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Possible application of MiRNA expression used as a biomarker for malignant cell transformation

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With the markedly increasing numbers in cancer diagnostics and progression, it has become imperative for researchers to determine biomarkers that could serve as early detection devices. The focus lies primarily on microRNA (miRNA) expression levels, secretory mechanisms and their role in malignant cell transformation. A recent study showed that miRNAs are exported from malignant cells in particles that are specific to that miRNA. Through sequencing of malignant cell RNA, researchers have found that sorting s-miRNA and n-miRNA is achieved through a sequence-specific mechanism; therefore, this indicates that these two proteins will perform different functions and could unveil specific biomarkers associated with malignant cell transformation. The s-miRNA will affect the cell-to-cell communication and information transfer; therefore, these proteins will alter the signaling pathways of the cells they come into contact with. Secretory mechanisms and intercellular transfer also dictate the signaling pathways employed by the miRNAs. Secretory mechanisms and intercellular transfer also dictate the signaling pathways employed by the miRNAs. The type of pathway used to secrete miRNA is significant because it determines whether extracellular miRNA will act on a cell the same way that cellular miRNA exerts its effects. Further progress in this research needs to be conducted in order to tie all the details together and determine how expression of miRNA affects malignant cell transformation as a whole.

Introduction

Cancer is among the most researched topics in science today. With the markedly increasing numbers in cancer diagnostics and progression, it has become imperative for researchers to determine biomarkers that could serve as early detection devices. The focus lies primarily on microRNA (miRNA) expression levels, secretory mechanisms and their role in malignant cell transformation. MiRNAs are short, non-coding molecules of RNA that are post-transcriptional regulators that bind to target messenger RNA (mRNA). This binding to the mRNA usually results in repression or silencing of the target gene. Studies have shown that miRNA plays a key role in regulation of several cellular processes such as: growth, reproduction, differentiation, and apoptosis (Wang and Sen, 2007). Since miRNAs are associated with a variety of cellular processes and regulation, increased expression levels of miRNA could indicate the presence of malignant cells and tumors. Previous studies have shown that abnormal miRNA expression has been detected and observed in most every human cancer signifying it as a common event associated with the genetics of cancer cells. Emphasis has been placed on miRNA expression as a possible biomarker for detecting malignant breast cancer cells.

To further determine the degree to which this protein can be used as a diagnostic marker, we must investigate the secretory mechanisms and the export selectivity of the miRNA from the malignant cells. Cell release of miRNAs is achieved through several different transport vesicles and protein complexes. Exosomes are pivotal in miRNA research, for theses vesicles contain miRNA and are thought to transfer material to other cells. An important part of cancer detection and proliferation is cell-to-cell communication and exosomes contribute to the communication and transmission of information from

one cell to another. Through this transfer of material, from one cell to another, exosomes have shown to play a large role in tumor progression and proliferation (Duelli *et al.,* 2005). Recent breast cancer research has suggested that differences in the vesicles that transport miRNA could possibly indicate the association of specific particles, namely cancer cell, and selectivity of the protein.

Another important area of study is the secretory mechanisms and intercellular transfer of miRNA information. Although the function of these mechanisms and significance of miRNA in the blood is not completely understood, scientists are hopeful that this could be used as early detection diagnostic tools. Once the regulation of these mechanisms are determined, it will be easier to observe the regulation roles the miRNA has on both malignant and healthy cells. It was found that the release of miRNAs is actively controlled through ceramide-dependent machinery associated with exosome secretion (Iguchi *et al.,* 2010). Synthesis of ceramide is regulated by the enzyme sphingomyelinase 2 (nSMase2). This enzyme also triggers the secretion of exosomes (Kosaka *et al*., 2010). Through manipulations of this enzyme, studies have shown the down-regulation or up-regulation of miRNA secretion and the pathways the proteins employ. These pathways are significant because the physiology of miRNAs are made relevant and could possibly be applied to therapeutic treatment of malignant cell transformation. This study also supported the hypothesis that released miRNA, whether selective or neutral, is integrated with a specific breast cancer cell population based on size and density of that miRNA.

Recent Progress

A recent study showed that miRNAs are exported from malignant cells in particles that are specific to that miRNA. Selectively released miRNAs (s-miRNA) exhibit increased export from cancer cells. These miRNAs are packaged and released from structures different from those structures that carry neutrally released miRNAs (n-miRNAs) (Palma *et al*., 2012). These n-miRNAs are not released from cancer cells, which indicate that the s-miRNAs are associated with nucleosomes and exosomes. Nucleosomes are often released in response to apoptosis. This is significant because many cancer cells frequently experience apoptosis, so increased amounts of circulating nucleosomes is likely to indicate malignant cells. These results indicate that both exosome and nucleosome detection in blood are strong possible biomarkers that malignant cells are present in the body. Selective release of the miRNA from breast cancer cells is guarded by malignant cell transformation, meaning several different types of miRNA could indicate the presence of cancer cell proliferation. Recent progress has been made in regards to the finding that s-miRNAs and n-miRNAs are exported from separate and distinct vesicle types. The identification of these "sub-populations" of vesicles allows for determination of cellular targets as well as cancer prognosis based on affiliation of s-miRNA with their specific vesicle. Through sequencing of malignant cell RNA, researchers have found that sorting s-miRNA and n-miRNA is achieved through a sequence-specific mechanism; therefore, this indicates that these two proteins will perform different functions and could unveil specific biomarkers associated with malignant cell transformation. These findings were supported by other studies.

Recent studies show that release of malignant cell related miRNAs suggest a role for selective export in cancer cell transformation (Palma *et al.,* 2012). The s-miRNA will affect the cell-to-cell communication and information transfer; therefore, these proteins will alter the signaling pathways of the cells they come into contact with. Secretory mechanisms and intercellular transfer also dictate the signaling pathways employed by the miRNAs. The authors concluded that miRNA secretion is controlled by the enzyme nSMase2, which is also the rate-limiting enzyme for the synthesis of ceramide (Kosaka *et al*., 2010). When comparing the different types of miRNA secretory pathways, it was noted that secretion of the protein only employs this ceramide secretory pathway as opposed to another type called ESCRT system. The type of pathway used to secrete miRNA is significant because it determines whether extracellular miRNA will act on a cell the same way that cellular miRNA exerts its effects. The exosomes used to transport miRNA through the ceramide pathway are important for allowing horizontal propagation of malignant cell transformation (Kosaka *et al.,* 2010). These pathways are used to aid in determining the type of cancer cells present by observing changes in the pathways themselves. This is yet another significant finding that could potentially contribute to understanding the role s-miRNA, and other types of miRNA, plays in the maintenance system against cancer proliferation and progression. Another study supported these results by determining that miRNA expression in solid tumors characterizes cancer targets (Voliana *et al.,* 2006). Further progress in this research needs to be conducted in order to tie all the details together and determine how expression of miRNA affects malignant cell transformation as a whole.

Discussion

A thorough understanding regarding the significance of circulating miRNAs in cell communication is an integral part of cancer cell biomarker identification. Once this is understood, scientists can use this information to synthesize therapeutic treatments for malignant cell transformation via different types of miRNA. An important finding showed that malignant cell s-miRNAs

and n-miRNAs are exported from completely separate and distinct vesicles from one another as well as different vesicles used to export normal, healthy cells. Though, further studies need to address malignant cell s-miRNA and the proteins correlation with other specific vesicles. The question still remains as to which, if any, specific microvesicles associate with different kinds of miRNA exported from malignant cells.

Much of the studies focus on the effects up-regulation of miRNA has on the biology of the cell. This is because it is easier to observe the presence of a chemical or cell secretion as opposed to the absence of a marker (Wang and Sen, 2011). Future studies will need to focus more on observing the down-regulation of the miRNA to see the complete consequences of the differing expression levels. This will allow for researchers to further progress their understanding of potential therapeutic applications this may harbor.

Several questions remained unanswered, as with most research, but increased analysis and investigation into the intricate specifics associated with circulating miRNAs is needed. This will help to determine the miRNAs probability as a reliable biomarker for malignant cell transformation. Practical implications of this research could be put towards early cancer detection as well as treatment and termination of cancer cells by antagonistically altering the cell that is undergoing transformation.

References

Duelli,D.M., Hearn,S., Myers,M.P. and Lazebnik,Y. (2005) A primate virus generates transformed human cells by fusion. J. Cell Biol., 171, 493–503.

Iguchi H., Kosaka N., Ochiya T. 2010 Secretory microRNAs as a versatile communication tool. *Journal of Biological Chemistry, 3(5):* 478-481.

Kosaka N., Iguchi H., Yoshioka Y., Takeshita F., Matsuki Y., Ochiya T. 2010 Secretory Mechanisms and Intercellular Transfer of MicroRNAs in Living Cells. *Journal of Biological Chemistry, 285 (23):* 17442-17452.

Palma J., Yaddanapudi S., Pigate L., Havens M., Jeong S., Weiner J., Weimer K.M., Stern B., Hastings M., Duelli D. 2012 MicroRNAs are exported from malignant cells in customized particles. *Nucleic Acids Research* 1-14.

Volinia S., Calin G., Liu C., Ambs S., Cimmino A., Petrocca F.,Visone R., Iorio M., Roldo C., Ferracin M., Prueitt R., Yanaihara N., Lanza G., Scarpa A., Vecchione A., Negrini M., Harris C., and Croce C. 2006. A microRNA expression signature of human solid tumors defines cancer gene targets. *PNAS 103(7)* 2257-2261.

Wang J. and Sen S. 2011 Micro RNA functional network in pancreatic cancer: From biology to biomarkers of disease. *J. Biosci.* 36: 481-491.

