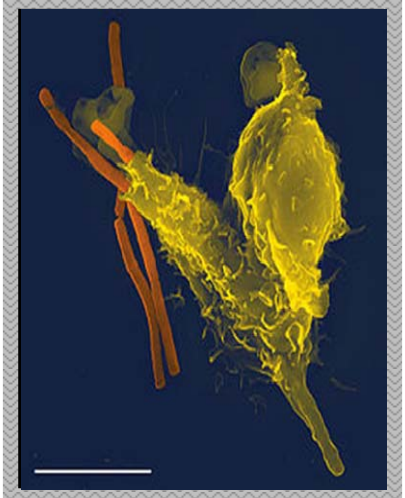

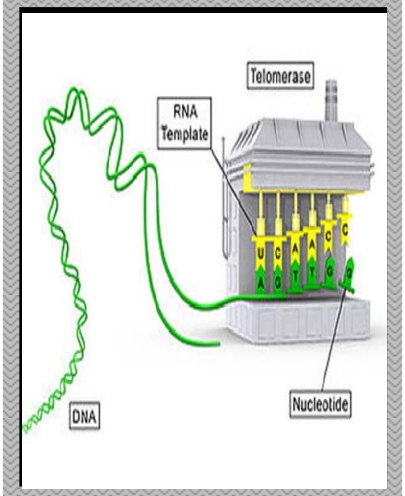

EMGEN Newsletter

Vol. 3, Issue 10, April-May, 2010

INSIDE THIS ISSUE:

Eastern Mediterranean Health Genomics and Biotechnology Network (EMHGBN) was created in 2004 with collaboration of representatives of selected centre of excellence in (health related) molecular biology, biotechnology & genomics in the Eastern Mediterranean region by recommendations and efforts of WHO/EMRO.

Address:
Biotechnology building, #69, Pasteur Ave., Pasteur Institute of Iran
Tehran, Iran, 13164
Tel: +98-21-66954324
Fax: +98-21-66465132
E-mail: emhgbn@gmail.com, secretariat@emhgbn.net
Website: www.emhgbn.net

Prepared by: Elham Jahangiri, Dr. Afshin Peirovi
Page design: Elham Jahangiri
Helped by: Behrad Shaghaghi, Shiva Shajari
Editor: Dr. S. Sardari

Modulations of Cell Cycle Checkpoints during HCV-Associated Disease

The article entitled "Modulations of Cell Cycle Checkpoints during HCV-Associated Disease" reports about the changes in regulation of cell cycle proteins that could significantly contribute to HCV disease evolution and following hepatocellular carcinoma (HCC). The study was done by Saira Sarfraz, Saeed Hamid, Syed Ali, Wasim Jafri and Anwar A Siddiqui. Corresponding author of this paper, Dr. Anwar A Siddiqui is Professor and Associate Dean for Research of Department of Biological and Biomedical Sciences Aga Khan University of Pakistan, and the paper was published in Journal of BMC Infect Disease. 2009; 9.125

Dr. Saira Sarfraz

The disease burden caused by hepatitis C virus (HCV) infection entails a strong public health trouble. A new report suggests its prevalence in geographically various regions, affecting around 130 million people worldwide (Alter 2007). In Pakistan, the frequency of HCV infection is estimated to be about 6% in general population (Hamid, Umar et al. 2004). These figures are frightening, as patients currently asymptomatic with relatively mild disease may finally progress to complications of chronic liver disease like, end-stage liver disease, cirrhosis and hepatocellular carcinoma (HCC).

Liver hurt is commonly supposed to be started by the death of infected cells afflicting problems of inflammation, regenerative hepatocyte proliferation and fibrosis in the surroundings (Tillmann, Manns et al. 2005). Together, these events keep the dividing hepatocytes susceptible to cellular insult and therefore putting them at a bigger risk of acquiring mutations. The cell cycle regulators controlled the molecular events through proliferation tightly. In the cell cycle, perfect DNA replication and chromosome separation are done by means of quality control checkpoints which are active during all phases of cell cycle namely Gap1 (G1), DNA synthesis (S), Gap2 (G2) and Mitosis (M). In reaction to cellular insults, for example DNA damage and oxidative stress, inhibitory checkpoint regulators are turned on at G1, S and G2 phases and stop the development of cell cycle (Sherr 1996). These regulators consist of DNA damage response genes (RAD1, HUS1, RAD9, ATM, ATR and p53) (Houtgraaf, Versmissen et al. 2006) and members of CDK inhibitor family, p16 (CDKN2A), p15 (CDKN2B), p21 (CDKN1A) and p27 (CDKN1B) (Sherr and Roberts 1999). In the subsequent M phase, mi-

totic checkpoint proteins of MAD and BUB families are liable for a suitable chromosomal separation in the period of cell division (Molinari 2000).

Studies done on hepatic biopsies from chronic hepatitis C patients have explained an arrested cell division cycle in hepatocytes (Werling, Szentirmay et al. 2001; Marshall, Rushbrook et al. 2005). In our earlier study, we have proved this arrest in hepatocytes distinguished by over expression of CDK inhibitor p21 that was further explained by screening changed expression of tumor suppressor p53 and apoptotic proteins Caspase-3 and Bcl -2 (Sarfraz, Hamid et al. 2008). Considering the basic function done by cell cycle proteins in managing cell proliferation, altered regulation of these proteins could considerably contribute to HCV disease progression and subsequent HCC. Our study aimed to recognize the altered expression of cell cycle genes expression with respect to early and advanced disease of chronic HCV infection.

By freshly frozen liver biopsies, mRNA levels of eighty four cell cycle genes were studied in RNA samples separately pooled from patients with early HCV, advanced HCV and commercial preparation of human usual liver RNA. Studies illustrated early HCV disease with increased mRNA levels of cell proliferation genes (MCM2, CCNE1) as well as cell cycle inhibitor genes (CDKN1A/p21, CDKN1B/p27). These transcriptional modifies imply a mixed population of cells in the infected livers of chronic hepatitis C, some of which are in proliferating state while others in an arrested state. In advance HCV, DNA damage response genes (RAD1, HUS1, RPA3) were up-regulated signifying raised DNA damage whereas those associated with chromosomal stability (MAD2L1, CDC34) were down-regulated. Taken together these effects are suggestive of deregulated cell cycle in chronic HCV infection that is considered as a consistent happening in HCC.

To associate mRNA levels with respective protein levels, four genes CDKN1B/p27, CDKN2B/p15, MAD2L1 and KNTC1 with considerable changes in mRNA levels (> 2-fold, p-value < 0.05) were picked and their protein expressions were studied by immunohistochemistry or western blotting. These genes were also chosen on the basis of their crucial role in cell cycle regulation. Increased expression of CDK inhibitor protein p27 was watched by immunohistochemistry that was consistent with its mRNA level detected in early HCV. Interestingly, a converse relationship between p27 protein expression and progression of liver fibrosis from F0 to F4 was found. It would be related to mention here that mouse models of chronic injury induced-liver tumorgenesis have explained that loss of p27 in advanced stages of disease promotes tumor cell proliferation (Sun, Ren et al. 2007). These examinations indicate the potential of p27 as a reporter for prognostic implications in chronic HCV disease. In contrast to p27, immunohistochemical evaluation of G1 phase inhibitor p15 showed no considerable difference in advanced HCV as compared to early HCV that conflicts with the changes observed at mRNA level. This inconsistency might have been because of declined stability of p15 protein in ad-

vanced disease stages that increased the transcription through a regulatory feedback system to keep its levels. Another elucidation for this irregularity could be the use of pooled samples of whole liver lysates for mRNA expression whereas immunohistochemistry was acted on individual formalin fixed paraffin embedded biopsies. Expression of p15 was also viewed in one of the non-viral hepatitis patients that indicate; it might be a general change in chronic liver diseases other then hepatitis C. Nevertheless, this analysis was made on a small number of patients and further studies are needed to support this postulation.

Between the differentially expressed mitotic checkpoints genes in early and advanced HCV, expression of KNTC1 and MAD2L1 proteins were studied by western blotting. KNTC1, as well known as "rough deal protein" (ROD) is element of a compound involved in elaborating an inhibitory sign for improper chromosomal alignment in cell separation (Chan, Jablonski et al. 2000), as member of Mad2 family, MAD2L1, participates in inhibiting Anaphase promoting complex (APC) (Fang, Yu et al. 1998) from ubiquitinating securin, whose degradation is a prerequisite for sister chromatid division and mitosis. Results illustrated increased levels of both of these proteins in early HCV that is indicative of mitotic checkpoint activation or mitotic arrest in the liver cells. This is constant with the down-regulation of CDC16 that is a component of APC and promote chromosomal segregation. Significantly, KNTC1 expression considerably decreases in advanced stages of HCV group as compared to early HCV. This mitotic protein does not show to be hepatitis C specific as its expression was also remaindered in non-viral hepatitis. In comparison, expression of MAD2L1 appears to be HCV-specific as it was found completely in chronic hepatitis C patients with a modest decreasing in advanced HCV group. The function of KNTC1 in human cancers is not well studied, but an only report shows mutation in KNTC1 gene in colorectal carcinoma (Wang, Cummins et al. 2004). Alternatively, reduced expression of MAD2 gene has been informated in mammalian cells with loss of mitotic checkpoint and later chromosomal instability (Michel, Liberal et al. 2001; Wang, Jin et al. 2002). Therefore, low expression of these mitotic checkpoint regulators in advanced HCV might mirror loss of mitotic checkpoint manage that could render cells to chromosomal instability. This hypothesis is too supported by a prior study that proved existence of near-aneuploidy DNA content in liver samples from chronic hepatitis C patients (Werling, Szentirmay et al. 2001).

Conclusion

The study of cell cycle regulators exhibited altered expression of G1/S and M phase inhibitors in chronic HCV infection that either seize or delays the progression through G1 and S phase as well as M phase, latter being not reported prior in chronic hepatitis C patients. These cell cycle perturbations might be a consequence of increased DNA damage or constant inflammation throughout viral infection that in turn could raise genetic in-

stability. This in turn could cause to aberrant DNA replication and cell division that might incite cell transformation and development of HCC. The information existing here highlights the potential of these proteins as early markers to recognize patients with high risk of cell transformation and HCC development.

References:

1. M. J. Alter, Epidemiology of hepatitis C virus infection: World J Gastroenterol (2007). Vol. 13: 2436-41.

2. G. K. Chan, S. A. Jablonski, et al. Human Zw10 and ROD are mitotic checkpoint proteins that bind to kinetochores: Nat Cell Biol (2000). Vol. 2:944-7.

3. G. Fang, H. Yu, et al. The checkpoint protein MAD2 and the mitotic regulator CDC20 form a ternary complex with the anaphase -promoting complex to control anaphase initiation: Genes Dev (1998). Vol. 12: 1871-83.

4. S. Hamid, M. Umar, et al. PSG consensus statement on management of hepatitis C virus infection: J Pak Med Assoc (2004). Vol. 54: 146-50.

5. J. H. Houtgraaf, J. Versmissen, et al. A concise review of DNA damage checkpoints and repair in mammalian cells: Cardiovasc Revasc Med (2006). Vol. 7: 165-72.

6. A. Marshall, S. Rushbrook, et al. Relation between hepatocyte G1 arrest, impaired hepatic regeneration, and fibrosis in chronic hepatitis C virus infection: Gastroenterology (2005). Vol. 128: 33-42.

7. L. S. Michel, V. Liberal, et al. MAD2 haplo-insufficiency causes premature anaphase and chromosome instability in mammalian cells: Nature (2001). Vol. 409: 355-9.

8. M. Molinari, Cell cycle checkpoints and their inactivation in human cancer: Cell Prolif (2000). Vol. 33: 261-74.

9. S. Sarfaraz. S. Hamid, et al. Altered expression of cell cycle and apoptotic proteins in chronic hepatitis C virus infection: BMC Microbiol (2008). Vol. 8: 133.

10. C. J. Sherr, Cancer cell cycles: Science (1996). Vol. 274: 1672-7.

11. C. J. Sherr, J. M. Roberts, CDK inhibitors: positive and negative regulators of G1-phase progression: Genes Dev (1999). Vol. 13: 1501-12.

12. D. Sun, H. Ren, et al. Loss of p27Kip1 enhances tumor progression in chronic hepatocyte injury-induced liver tumorigenesis with widely ranging effects on Cdk2 or Cdc2 activation: Carcinogenesis (2007). Vol. 28: 1859-66.

13. H. L.Tillmann, M. P. Manns, et al. Merging models of hepatitis C virus pathogenesis: Semin Liver Dis (2005). Vol. 25: 84-92.

14. X. Wang, D. Y. Jin, et al. Significance of MAD2 expression to mitotic checkpoint control in ovarian cancer cells: Cancer Res (2002), Vol. 62: 1662-8.

15. Z. Wang, J. M. Cummins, et al. Three classes of genes mutated in colorectal cancers with chromosomal instability: Cancer Res (2004). Vol. 64: 2998-3001.

16. K. Werling, Z. Szentirmay, et al. Hepatocyte proliferation and cell cycle phase fractions in chronic viral hepatitis C by image analysis method: Eur J Gastroenterol Hepatol (2001). Vol. 13: 489-93.

Green Fluorescent Protein

Green fluorescent protein or GFP refers to the protein that isolated from the jellyfish *Aequorea victoria (figure 1)*. Wild type GFP absorbs light at 395 and 470nm. It absorbs blue light with calcium activated photoprotein, and emits green fluorescence at 509nm. The reason of both bioluminescence and GFP fluorescence in jellyfish *Aequorea* is unknown, but this protein becomes a standard tool for observing the various mechanisms of cells, scientists can use it as a beacon.

The three researchers, Osamu Shimomura, Martin Chalfie and Roger Tsien, were awarded the 2008 Nobel Prize in chemistry, "for the discovery and development of green fluorescent protein". They witnessed the rapid development of tools based on the wtGFP, it's siblings from other organisms and engineered variants of members of the "GFP family" of proteins.

Figure1. The edges of the skirt (umbrella) of this specie of jellyfish emit a green light.

Structure of GFP

The primary structure of wtGFP is a chain of 238 amino acids (26.9 kDa). The secondary structure is a series of helices and pleated sheets that is caused by hydrogen-bonding within the chain. The tertiary structure of wtGFP comprises 11 stranded ß- sheet, threaded by a α-helix and short helical segments at the ends of the cylinder.

The chromophore is in the α-helix. The barrel structure keeps the chromophore away from solvents and let the fluoresce to be near the point at which the protein is denatured by factors such as heat or pH. The tripeptide Ser65-Tyr66-Gly67- in the primary structure of unfolded or denatured GFP does not show any striking feature. In the native conformation of protein, these three amino acids are forced into a sharp turn. At this

position, GFP does not fluoresce but in the presence of molecular oxygen, the α–β bond of Tyr66 is subsequently dehydrogenated into conjugation with the imidazolinone (Gly67) which results in maturation of the GFP chromophore to its fluorescent form.

Family of GFP

Because of the potential of widespread usage and the evolving needs of researchers, many different mutants of GFP have been engineered. Some mutations have been improved the spectral characteristics of GFP (increased fluorescence, rapidly maturing, photo stability), like EGFP (enhanced GFP) that is a form of GFPs used in mammalian cells. Many other mutations have been made, including color mutants, in particular blue protein, cyan fluorescent protein and yellow fluorescent protein derivatives. The latest efforts in jellyfish variants have led to new and improved monomeric RFP, YFP, GFP, CFP and BFP variants.

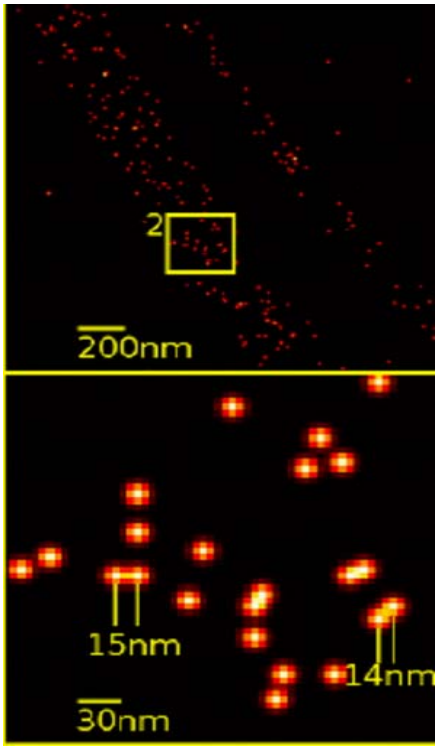

Figure2. The yellow fluorescent protein (YFP) detection in a human cancer cell (standard distance is 5 nm).

General applications of GFP

GFP's fluorescence needs no other cofactor, only requires oxygen and is not toxic for cell, these specifications make GFP so useful as a bio marker. In cellular Molecular biology, GFP has been used as cell marker, reporter gene, fusion tag, transcription factor of dimerization, calcium sensitivity and etc. GFP fluorescence can be used to quantitatively monitoring gene expression *in vivo*, *in situ*, and in real time.

The GFP derivatives allow the monitoring in time and ever-increasing number of phenomenon in living cells and organisms like gene expression, protein dynamics and localization, protein-protein interactions, pharmaceutical drug discovery ,chromosome replication and organization, cell division, intracellular transport pathways, organelle inheritance and has been used to make biosensors in modified forms. While most small fluorescent molecules like FITC (fluorescein isothiocyanate) are too phototoxic for living cells, fluorescent proteins like GFP are usually much less harmful when illuminated in live cells. The most common use of GFP has been to monitor the location, movement and chemical reactions.

GFP as a reporter gene

By evaluating the amount of mRNA or protein gene expression can be recognized, but these procedures are difficult and destructive. Reporter gene can solve this problem. A reporter is a gene. Scientists link it to a regulatory sequence of another gene and is used in molecular biology, biomedical, pharmaceutical research and biochemistry. When this gene is expressed, produces a signal, this product is easy to measure. These genes are usually used in positions in which the product of some gene of interest is difficult to assay quantitatively. The reporter gene is attached to the gene of interest and produces a gene fusion. These two genes are under the same promoter factors and are transcribed into a single mRNA. The mRNA is then translated into protein. In these cases it is significant that both proteins be able to fold into their active conformations and interact with their substrates despite being fused. There are different reporter genes. Here, we describe some of them:

A: The Escherichia coli lacZ gene is used as a first reporter gene. lacZ gene encodes a β-galactosidase protein, that has been extensively characterized genetically and biochemically. The major problem with this reporter gene is the background level of the β-galactosidase enzyme in plants.

B: Another reporter gene is the chloramphenicol acetyltransferase (CAT) gene. It was successfully introduced into different eukaryotic organisms, such as yeast, plants and etc. This gene was a suitable reporter for some but not all plant species, but there are unknown inhibitors in some plant species. This reduced the levels of expression of the bacterial CAT in transgenic plants. This problem limited the applications of CAT as a marker

gene for plant transformation.

C: A reporter gene that overcomes the problems with the cat and lacZ genes is (uidA) the E. coli β-glucuronidase (GUS) gene. For the low content or lack of endogenous GUS activity in most plants, this gene became one of the most widely used reporters in eukaryotic cells. Another advantage of this reporter gene is easily quantifying by using a fluorometric assay but its detection assay is destructive.

D: One gene has been very successful as a gene reporter and don't have these problems is Green Florescent Protein. This gene can be introduced into organisms by injection with a viral vector, breeding, or cell transformation and maintained in their genome. It has been expressed in many bacteria, yeast and other fungi, fish, plant and mammalian cells like human. In cells where the gene is expressed, and the proteins are produced, GFP is produced at the same time. Only those cells in which the target proteins are produced or the tagged gene is expressed will fluoresce when studied under fluorescence microscopy. This reporter can be used to assay for the expression of interest gene that may generate a protein which has immediate effect or little obvious on the cell culture or organism.

For achieving to plasmid-reporter gene, the white colonies analyzed by lysis of the bacteria and purified the plasmid construct with gel chromatography or ultracentrifugation on cesium chloride (CsCl) gradients. GFP expression can be followed over extended periods of time. It is noticeable that GFP expression measured as GFP intensity. It was first studied on several times for each GFP construction. The model of GFP expression in general was similar. Expression has 3 phases. In initial period, there isn't any expression, then this followed by a logarithmic raise in intensity, and in final there is a fixed phase after reaching maximum levels of expression.

By using a spectrophotometer or digital imaging, the GFP expression can be easily distinguished in transgenic cells or in living cells. Digital imagining has become the preferred method for easiness of this method. By using of this technique can be evaluated the volume, area, shape, length and etc of cells. One technique in imaging is changing the gray values to show different images. Illumination intensity is represented in gray values. In gray scale system, low levels of GFP fluorescence can be reached to zero, while high levels of GFP expression can be represented with high gray values closer to 255. The intensity of the gray values will also depend on the imaging hardware sensitivity, exposure times, illumination, spectral properties of the sample, etc. In this method it is important to control the items affecting the detection of Color digital images contain spatial and spectral information about objects.

gene for plant transformation.

C: A reporter gene that overcomes the problems with the CAT and lacZ genes is (uidA) the E. coli β-glucuronidase (GUS) gene. For the low content or lack of endogenous GUS activity in most plants, this gene became one of the most widely used reporters in eukaryotic cells. Another advantage of this reporter gene is easy quantification by using a fluorometric assay, but its detection assay is destructive.

D: A gene which has been very successful as a gene reporter and does not have these problems, is Green Florescent Protein. This gene can be introduced into organisms by injection with a viral vector, breeding, or cell transformation and maintained in their genome. It has been expressed in many bacteria, yeast and other fungi, fish, plant and mammalian cells like human. In cells where the gene is expressed, and the proteins are produced, GFP is produced at the same time. Only those cells in which the target proteins are produced or the tagged gene is expressed will scatter fluoresce when studied under fluorescence microscopy. This reporter can be used to assay for the expression of interest gene that may generate a protein which has immediate effect or little obvious on the cell culture or organism.

GFP expression can be followed over extended periods of time. It is noticeable that GFP expression measured as GFP intensity. It was first studied on several times for each GFP construction. The model of GFP expression in general was similar. Expression has 3 phases. In initial period, there isn't any expression, then this followed by a logarithmic raise in intensity, and finally there is a fixed phase after reaching maximum levels of expression.

By using a spectrophotometer or digital imaging, the GFP expression can be easily distinguished in transgenic cells or in living cells. Digital imagining has become the preferred method for convenience of this method. By using this technique the volume, hyarea, shape, length and other parameters of cells can be evaluated. One technique in imaging is changing the gray values to show different images. Illumination intensity is represented in gray values.

In gray scale system, low levels of GFP fluorescence can be reached to zero, while high levels of GFP expression can be represented with high gray values close to 255. The intensity of the gray values will also depend on the imaging hardware sensitivity, exposure times, illumination, spectral properties of the sample, etc. In this method it is important to control the items affecting the detection of Color digital images contain spatial and spectral information about objects.

Example: GFP insertion in lima beans

Lima bean seeds were incubated overnight and when their coating removed, they were placed in an empty cell culture dish for bombardment. Half–cotyledons were transformed with the pUC-sgfp-TYG-nos plasmid or pUC-mgfp5-ER plasmid by using a particle inflow gun. The 35S promoter controls both GFP genes regulatory. The experimental samples and the control one were imaged for 2 days by gray imaging system.

The sgfp-TYG gene has been expressed earlier and it has a shorter log phase than the mgfp5-ER gene. For this reason sgfp-TYG gene has a higher expression than the mgfp5-ER gene during the log phase. The delay in reaching the maximum expression of the mgfp5-ER gene is a time requirement for maturation of the protein and its labeling to the lumen of the ER. The competition between ER–targeted GFP and other ER–targeted proteins could decrease the value of GFP that can be properly folded and accommodated in the lumen of the ER. The high fluorescence intensity detected from the sgfp-TYG gene might be because of its high solubility.

Cells, with high levels of GFP expression, may be associated with a high number of plasmids that were inserted during the transformation. Low levels of GFP fluorescence may be caused by low number of plasmids or cell damage.

References:

http://en.wikipedia.org/wiki/Green_fluorescent_protein.

Ehrenberg, The Green Fluorescent Protein: Discovery, Expression and. Development, The Royal Swedish Academy of Sciences, 2008.

Buenrostro-Nava, Marco T. Characterization of GFP gene expression an automated image collection system and image analysis, Doctor of Philosophy, Horticulture and Crop Science, 2002, Ohio State University.

Announcement

Announcement

23-25 May 2010

Dubai International Convention and Exhibition Centre, UAE

For more information please refer to: http://www.pabme.com/

News

Anticholera Vaccine

Transgenic plants constitute a novel basis for the production of vaccines against major human diseases such as malaria and cholera., Investigators engineered tobacco and lettuce plants that encoded for the production of the toxin-B subunit (CTB) of *Vibrio cholerae* (cholera antigen), fused to malaria vaccine antigens apical membrane antigen-1 (AMA1) and merozoite surface protein-1 (MSP1) by inserting genes into tobacco and lettuce chloroplasts. Chloroplasts Analysis from the transgenic plants showed, CTB-AMA1 and CTB-MSP1 fusion proteins accumulated up to 7.3% and 6.1% (total soluble protein) in lettuce and up to 13.17% and 10.11% in tobacco.

The researchers revealed that animals receiving the plant antigens by injection lyophilized chloroplast proteins to them produced significant levels of antigen-specific antibody titers. These antibodies completely inhibited proliferation of the malaria parasite and cross-reacted with the native parasite proteins in immunoblots and immunofluorescence studies. Protection against cholera toxin challenge correlated with CTB-specific titers of intestinal or serum IgA and IgG1. The vaccinated animals demonstrated long-term (>300 days or 50% of mouse life span) dual immunity against these two major infectious diseases. Producing vaccines in plants is less expensive than traditional methods because it requires less labor and technology.

Source: biotech daily

www.nature.com

Nature is the world's most highly cited interdisciplinary science website, sponsored by **Macmillan Publishers Company** which was established in 1843. Some features of Nature website are presented below:

- Nature.com has a very strong search engine, which could retrieve data through a wide range of web pages.

- Valid updated articles are the most important feature of the Nature website.

- In the "**Last News"** section you can read the recent scientific news which is gathered from different scientific fields.

- Latest research results are available at the bottom of the homepage.

- In "**Explore Nature"** section, the most visited and popular sections are listed.

- "**Special Feature"** contains interesting information, which is selected by consultants of the site.

- The next section is "**Browse Subjects";** this is divided into five groups:

1- **"Life Sciences",** which is so related with EMGEN network activities including genetics, neuroscience and cancer related issues.

2- "**Physical Sciences"** is about physics and materials topics.

3- "**Chemistry"**, in this section you can also find biotechnological concepts and topics.

4- "**Clinical Practice & Research"** is ranging over a variety of topics from cancer, dentistry to gastroenterology and hematology.

5- "**Earth and Environment"** contains earth science, evolution and ecology issues.

Cover Picture

Title: Neutrophil ingests anthrax bacteria

Description: Neutrophils are the most common type of white blood cell. They are the first immune cells to arrive at a site of infection, through a process called chemotaxis. Neutrophils can produce a net of fibers called a neutrophil extracellular trap (NET) and release it, which can serve to trap and kill infections outside of the cell. Among the ingestion of microbes, Neutrophils release a number of proteins in granules that help kill the microbe. They also produce superoxide that becomes changed into hypochlorous acid, which is theorized to play a part in killing microbes as well. The neutrophil has a lifespan of about 3 days. This picture shows a scanning electron microscope image of a single neutrophil (yellow), engulfing anthrax bacteria (orange).

Source: http://en.wikipedia.org/wiki/Neutrophil_granulocyte

Title: Telomerase

Description: Telomerase is a repeating DNA sequence (TTAGGG) at the 3' of the eukaryotic chromosomes. Telomerase prevents chromosomes from losing base pair sequences at their ends and also stops chromosomes from fusing to each other. This enzyme is a reverse transcriptase that carries its own RNA molecule (as a template when it elongates telomeres). Telomeres are shortened after each replication cycle. When the telomere becomes too short, the chromosome reaches a "critical length" and can no longer replicate. This means that a cell becomes "old" and dies by an apoptosis process. Two mechanisms control telomere activity: addition and erosion. Addition is determined by the activity of telomerase and erosion occurs each time a cell divides.

Source: http://en.wikipedia.org/wiki/Telomerase

Title: *Mycobacterium tuberculosis*

Description: *Mycobacterium tuberculosis* (MTB) is a pathogenic bacteria species in the genus *Mycobacterium* .It has an unusual, waxy coating on the cell surface which makes the cells resistant to Gram staining so acid-fast detection methods are used instead. *M. tuberculosis* has a tough cell wall that stops passageway of nutrients into and send out from the cell, so giving it the characteristic of slow growth rate. It forms a complex with other higher associated bacteria called the *M. tuberculosis* complex that comprises of 6 members: M. tuberculosis and *M. africanmum*, which infect humans; *M. microti*, which infects vole; *M. bovis*, which infects other mammalian species as well as humans; *M. bovis* BCG, a variant of *Mycobacterium bovis* and *Mycobacterium canettii,* a pathogen that infects humans. *M. tuberculosis* infected humans. It has been found in early hominids originating in East-Africa. Therefore, studying the population structure of the species could make available insights about Homo sapiens' migratory and demographic history.

Source: http://en.wikipedia.org/wiki/Mycobacterium_tuberculosis