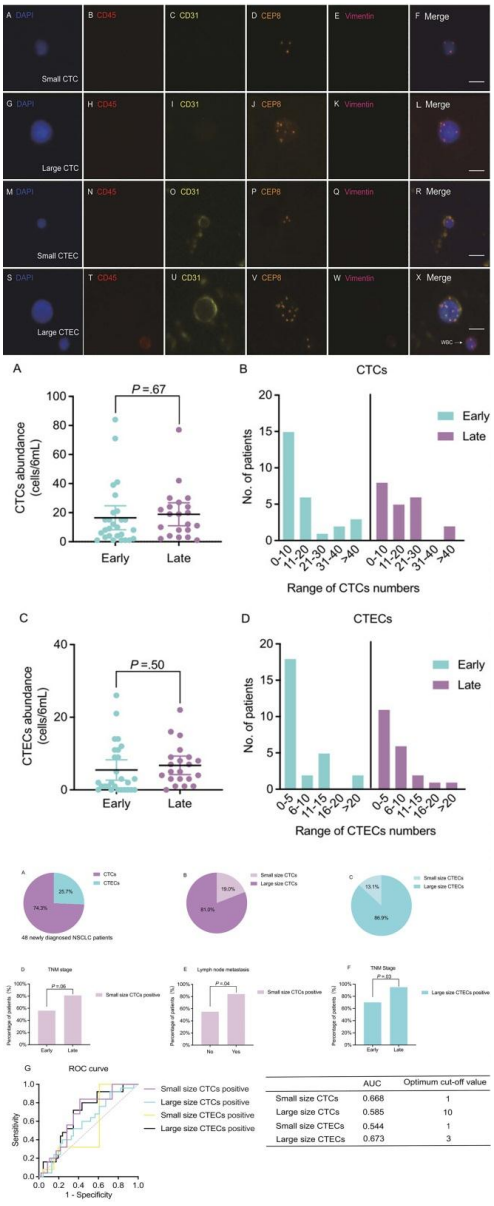
本文要点

- 应用赛特 SE-i•FISH 检测 48 例入组患者 (27 例 I-IIIA 早期及 21 例 IIIB-IV 晚期病人)：所有患者 (100%) 均可检出 $CD31^-$ 异倍体 CTC；81.3% 患者可检出 $CD31^+$ 异倍体 CTEC，其中，晚期患者 CTEC 阳性检出率高于早期患者，并以大细胞 CTEC 为主
- 晚期或伴有淋巴转移的患者，其小细胞 CTC 阳性率明显高于早期或无淋巴转移患者
- 患者检出 ≥ 1 小细胞 CTC，预后较差，PFS 明显缩短
- 小细胞 CTC 可作为预测 NSCLC 较差预后的可靠、独立风险评估因素

定量分析

在检测出的所有循环异倍体细胞 (circulating aneuploid cells, CACs)中，$CD31^-$ CTC 占比 74%，$CD31^+$ CTEC 占比 26%。无论 CTC 或 CTEC，其在早期与晚期 NSCLC 患者体内的数量不具备统计学显著差异，提示 CTC、CTEC 总体数量与 TNM 分期并不相关。将 CTC、CTEC 进一步细分为大细胞 (≥5 μm)、小细胞 (<5 μm) 进行分析，结果显示小细胞 CTC 与大细胞 CTC 的比例分别为 19%及

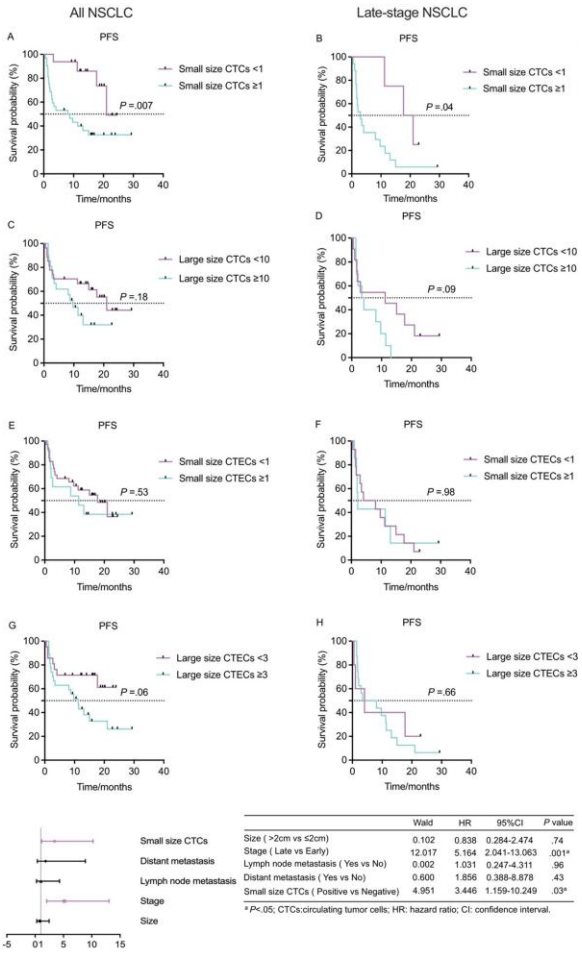
81%，小细胞 CTEC 则占比 13%，其余 87%为大细胞 CTEC。类似 CTC 总类 (包含大、小两类细胞)，小细胞 CTC 也与 TNM 分期不相关，但与淋巴结转移密切相关。不同于 CTEC 总类，大细胞 CTEC 与 TNM 分期密切相关 (p=0.03)，更多的晚期 NSCLC 患者体内可检测出大细胞 CTEC。



	AUC	Optimum cut-off value
Small size CTCs	0.668	1
Large size CTCs	0.585	10
Small size CTECs	0.544	1
Large size CTECs	0.673	3

ROC 曲线分析显示，小细胞 CTC 及大细胞 CTEC 具有较高的 AUC values, 分别为 0.668、0.673，即具备最高的诊断价值。若将小细胞 CTC cut-off 值定为 1 cell，大细胞 CTEC cut-off 定为 3 cells，则它们的阳性检出率分别为 84% 及 72%。

预后分析



	Wald	HR	95%CI	P value
Size (>2cm vs ≤2cm)	0.102	0.838	0.284-2.474	.74
Stage (Late vs Early)	12.017	5.164	2.041-13.063	.001*
Lymph node metastasis (Yes vs No)	0.002	1.031	0.247-4.311	.96
Distant metastasis (Yes vs No)	0.600	1.856	0.388-8.878	.43
Small size CTCs (Positive vs Negative)	4.951	3.446	1.159-10.249	.03*

* P<.05; CTCs:circulating tumor cells; HR: hazard ratio; CI: confidence interval.

根据 ROC 曲线分析结果，作者分别比较了小细胞 CTC (cut off =1)、大细胞 CTC (cut off =10)、 小细胞 CTEC (cut off =1) 及大细胞 CTEC (cut off =3) 与所有病人及晚期患者预后的相关性，Kaplan-Meier 生存分析显示，

只有小细胞 CTC<1 时 (即阴性检出)，所有 NSCLC 病人或晚期患者的无进展生存期 (progression-free survival, PFS) 明显高于小细胞 CTC≥1 患者，提示小细胞 CTC 的阳性检出预示 NSCLC 较差预后。针对所有患者的进一步多变量生存分析 (森林图，forest plot) 显示，小细胞 CTC 及 TNM 分期是影响预后的独立风险因素。小细胞 CTC 阳性检出的 NSCLC 患者，其肿瘤进展的风险是不含此种细胞的 3.446 倍！

展望

与日常体外培养的肿瘤细胞系细胞不同，患者体内的肿瘤细胞 TC/CTC、肿瘤血管内皮细胞 TEC/CTEC 千变万化，具有极高的异质性。在检测与分析过程中避免以偏概全，应根据细胞三要素 (染色体、瘤标、细胞形态) 将各种细胞进行亚类分型，并充分分析、锁定各个不同亚类细胞的临床意义，从而为肿瘤的精准防治奠定坚实基础。

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