

COMPUGEN'S NOVEL LUNG CANCER BIOMARKER CANDIDATES

INTRODUCTION

Compugen has discovered two potential biomarkers for Lung Cancer. The first, CGEN-438 is a novel secreted splice variant form of the known transmembrane protein Delta-Like Protein 3 Precursor (DLL3), also called Drosophila Delta homolog 3 (Bulman et. al, Nature Genetics 2000 24: 438 – 441). The second, CGEN-6 is a secreted splice variant form of the known secreted Gastrin Releasing Peptide precursor. Clinical evidence suggests that these two novel peptides, CGEN-438 and CGEN-6, may serve as novel serum biomarkers for the diagnosis of Small Cell Lung Cancer (SCLC). CGEN-438 may also be used in a biomarker combination test for the diagnosis of Non-Small Cell Lung Cancer (NSCLC). A non-invasive screening test for Lung Cancer with high sensitivity and specificity would provide a significant medical contribution to the early detection of Lung Cancer. Compugen is currently seeking partners with a strategic focus in Lung Cancer diagnosis and access to clinical samples for further validation and commercialization of both CGEN-438 and CGEN-6.

BIOMARKER DISCOVERY APPROACH

Biomarker Identification

CGEN-438 and CGEN-6 were identified through Compugen's Immunoassay Discovery platform which consists of a database of novel predicted mRNA's and proteins. This platform utilizes a combination of computational algorithms and biological data to identify differential expression of mRNAs and proteins in specific epithelial cancers. CGEN-438 and CGEN-6 were further validated experimentally by establishing the presence of each as a circulating serum protein and analyzing the differential expression of the serum protein in healthy and diseased populations. The collective data suggests that the two candidates may serve as biomarkers for Lung Cancer.

Characterization of CGEN-438 and CGEN-6 Variants

CGEN-438 is a novel splice variant form of the DLL3 gene. However, unlike the wild-type, the splice variant (CGEN-438) is a secreted peptide which doesn't share a sequence similarity to DLL3. Given the differential characteristics of the Compugen variant, it may serve as a biomarker for serum-based immunoassay tests, unlike its membrane wild-type form, DLL3.

CGEN-6 is a novel splice variant form of the GRP (Gastrin Releasing Peptide Precursor) gene. Today, there are three known isoforms of the wild type GRP protein, all of which are secreted proteins. CGEN-6 is a novel secreted splice variant protein with a unique combination of amino acids derived from various known GRP isoforms.

CGEN-438 AND CGEN-6 AS SERUM BIOMARKERS FOR LUNG CANCER

RNA Profile in Lung Cancer Tissues

The mRNA expression pattern of the two molecules, predicted by Compugen's Immunoassay Discovery platform, was tested using quantitative RT-PCR analysis on a panel of cancerous and healthy lung tissues and was shown to be specific for small cell lung carcinomas (Fig 1 and Fig 2). The columns correspond to various RNA samples isolated from about 50 Lung Cancer tissues, including adenocarcinoma, squamous cell carcinoma, large cell carcinoma, small cell carcinoma and about 13-20 normal lung tissues. The Y-axis represents expression relative to the median of the normal samples. Significant over expression is defined by a greater than 5-fold increase in relative expression.

Figure 1: mRNA expression pattern of CGEN-438 in cancerous lung samples relative to normal samples.

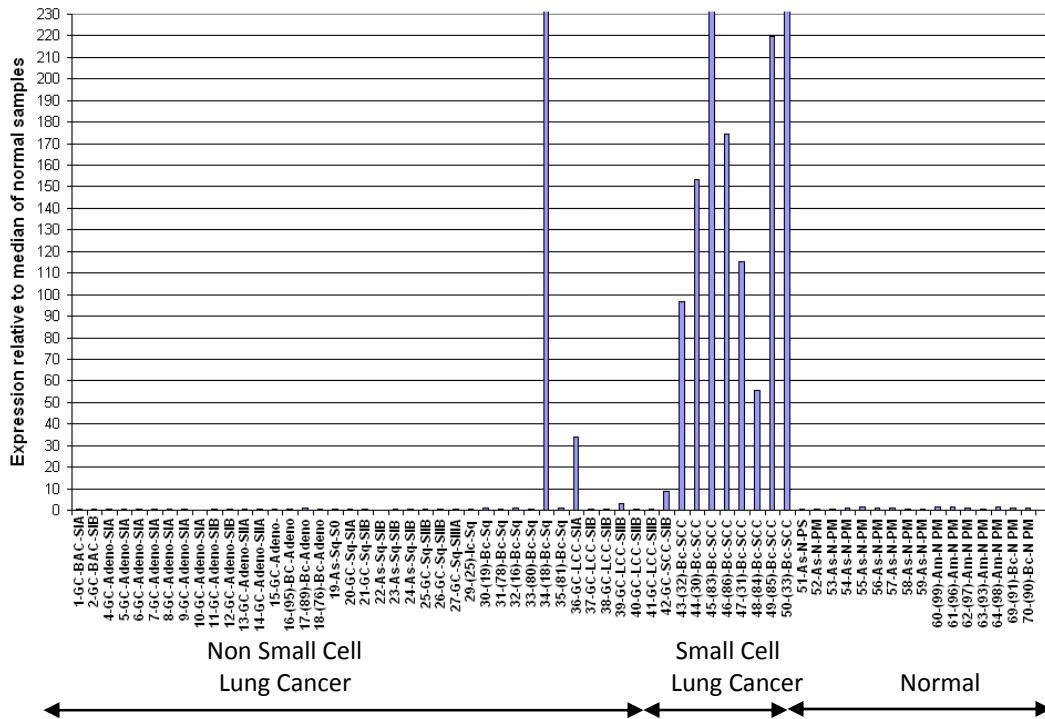
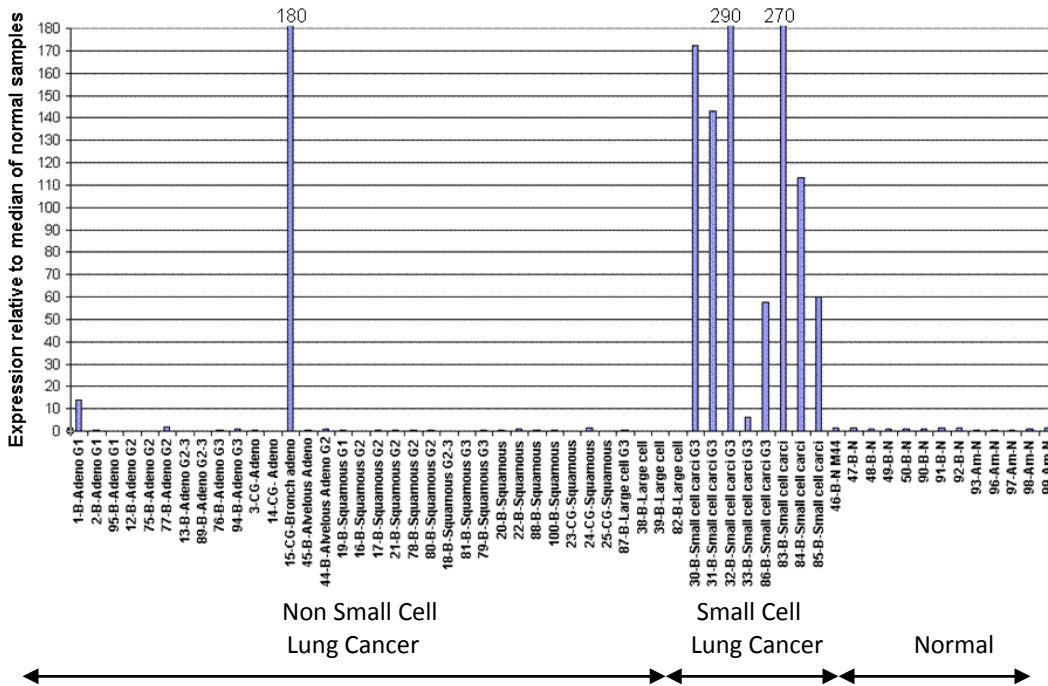


Figure 2: mRNA expression pattern of CGEN-6 in cancerous lung samples relative to normal samples.



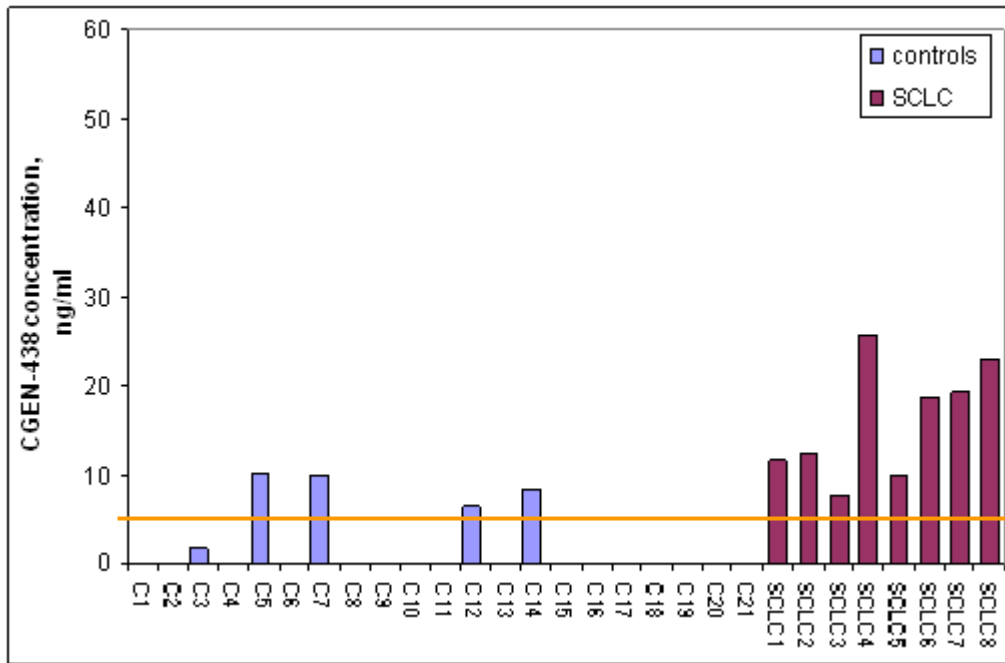


Protein Profile in Lung Cancer Serum Samples

The protein expression pattern of CGEN-438 and CGEN-6 in serum samples of diseased and healthy individuals was tested using polyclonal antibody preparations developed to specifically recognize each of these molecules.

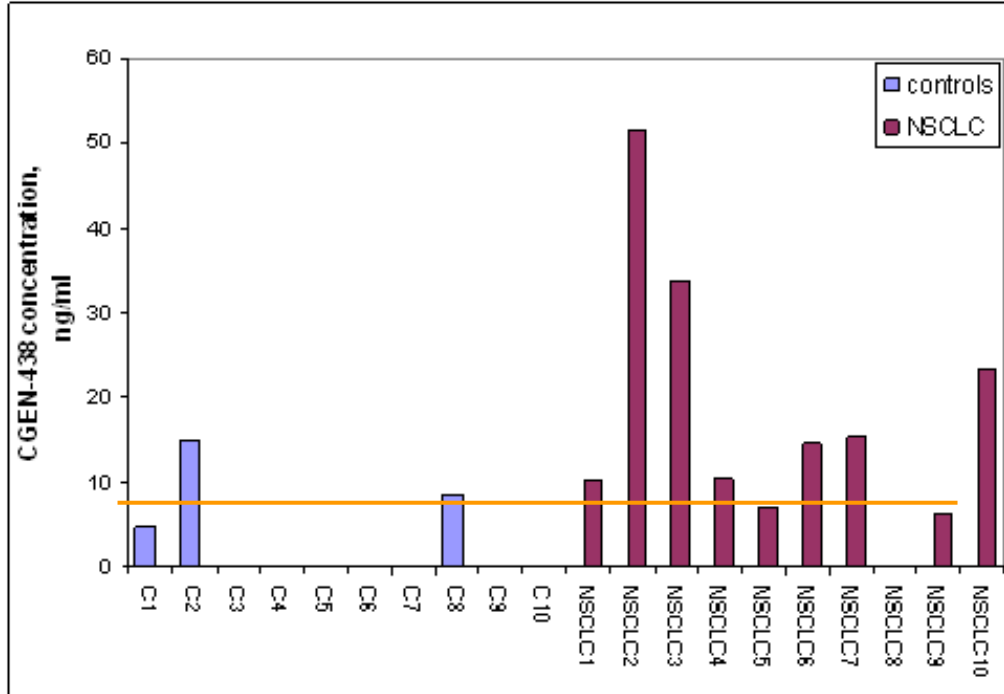
CGEN-438 in serum: Using an antibody competitive ELISA test, the level of CGEN-438 was analyzed in the sera of eight SCLC patients and 21 controls. Serum protein was detected in all eight SCLC patients and in four out of the 21 controls, above a given cut-off (Figure 3). CGEN-438 concentrations in SCLC patients were 16 ± 7 ng/ml relative to 2 ± 4 ng/ml in the control population.

Figure 3: CGEN-438 concentration in serum samples of SCLC and controls.



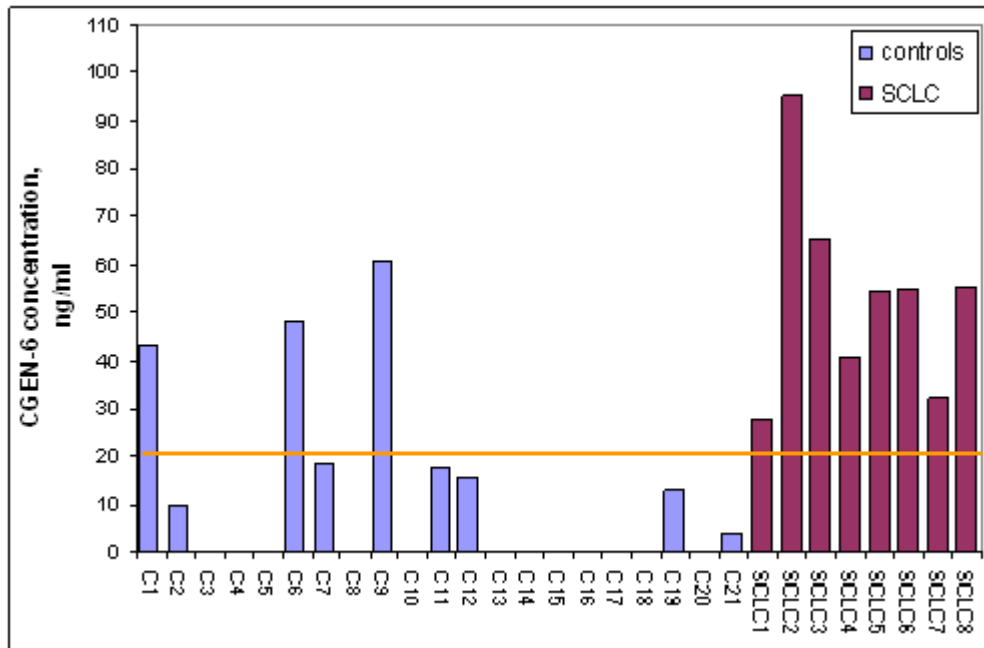
The same ELISA format was employed to detect the level of CGEN-438 in Non-Small Cell Lung Cancer (NSCLC) patients. For this purpose, serum samples of 10 NSCLC and 10 additional new controls were tested. Out of 10 NSCLC patient's sera tested, nine showed detectable levels of CGEN-438, above a given cut-off (Figure 4). CGEN-438 concentrations were 17 ± 15 ng/ml relative to 3 ± 5 ng/ml in the control population.

Figure 4: CGEN-438 concentration in serum samples of NSCLC patients and controls.



CGEN-6 in serum: Using an antigen competitive ELISA test, CGEN-6 was detected in the sera of all eight SCLC patients and only three out of 21 controls, above a given cut-off (Figure 5). The control samples showing positive CGEN-6 values were different than those showing positive CGEN-438 values, implying that a combination test using both markers may enhance assay specificity.

Figure 5: CGEN-6 concentration in serum samples of patients and controls.



SUMMARY

Compugen has identified two novel secreted proteins that may serve as serum biomarkers for the diagnosis of Lung Cancer. CGEN-438 was detectable at higher protein concentrations in the serum samples of the SCLC and NSCLC patients as compared to those of the control samples. As a molecule that can be found in the bloodstream, CGEN-438 has the potential to serve as a better biomarker for serum-based immunoassay tests than its membrane wild-type form, DLL3. CGEN-6 demonstrated a similar differential expression pattern in serum samples of SCLC and may improve CGEN-438 assay performance in a combination test for Small Cell Lung Cancer. Compugen is seeking both development and commercialization partners who have a strategic focus in the area of Lung Cancer diagnosis. Compugen aims to discover additional novel candidate biomarkers for additional types of epithelial cancers, mainly colorectal, ovarian and breast cancers.