Cambridge Polymer Group

Sniffing Out Biomarkers with HS-GC-MS

By Becky Bader, Ph.D., Associate Director of Chromatography

Summary

Researchers from MIT and Johns Hopkins collaborated with Cambridge Polymer Group (CPG) to develop an electronic "nose" with greater sensitivity to prostate cancer than a dog's nose. This innovative project combined advanced analytical chemistry techniques with artificial intelligence to pave the way for non-invasive cancer detection methods.

Introduction

Figure 1: A sampling of the large variety of highly sensitive dog noses capable of detecting trace levels of compounds.

Canine olfaction has been demonstrated as an effective method for detection of disease, including cancer, over the past three decades.[1] One of the most notable studies on the detection of prostate cancer by dogs from urine reported a mean sensitivity and specificity of 99% and 98%, respectively (2 dogs), from 362 cases and 540 healthy controls.[2] As part of a larger study aimed towards the long-term goal of replicating the dogs' early detection ability with an artificial network (ANN) deployed in conjunction with machine olfaction, CPG was asked to analyze urine samples by **headspace solid phase microextraction gas chromatography—mass spectrometry (HS SPME GC-MS)** to assess differences in the chromatograms (fingerprint patterns) of individuals with prostate

cancer and biopsy-negative controls.

Experimental

GC-MS was coupled with headspace solid-phase microextraction (HS-SPME) to analyze urine samples obtained from prostate cancer patients and biopsy-negative controls. Frozen urine samples provided to CPG were thawed to room temperature and transferred via pipette to headspace vials (Restek, Bellefonte, PA). Volatiles were extracted from the headspace of the urine with carbon wide range SPME arrows (Restek). To facilitate equilibration, the headspace vial-SPME arrow assembly was gently agitated at 172 rpm and 80°C for 30 minutes. The SPME arrow fiber was thermally desorbed in the injector of a 6890 GC system coupled with a 5973 mass spectrometer (Agilent Technologies, Palo Alto, CA). The injector was used with a 2:1 split ratio and a 2.0 mL/min split flow at 300°C. For GC separation, a ZebronTM ZB-624 column (30 m x 0.32 mm x 1.80 mm, 5% cyanopropylphenyl-94% dimethylpolysiloxane, Phenomenex, Torrance, CA) was used, and the carrier gas flow was maintained at 1 mL/min. The oven program was as follows: initial temperature of 40°C for 1.0 minutes, 10°C/min ramp up to 300°C, and 300°C isothermal for 10 minutes. The MS transfer line temperature was maintained at 240°C, and MS spectra were recorded in scan mode from m/z (mass to charge ratio) 35–500. GC-MS data was analyzed via Agilent MSD ChemStation software (E.02.02.1431) in combination with MZmine 3.6.0 open-source software.

Results

Overlays of the LC-MS chromatograms of the VOC profiles obtained from the urine of several cancer patients and biopsy-negative controls are provided in **Figure 2** and **Figure 3**, respectively. For additional visual comparison, three dimensional GCMS profiles of the urine from cancer patients and the biopsy-negative controls are shown in **Figure 4** and **Figure 5**, respectively.

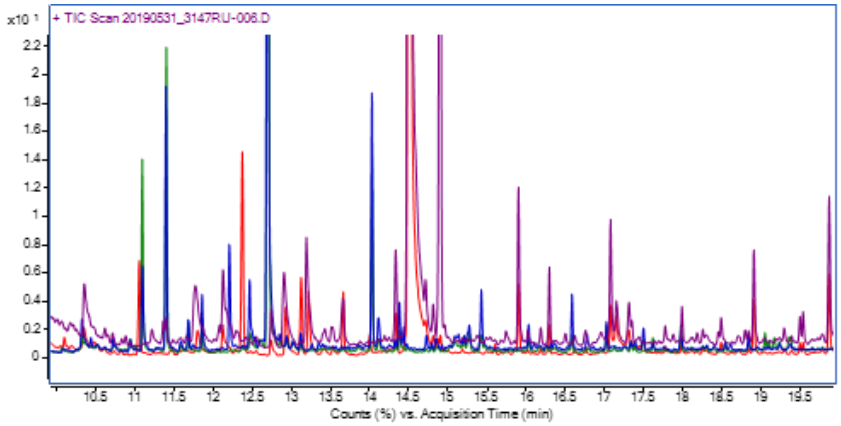

Figure 2: Overlay of the GCMS chromatograms obtained from the urine of several prostate cancer patients.

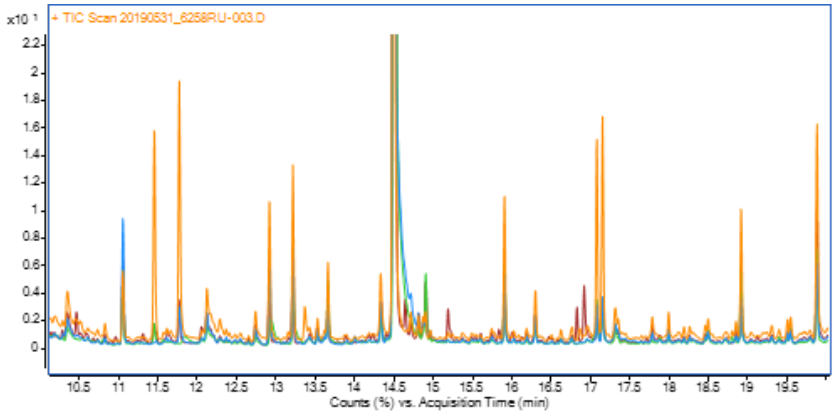

Figure 3: Overlay of the GCMS chromatograms obtained from the urine of several biopsy-negative controls.

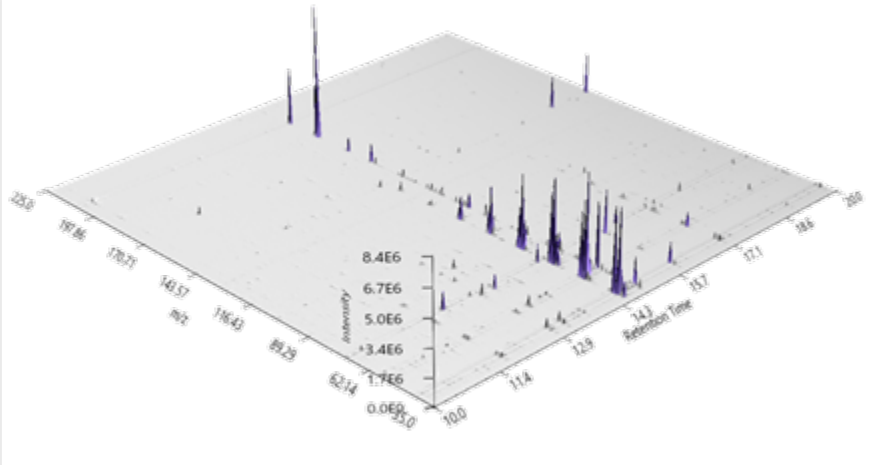

Figure 4: Representative 3D GCMS chromatogram obtained from the urine of a prostate cancer patient.

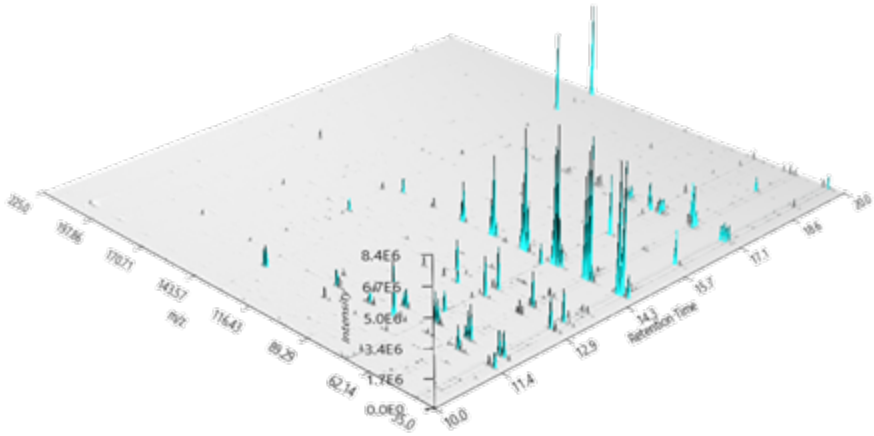

Figure 5: Representative 3D GCMS chromatogram obtained from the urine of a biopsy-negative control.

Integration with Canine Olfaction

Parallel to CPG's GC-MS analysis, the same urine samples were used to train cancer detection dogs at Medical Detection Dogs (MDD) in the UK. This approach leveraged the remarkable olfactory capabilities of dogs, which have been shown to detect prostate cancer with high sensitivity and specificity.

Artificial Neural Network Development

The Johns Hopkins and MIT researchers combined CPG's GC-MS data with the results from the trained cancer detection dogs to create an artificial neural network (ANN). This ANN was designed to emulate canine olfactory diagnostics, effectively translating the dogs' cancer detection abilities into a digital format.[3]

Significance and Future Applications

This innovative approach to cancer detection, facilitated by CPG's analytical expertise, offers several advantages:

1. **Non-invasive**: Uses urine samples instead of more invasive biopsy procedures.
2. **Early detection**: Potential for identifying cancer at earlier stages.
3. **Scalability**: Once developed, the electronic "nose" could be widely deployed.

The successful integration of CPG's analytical chemistry capabilities, canine olfaction, and artificial intelligence demonstrates the power of interdisciplinary research in tackling complex medical challenges.

Conclusions

This case study highlights CPG's role in providing critical analytical results for medical device R&D. By working with cross-disciplinary teams to analyze complex chromatographic datasets, CPG contributed significantly to the development of a novel approach for cancer detection. The trained ANN, based on CPG's GC-MS data and canine cancer diagnostics, opens up new possibilities for non-invasive, early detection of cancer.

References

1. Juge, A.E., M.F. Foster, C.L. Daigle. "Canine Olfaction as a Disease Detection Technology: A Systematic Review." *Applied Animal Behaviour Science* (2022) 253: 105664. https://doi.org/10.1016/j.applanim.2022.105664
2. Taverna, G., L. Tidu, F. Grizzi, V. Torri, A. Mandressi, P. Sardella, et al. "Olfactory System of Highly Trained Dogs Detects Prostate Cancer in Urine Samples." *Journal of Urology.* (2015) 193(4):1382–7. https://doi.org/10.1016/j.juro.2014.09.099
3. Guest, C., R. Harris, K.S. Sfanos, E. Shrestha, A.W. Partin, B. Trock, L. Mangold, R.A. Bader, A. Kozak, et al. (2021) "Feasibility of Integrating Canine Olfaction with Chemical and Microbial Profiling of Urine to Detect Lethal Prostate Cancer." *PLoS ONE* (2021) 16(2): e0245530. https://doi.org/10.1371/journal.pone.0245530

About Becky Bader, Ph.D.

Becky Bader, Ph.D., is the Associate Director of Chromatography and Biocompatibility Specialist at Cambridge Polymer Group. Prior to returning in 2023 after initially working for Cambridge Polymer Group from 2017-2020, Becky was Biocompatibility Engineering Manager at ZOLL Medical Corporation where she ensured biocompatibility and material compliance with harmonized standards and global regulatory requirements. Becky has over 20 years of experience in polymeric materials, drug delivery, medical device, and analytical chemistry. She is currently advancing analytical techniques for materials characterization at Cambridge Polymer Group.

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