Molecular Med Tri-CON, San Francisco, CA, Feb. 11-16, 2018

Contact: cmtang@creatvmicrotech.com
301-983-1650

Cancer associated macrophage-like cells are prognostic indicators of survival in a variety of solid malignancies

Daniel Adams[1], Diane K Adams[2], Steven H. Lin[3], Massimo Cristofanilli[4] Raymond C. Bergan[5], Jeffrey Marks[6], Stuart S Martin[7], Saranya Chumsri[8], Thai H. Ho[9], Rena G. Lapidus[7], Susan Tsai[10], Cha-Mei Tang[11], R. Katherine Alpaugh[12]

[1] Creatv MicroTech, Inc., Monmouth Junction, NJ 08852, [2] Rutgers, the State University of New Jersey, New Brunswick, NJ 08901, [3] MD Anderson Cancer Center, Houston, TX 77030, [4] Northwestern University, Robert H Lurie Cancer Center, Chicago IL 60611, [5] Oregon Health and Sciences University, Portland, OR 97239, [6] Duke University Medical Center, Durham, NC 27710, [7] University of Maryland Greenebaum Cancer Center, Baltimore, MD 21201, [8] Mayo Clinic Cancer Center, Jacksonville, FL 32224, [9] Mayo Clinic Cancer Center, Scottsdale, AZ 85259, [10] Medical College of Wisconsin, Milwaukee, WI 53226, [11] Creatv MicroTech, Inc., Potomac, MD 20854, [12] Fox Chase Cancer Center, Philadelphia, PA 19111

ABSTRACT

Cancer associated macrophage-like cells (CAMLs) are a recently described circulating stromal cell subtype commonly found in the peripheral blood of cancer patients. While CAMLs have been identified in all stage of solid malignancies and in a variety of cancer subtypes, no study has evaluated their clinical use as it relates to cancer prognosis. In an effort to elucidate the clinical utility of CAMLs as it relates to cancer progression, we ran a multi-institutional prospective 2-year study of 293 cancer patient samples in 6 types of solid tumors.

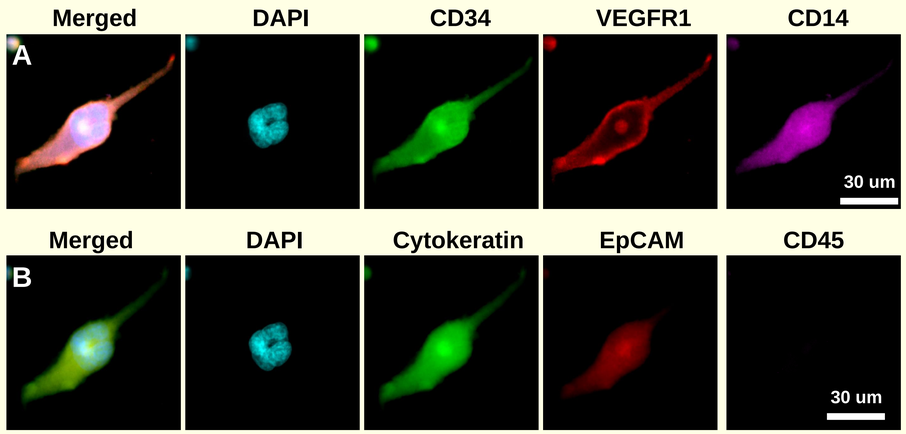


Figure 1. Example of QUAS-R screening for 6 subtyping markers on one CAML, including epithelial (Cytokeratin/EpCAM), white blood cell (CD45), myeloid (CD14), & stem (CD34).

INTRODUCTION

CAMLs are specialized myeloid polyploid cells transiting the circulation of patients with various types of solid malignancies and appearing in all stages of cancer[1-4]. While CAMLs are easy to identify by their large size and polyploid nucleus, they appear to present as stem cell like phenotype with multiple heterogeneous epithelial, myeloid, and endothelial markers.

Size exclusion is the only known technique for isolating large cells from peripheral patient blood irrespective of their surface marker expression. CellSieve™ microfilters are size exclusion membranes which efficiently isolate CAMLs and circulating tumor cells (CTCs) from whole blood, making it possible to study both cell types in relation to malignant disease[1-4].

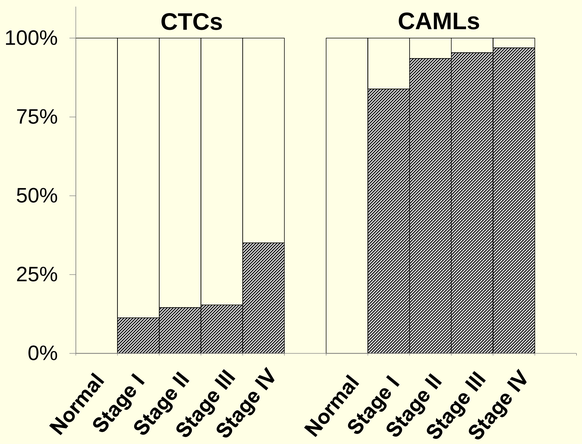
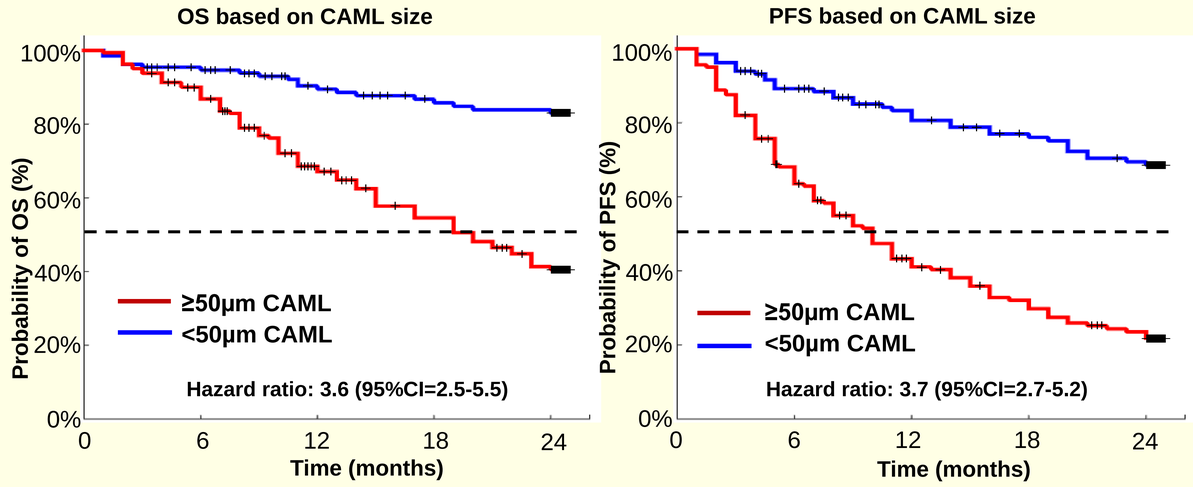


Figure 2. Percentage of patients positive for CTCs and CAMLs by stage

MATERIALS & METHODS

This single blind study consisted of 293 stage I-IV patients; breast (n=59), esophageal (n=27), prostate (n=52), pancreatic (n=59), lung (n=59), and renal cell (n=37), in treated (n=123) and untreated baseline (n=170). 7.5mL of whole peripheral blood was filtered by CellSieve™ microfiltration. CAMLs were identified as giant polyploid cells that express cytokeratin 8, 18 & 19, CD45, and/or CD14, as previously described[1]. Patients were grouped by CAML number (<6 or ≥6) and by size (<50 or ≥50 µm) to determine hazard ratios (HR) by censored univariate & multivariate analysis.

Figure 4. Analysis of OS and PFS based on size of CAMLs.



RESULTS

- In 7.5 mL blood, CAMLs were found in 91% of all cancer patients (n=266/293), but in none of the healthy control samples (Fig. 2)
- CAMLs were common, found in stage 1 (84%), stage 2 (94%), stage 3 (95%) or stage 4 (97%). N
- Number of CAMLs was exponentially associated with disease stage averaging 4.7 (Stage I), 4.7 (Stage II), 8.7 (Stage 3), 12.0 (Stage IV).
- In a univariate analysis of CAML number, optimal HR stratification for progression free survival (PFS) and overall survival (OS) occurred in patients with ≥6 CAMLs per sample (Fig. 3): OS (HR=1.9, p=0.006) and PFS (HR=1.8, p=0.003).
- In a multivariate analysis CAML size of ≥50 µm was the most significant factor, Fig. 4: OS (HR= 3.6, p<0.001) and PFS (HR=3.7, p<0.001).

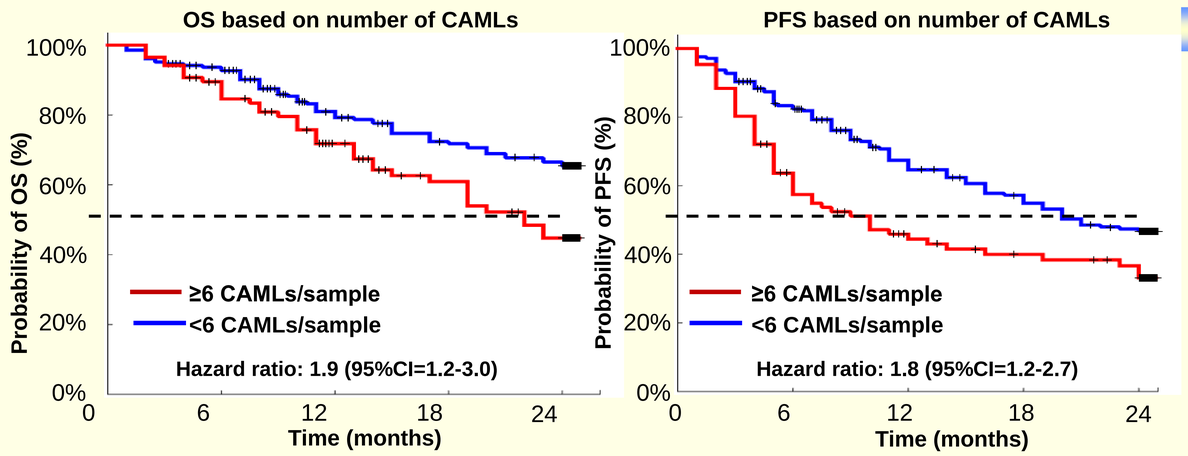


Figure 3. Analysis of OS and PFS based on number of CAMLs.

CONCLUSIONS

- In a multivariate analysis CAML size was the most predictive variable for PFS & OS and was independent of other clinical variables tested, including CTCs.
- Our data suggests that in solid malignancies CAML number and size clinically correlate with OS and PFS in early and late stage solid malignancies.
- Additional studies are warranted to determine if CAMLs can serve as a general clinically prognostic blood based marker.

References

1. Adams DL, et al "Circulating giant macrophages as a potential biomarker of solid tumors." Proc Natl Acad Sci, 2014 111(9):3514-3519
2. Mu et al, Detection and Characterization of Circulating Tumor Associated Cells in Metastatic Breast Cancer" *Int. J. Mol. Sci.* 2016, *17*(10), 1665
3. Cristofanilli M, "Liquid Biopsies in Solid Tumors" *Springer International Publish.* 2017
4. Adams DL, et al, "Multi-Phenotypic subtyping of circulating tumor cells using sequential fluorescent quenching and restaining." *Sci Rep. 6(33488) 2016*

Funding Sources

This work was supported by a Maryland (TEDCO) MTTCF award, grant R01-CA154624 from the National Cancer Institute, grant KG100240 from the Susan G. Komen Foundation, and the U.S. Army Research Office (ARO) and the Defense Advanced Research Projects Agency (DARPA) (W911NF-14-C-0098). The content of the information does not necessarily reflect the position or the policy of the US Government.