Advances in Fieldable Chemical Threat Sensing

Engineered Olfactory Receptors for Chemical Detection

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Current state-of-the-art inorganic hardware sensors for biological and chemical agent detection are highly tailored for specific chemicals and cannot be used to detect compounds outside of a highly defined analyte set. With the growing threat of chemical and biological warfare, new diseases, and food borne pathogens, there is an increased need for rapid and sensitive recognition to identify such agents. Olfaction, or the sense of smell, is a detailed and nuanced selection process that organisms employ to detect and discriminate thousands of possible odorants. Specifically, humans contain over four hundred olfactory receptors while animals such as dogs and rats have close to 1,000 that when combined in different permutations can detect trillions of different smells. Olfactory receptors are G protein coupled receptors (GPCRs) that cause cells to fire an electrical action potential that can specifically detect agents (based on organic functional groups, lipids, sugars, nucleotides, and proteins) produced through organic synthesis or through biological means such as viruses, bacteria, spores, and biological toxins. Therefore, it is feasible that the unique combinations and selectivity of olfactory receptors could be comprehensively utilized to create a sensor that detects trace amounts of molecules in gas or liquid environments. Here, we modify chicken sensory neurons and HT-22 cells by adding individual olfactory receptors by transfecting with plasmids specific to each receptor. In the presence of an odorant, the cells produce an action potentials that is measured optically with calcium imaging and via a field potential measurement using a microelectrode array. Seven different plasmids encoded with individual olfactory receptors (OR2W1, OR5A1, OR5P3, OR51E1, OR2C1, and OR10G4) were used to nucleofect cell colonies. To ensure nucleofection was successful, immunohistochemistry was conducted with antibodies specific to individual olfactory receptors. Additionally, Western Blotting was conducted to determine olfactory receptors were produced. Finally, physiological measurements were completed to ensure proper function from cell colonies in the presence of specific odorants and in the presence of non-specific odorants. Western Blots show bands at ~37 kDa stained specifically for individual olfactory receptors which is corroborated with immunohistochemistry fluorescent microscope images reveal that olfactory receptors produced in HT-22 cells. Preliminary results show action potential firing from HT-22 cells nucleofected with OR2W1 and stimulated with hexanol (a specific odorant). Additionally, chicken sensory neurons nucleofected with OR2W1 shows stimulation via fluorescent calcium imaging after the additional of hexanol (a specific odorant). Analysis of modified sensory neurons that serve as a representative model for exploiting the sensitivity and selectivity of native olfactory systems can be used as rapid detection systems. Additionally, genetic engineering of unique GPCRs could yield detection of other chemicals that are mission critical that are easily created from engineered nucleotide sequences. Rapid detection of biological material can be applied to several areas of operation interest such as defense, security, force protection, Special Forces, homeland security, and medical & health capacities.

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