See inside rodent cell lines retroviruses also monkey bovine etc

CONTINUOUS CELL LINES AS SUBSTRATES FOR BIOLOGICALS

ABSTRACTS

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Material to be published later this year will show that SV_{40} was directly responsible for leukemia in some recipients, as well as brain tumours (and other problems - but these may not be included because they are not yet proven)

#6 A REVIEW OF THE CIRCUMSTANCES AND CONSEQUENCES OF SIMIAN VIRUS 40 (SV40) CONTAMINATION OF HUMAN VACCINES

Keerti V. Shah

Department of Immunology and Infectious Diseases
Johns Hopkins School of Public Health
Baltimore, Maryland

Between 1955 and 1961, about one hundred million people in the USA were immunized with vaccines which were prepared from virus pools made in simian kidney cell cultures and which were at risk of contamination by SV40, a latent renal infection of macaque species. SV40 grows in rhesus cell cultures without overt cytopathic effect and was not recognized until 1960. The practice of holding recently captured juvenile animals in gang cages provided opportunities for transmission of SV40 between cage mates. Pooling of kidneys from different animals for cell cultures further increased the risk of SV40 contamination of the virus pools. The major source of human exposure to SV40 was the inactivated poliovirus vaccine which was administered to 98 million individuals in the USA during the time period when a proportion of the vaccine lots was contaminated with the simian virus. The formalin treatment employed for inactivation of polioviruses was not equally effective for inactivation of SV40. Therefore, some lots contained small amounts of residual live SV40, in addition to inactivated SV40. Approximately 20% of the vaccinees developed neutralizing antibodies to SV40, indicating that they were immunized with contaminated vaccines. The inadvertent exposure of millions of people to SV40 raised public health concern, especially because SV 40 was oncogenic for laboratory rodents. Epidemiologic and clinical studies were conducted to determine if administration of contaminated vaccine was associated with increased cancer mortality in the population; with malignant disease in exposed children; with malignant disease in children born to potentially exposed mothers; and with neurological disease. The studies did not reveal unequivocal evidence of any ill effects attributable to SV40 in the vaccine, but it was also evident that the investigations were not sufficiently extensive or sufficiently detailed to exclude SV40-related complications which may have occurred at a low frequency and after a long incubation period. Experimental lots of live oral poliovirus vaccines, which were administered to several thousand individuals, contained large amounts of SV40 but the oral vaccine commercially licensed in the USA was required to be free of SV40. It was not clear if orally administered SV40 multiplied in the exposed individuals. SV40 was also an unrecognized contaminant of a respiratory synctial virus preparation which was used to study experimental infection in adult human volunteers and of inactivated adenovirus vaccines administered to military recruits. The experience with SV40 contamination of human vaccines highlights the potential hazard associated with the use of primary cell cultures for vaccine production. It also illustrates the difficulties which may be encountered in determining if a vaccine contaminant caused adverse effects in the population at risk.

NB

It is clear. 3 articles show it reproduced was excreted in the environment

During Discussion

French Scientist stood up and said that the French, in the early 1960s, took 1950's polio vaccine from the freezer and retested it and were very surprised to find that many vials had higher concentrations of SV_{40} than poliovirus.

Cell lines were generally believed to be poor candidates for the production of human biologicals because most had properties typical of cancer cells.

In 1962, an NIH committee met to address the issue of cell substrates for the production of human virus vaccines. Unfortunately this was not a public meeting and several conclusions were published for which refutation was subsequently offered in print. For example they continued to embrace the dogma, disproven by Moorhead and me, that " . . . continuously cultured cells eventually develop characteristics suggestive of malignant change." Although this committee was unable to distinguish between cell lines and cell strains they were, in fact, the first group to recommend the use of cell lines for the production of human virus vaccines.

The suggestion that human diploid cells could provide a safe and superior substrate for the production of human virus vaccines was met with considerable resistance for the next decade. The major reason for this resistance was (1) the possibility that they contained an hypothetical human cancer virus or some other latent virus, (2) the possibility that they would spontaneously transform, (3) when compared to commonly used primary monkey kidney cells known to contain many latent viruses (some of which killed several workers) "The devil you know is better than the devil you don't know" and (4) "vaccines are licensed not cell substrates."

In 1963 it became clear to several of the original advocates of the use of HDCS for the production of human biologicals that a program of developmental research and education should be mounted in order to further investigate the safety of the cells and to overcome the resistance to their use. The Cell Culture Committee (CCC) of the International Association of Biological Standardization was thus born. In the next eight years this committee was to play a significant role in the subsequent worldwide licensing of human biologicals produced in WI-38 and similar HDCS.

The concept of the well-characterized and standardized master cell bank, working cell bank, and the first "Minimum Requirements" emerged from a small CCC workshop organized by Drs. Hayflick, Perkins and Ikic in Opatija, Yugoslavia in 1963.

Current interest in using cell lines for the production of human biologicals has resulted in the development of guidelines virtually identical to the procedures originated by the CCC. They have been made a part of all National Control Authority regulations where cell strains or cell lines are sanctioned for use.

The history of the licensing of human vaccines produced in WI-38 had its first tentative beginning in 1965 when the adenovirus type 4 vaccine, produced by Wyeth Laboratories, was used by the US military in recruit training centers where it enjoyed great success for many years. Technically this vaccine was

not licensed by the then Division of Biological Standards (DBS) for civilian use but was used in the military where their approval was not required.

In 1967, through the efforts of Dr. Drago Ikic, the first attenuated poliomyelitis vaccine produced in WI-38 was licensed in Yugoslavia. In 1970 an attenuated poliomyelitis virus vaccine manufactured by Pfizer Laboratories in WI-38 was licensed in the UK. A similar vaccine, produced by Merieux in Lyon, France, was licensed in that country in 1971. Another, produced in the USSR, was also licensed. In 1972 Pfizer Laboratories was licensed to sell their WI-38 produced poliomyelitis vaccine in the USA. Exactly ten years had elapsed from the time that we first reported on the safety and efficacy of a vaccine produced in a HDCS.

Today there are many virus vaccines produced in WI-38 or similar HDCS that are licensed for human use throughout the world. These include poliomyelitis, adenovirus types 4 and 7, rubella, rubeolla and rabies. Tens of millions of people have been inoculated or fed vaccines produced in HDCS. There are no reports of untoward effects in any of the recipients traceable to the cell substrate itself.

Human diploid cell substrate.

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Consideration of the use of cell lines for manufacture of human biologicals has become necessary much sooner than many of us had expected.

W+IY?? If not seriously contaminated AID scaremongering

Now that recombinant DNA and hybridoma technology have revolutionized the way in which human biologicals are produced, a consideration of the cell lines used in these technologies is unavoidable. It is unavoidable because many of the important human biologicals that can be made by recombinant DNA technology can only be produced as biologically active molecules in cell lines. And hybridomas themselves are cell lines.

The chief objection to using cell lines for the manufacture of human biologicals is the same one that was so vigorously held by those who first opposed the use of HDCS. That is, the possibility that they may contain adventitious viruses or a latent cancer virus. Furthermore, since all cell lines by definition are karyologically abnormal, and some produce tumors when inoculated into appropriate laboratory animals, these properties might subject the final product to the risk of contamination with proteins or DNA sequences theoretically capable of producing unwanted effects in recipients.

!!!

Nevertheless, these objections have been largely overcome by (1) utilizing cell lines that do not produce malignancies when tested in animals and (2) validating the manufacturing process such that the likelihood of biologically active DNA sequences or unwanted proteins surviving from the cell substrate is not possible. A good example of how the latter goal has been achieved is the technique employed by Merck, Sharp and Dohme for production of their FDA licensed Hepatitis B vaccine. Pooled human blood is used as the source of the antigen, and was withdrawn as soon as poss!!

Hep B.

Sawtsied named for a medium cultured from aborted fetuses — see M.S.D. insert.

after 64 it was decided that "aborted fetus" was too emotive.

It is unlikely that a substrate with greater actual or theoretical risk could be found for the manufacture of a human biological than is pooled human blood. Yet despite the use of a system in which, perhaps, the ultimate risks are present, the validation of down stream processing is such that the Hepatitis vaccine manufactured from human blood has produced no untoward effects attributable to the substrate used. This is not true!! Alaskan experience shows this to be untrue.

? Hep B

Thus the evolution of modern process technology for human biologicals and the means for validating the absence of unwanted DNA and protein has evolved to the point where any cell substrate can be considered safe.

Although it took ten years of effort for the first biological produced in a human diploid cell strain to be licensed in the USA it is remarkable to observe that only a relatively few months were needed to make the riskier leap to the use of cell lines. In my view this occurred because today scientific attitudes toward change are more receptive and the technical achievements made in down stream processing are such that a human biological can be made safely in virtually any substrate.

#7 DETECTION AND ELIMINATION OF ENDOGENOUS RETROVIRUS AND RETROVIRUS-LIKE PARTICLES IN CONTINUOUS CELL LINES

Masao Ono

School of Medicine, Kitasato University, Kanagawa, Japan

Detection of Retrovirus Genomes

Viruses which replicate their RNA genomes by virus-coded reverse transcriptase are known as retroviruses. This replication is followed by integration of reverse transcribed DNA into host chromosomal DNA to produce a provirus to be used for transcription. Retroviral proviruses transmitted from parents to offspring through germ cells just as in the case of chromosomal DNA are called endogenous retroviruses (ERV's) and all vertebrates are considered to have ERV genomes. In a retrovirus genome, a gene encoding reverse transcriptase (*pol*) is conserved to a greater degree than other genes coding for structural proteins (*gag*), protease (*prt*) and envelope proteins (*env*). By comparison of the deduced amino acid sequences of pol regions, a retrovirus family can be divided into five genetically related groups, one consisting of A-, B--, avian C-, and D-type genomes, one with many mammalian C-type genomes as in the case of the Moloney murine leukemia virus (mo-MuLV), one of several genomes such as the human T-cell leukemia virus type I (HTLV-I), one comprised of a lentivirus subfamily in which the human immunodeficiency virus (HIV) is present and finally one comprised of a relatively uncharacterized spumavirus subfamily. Since one *pol* DNA can be experimentally cross-hybridized with another belonging to the same group, it should certainly be possible to detect an unknown ERV genome and its transcriptional product by a particular *pol* DNA unless the *pol* region is lost in that genome. ERV genomes homologous to the 1st and 2nd group retroviruses specified above are abundantly present in the mammalian genome, whereas no ERV homologous to HTLV-I, lentivirus, and spumavirus group has yet been detected.

Human Endogenous Retrovirus

ERV genomes present in mammals such as humans and mice have been well characterized. Humans possess many ERV genomes related to the mammalian C-type, A-B-D-type and unidentified-type due to insufficient structural clarification. The number of genomes in each ERV varies from one to a thousand per haploid human genome. Some of the genomes are transcribed in particular cells and / or under special conditions, but no translation product of a human ERV genome has been identified and no retrovirus-like particle infrequently found in a human cell would be related to a specific ERV genome. At the present, there is no concrete evidence for an etiological relationship between a human ERV genome and a disease such as cancer or autoimmune dysfunction.

Why ? vaccine ?

N.B.

Mouse Endogenous Retrovirus

Laboratory mice have several groups of ERV and retrovirus-like particle genomes. Mammalian C type ERV genomes in mice may be divided into either a class of ecotropic ERV genomes present in a few copies and have the appearance of being recently integrated, or a class of non-ecotropic ERV genomes found up to a hundred copies and considered to possibly be integrated prior to establishment of inbred mouse strains. Some of these ERV genomes possess genetic information by which replication competent retroviruses can be formed and a leukemia-inducing virus is produced from recombinants between endogenous ecotropic and non-ecotropic retroviruses. Endogenous mouse mammary tumor virus (MMTV, B-type) genomes are strain specifically found up to several copies per haploid mouse genome. Intracisternal A particles (IAPs, A-type) are retrovirus-like structures consistently observable in a variety of tumor cells such as myeloma and hybridoma but nothing is known of their infectivity. IAP genomes are interspersedly present in rodents such as mice, rats, Syrian hamsters and Chinese hamsters in several hundred to a thousand copies per haploid genome. Haploid mice and rats each have about a hundred copies of the VL30 genome (a type unknown but possibly belonging to the mammalian C-type) which is transcribed into 30S RNA (virus-like 30S RNA). 30S RNA's are then packaged in exogenous infectious particles by a helper endogenous mammalian C-type retrovirus.

Expression and Elimination of Endogenous Retrovirus and Retrovirus-like Particles

Many continuous cell lines used to produce biological products are derived from mammalian cells in which a wide variety of ERV genomes is quite possibly present. Although their expression largely depends on their particular type, what cells they are present in and surrounding conditions, transformed cells generally show greater degree of expression than normal cells. The expression of some ERV genomes can be induced either by a halogenated pyrimidine, the DNA methylation inhibitor, a tumor promoter or a growth factor.

Although it is quite unlikely that endogenous retrovirus and retrovirus-like particles are a biohazard to human beings, there is nevertheless the possibility and thus, they should be eliminated from biological products for human use. Conventional safety tests are sufficient for inspecting biological products provided no retrovirus-like particle is present in substrate cells. But if so, changing the cell line to another would be the best course of action but of course unwise for cell lines derived from rodents due to the difficulty in obtaining a particle-free cell line. Otherwise, biological and/or physical means for eliminating such particles would be best. A general procedure should be available for inactivating retroviruses in vaccines. A standard method for selectively inactivating the biological activity of nucleic acid but not protein by physical (e.g., UV or radiation), chemical, or biological (e.g., nuclease treatment

NB

Remember that "conventional" testing procedures did not pick up SV_{40} for 6 years, and and the 140+ viruses which were detected as time passed.

following particle disruption) treatment should be developed since it is known that the essential biological activity of the particles is on the RNA genome. Should a product be too unstable to permit the application of these methods, elimination of the particles can be carried by physical means since they are sufficiently large.

#15 POLYOMA-TYPE VIRUS CONTAMINATION OF PRIMATE CELLS

G. van Steenis, C. J. A. Sol, J. van der Noordaa
A. D. M. E. Osterhaus

Rijksinstituut voor Volksgezondheid en Milieuhygiëne, Bilthoven,
Laboratorium voor de Gezondheidsleer,
Universiteit Amsterdam,
Amsterdam, The Netherlands

A polyoma-type virus was isolated from cynomolgus monkey (*Macaca fascicularis*) kidney (CK) cells. The virus proved not closely related to SV40 virus but resembled both biochemically and immunologically, similar virus isolates from stump-tailed macaque cells, Vero cells and fetal rhesus monkey kidney cells (1, 2).

No antibody was found in the serum of the cynomolgus monkey from which the kidney cell culture originated or other animals from the same colony. However, antibody was found to occur wide-spread in bovine serum, including fetal calf serum from geographically different areas, indicating bovine serum as the probable source for contamination. Antibody to the virus was not observed in porcine sera, making trypsin of porcine origin a less likely source.

NB.

Recently two more isolations were made on separate occasions from different batches of subcultured CK cells, which had in common that they had been maintained in serum free medium, after initial cultivation in different batches of bovine serum. The serum batches concerned had been examined for viral contamination using the methods indicated by WHO (3), with negative results. This finding suggests a need for changes in screening methods of bovine serum for virus contamination.

NB.

Cells permissive for replication of CK isolates included calf kidney cells, Vero cells, dog kidney cells and probably also human diploid cells, as indicated by the appearance of specific fluorescence in inoculated cells.

A recently reported observation on the occurrence of antibodies to these papova viruses in people with frequent contacts with cattle such as farmers and veterinarians (4) suggests man as a natural host. The widespread occurrence of this type of agent in cattle is also testified by the high frequency of its isolation from kidney cell cultures of young calves (5).

The wide-spread occurrence, extensive range of susceptible cells, and apparent infectivity for man make this virus a potential threat to the use of biologicals which depend for their production on the use of bovine serum. These papova viruses therefore merit more attention than they have been given so far in the control of biological products.

NOTE – Bovine serum CLEARED as squeaky clean was NOT

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#17 POSSIBLE SIGNIFICANCE OF RODENT VIRUS CONTAMINATION OF BIOLOGICAL PRODUCTS FOR USE IN HUMANS

P. Carthew

Medical Research Council Toxicology Unit, Carshalton, Surrey, England

Since Kohler and Milstein's publication on the preparation of monoclonal antibodies in 1975 much work has been devoted to the exploitation of this technique for diagnostic and therapeutic uses in humans. Monoclonal antibody preparations produced in both rats and mice are routinely used in vivo for techniques as diverse as tumor diagnosis and localization, and the treatment in vitro of lymphocyte preparations of patients, to ameliorate graft versus host disease after bone marrow transplants. Used in immunosuppressed patien

The possibility thus exists that rodent viruses may contaminate such products, due to the original source of cells involved in their production, and also because the most efficient method of bulk producing monoclonal antibodies is as ascites fluid induced in the peritoneal cavity in rodents.

We must therefore decide what materials we need to test to ensure the safety of patients who will receive these products, and also what we need to test these materials for exactly. Do we need to test the hybrid cell lines that are prepared, prior to their use in ascites fluid production, so as to guarantee the virological integrity of the animals they are subsequently injected into? We obviously need to exercise stringent controls over the virological status of the animals in which such cell lines will be passaged. However, will this be possible in practice? Most rodent colonies throughout the world are infected with a wide range of inapparent viral pathogens, except of course germ free rodents. Perhaps before we consider the expensive and restricting principle of using germ free animals for ascites fluid production, we should consider what possible risk there is to humans from rodent viruses.

One way of considering the possible pathogenicity of rodent viruses for humans, is to examine which of these viruses are recorded zoonoses. Certainly, Hantaan virus, lymphocytic choriomeningitis virus and the rat rotavirus, were well documented zoonotic viruses, and must certainly be excluded from antibody preparations, in the same way that they are currently excluded from animal facilities, by serological monitoring. Unfortunately we cannot rely on such a simple principle in considering the other rodent viruses. Just because some rodent viruses have not been recorded as being transmissible to humans under the normal conditions of contact between humans and rodents, does not mean that they may not be pathogenic when inadvertently administered intravenously, or attached to lymphocytes reintroduced into patients, after an in vitro procedure. This could give rodent viruses access to susceptible cell types, which they would not normally

encounter at mucosal surfaces, (the normal route of transmission in zoonoses) and thus allow their replication. It would seem reasonable to say that any rodent virus which is recorded in the literature as having been found to replicate in human or monkey cell cultures would also be considered as undesirable, and should be excluded from biological products for use in humans. This could be especially important where patients are immunosuppressed and the virus might have a much enhanced pathogenicity, and will also increase our list of undesirable pathogens by at least another ten viruses. Does this mean that the now small number of remaining viruses pose no threat to humans? Unfortunately we cannot even be sure of that, because most of these have not been tested extensively for replication in human cells. Prior to the advent of rodent monoclonals there was no need to know this type of information. We could test these viruses in a variety of human cell cultures, using sensitive techniques such as immunocytochemistry, or even radioimmunoassay to detect viral replication; but could this be considered to be a stringent enough safeguard for their lack of pathogenicity for humans? It would certainly be very laborious and at best could be considered equivocal.

Perhaps the only safe approach to the problem is to remove all of the viruses from biological products, because of the possibility that there are some rodent viruses which have not as yet been identified, because of their low pathogenicity for rodents; but which cannot be assumed to be so innocuous for humans (Hantaan virus is a good example of this). Of the many approaches to purification of antibodies from ascites fluids, perhaps the most attractive is a method using affinity chromatography coupled with a "spiking" quality control assurance. The "spiking" technique would use a range of viruses, and perhaps other microbial organisms, their toxic products and even allergens, which are deliberately introduced into batches of the product to be purified so as to validate the efficiency of the purification technique. This was used to good effect in evaluating the purification of human interferon some years ago, and could be an important concept in monoclonal antibody preparation, and quality assurance prior to use in human therapies.

#18 PREVENTION OF VIRAL CONTAMINATION IN PRODUCTS DERIVED FROM MAMMALIAN CELL CULTURE

Alfred M. Prince and Bernard Horowitz

The Lindsley F. Kimball Research Institute and Blood Derivatives Program of the New York Blood Center
New York, New York.

Increasingly injectable products for human use will be manufactured in mammalian cell cultures. Foremost among these will be products derived from recombinant DNA technology, lymphokines, and monoclonal antibodies. It is important that such products should be free of contaminating viruses.

Two sources of potential virus contamination exist: cells and culture medium.

Control of contamination introduced from cells has traditionally employed the use of standard virus isolation procedures using cell cultures and experimental animals, as well as the use of electron microscopy. More recently sensitive molecular hybridization methods have been introduced which make it possible to rule out the presence of less than one genome of known viruses per cell if appropriate probes are available. The sensitivity of these procedures can be further enhanced by up to a million fold by the use of the Polymerase Chain Reaction (PCR).

In the case of murine hybridoma derived products absence of viral contamination is further documented by mouse antibody production tests (MAP) capable of detecting Reovirus type 3, Hantaan virus, Polyoma, Pneumonia virus of mice, mouse adenovirus, Minute virus of mice, Mouse hepatitis, Ectromelia virus, Sendai, and GD VII. Additional tests must be done to detect Lymphocytic Choriomeningitis virus, Mouse salivary gland virus, Epidemic diarrhea of infant mice virus, Lactic Dehydrogenase virus and murine leukemia viruses.

In the case of human hybridoma derived products it is necessary to assure the absence of Epstein Barr virus DNA by tests for viral antigens on the cell line, and by DNA hybridization if these antigens are not detectable. In addition, attempts to culture CDV on cord blood lymphocytes should be carried out. Cytomegalovirus should be sought for in diploid fibroblast cell culture. Human retroviruses should be excluded by reverse transcriptase assay of culture supernatants, by electron microscopy, cocultivation, and by DNA hybridization. In addition, cell donors should be tested and found free of infection with known human retroviruses. Hepatitis B virus is best ruled out by DNA hybridization as well as by assays with third generation tests for HBsAg.

Viruses can also be introduced with the serum used for cell cultivation. In the future it is likely that these will be sterilized by one or more of the procedures which have been introduced for sterilization of human blood derivatives primarily to render these safe with respect to transmission of Hepatitis B and

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non-A, non-B viruses and HIV. These procedures include the use of solvents and detergents in combination, the combined use of ß-propiolactone and ultraviolet light, Pasteurization in the liquid state and heating in the lyophilised state.

In addition to providing protection against introduction of viruses by serum used in culture medium, the sterilization procedures have obvious application to sterilization of final products derived from cell culture and should be used for this purpose if maximal safety is to be achieved.

#34 ALTERATIONS IN THE INTRACELLULAR DISTRIBUTION OF CHO CELL INTRACISTERNAL A PARTICLES DETECTED BY ELECTRON MICROSCOPY AFTER TREATMENT WITH TUMOR PROMOTER (TPA) AND GM-CSF

Carol J. Marcus-Sekura, Michael Klutch, Marilyn Lundquist, and Ruth C. Dunlap

Division of Virology, Center for Biologics Evaluation and Research, Food and Drug Administration Bethesda, MD

what recombinant biologics.

Chinese Hamster Ovary (CHO) cells, which are being used as a cell substrate for the production of recombinant biologics, have been shown by electron microscopy to contain viral-like particles, sometimes called intracisternal A particles (IAP) or retroviral type-A particles. Repeated attempts in many laboratories to detect infectious virus or reverse transcriptase activity in these cells have been negative. Sequence data on cloned IAP particle genomes from cell lines including Syrian Hamster and human, suggest that these IAP particles have defective genomes.* Since viral-like particles do form in CHO cells, at least some of the viral genes must remain functional, although viral maturation or infectivity is impaired. This block in maturation permits investigation of intermediate stages of viral development. We have, therefore, examined the effects of the viral inducing phorbol ester 12-0-tetradecanoylphorbol 13-acetate (TPA) and the cellular growth factor GM-CSF on the nature, number and intracellular location of these particles.

CHO cells (ATCC #CCL CHO-K1) were grown in Ham's F12 medium supplemented with 10% fetal bovine serum and treated with either TPA (20ug/ml) or GM-CSF (recombinant from Genzyme, 5000u/ml) alone or in combination. Four days later, treated cells as well as untreated controls were harvested and prepared for microscopy. Cells were fixed, embedded in Epox resin and the blocks were then sectioned and examined in a Zeiss 10A electron microscope. A quantitative assessment of particles was made by counting the particles visible in one thin section through 200 different cells for each sample. Scoring included categorizing as to number of particles per cell, type of particle, and location of particle.

Control and treated cells contained predominantly type A particles. There was no significant increase in the number of extracellular particles observed in the samples treated with TPA or TPA + GM-CSF(10-11 particles) compared to untreated cells or cells treated with GM-CSF alone (3 particles). No differences in the number of budding or intravesicular particles were observed. In the TPA treated samples, there was an increase in the total number of cells scored as containing one or more particles (79 of 200 cells for TPA alone, 59 of 200 for TPA + GM-CSF) compared to untreated CHO cells (48 of 200) or treatment with GM-CSF (47 of 200), suggesting that TPA might trigger

*Which means that standardized testing cannot pick them up since it depends on non-defective genomes.

de novo synthesis or assembly of particles. In addition, a change in the distribution of the particles within the cell was observed: in the presence of TPA alone there was an increase in the total number of cytoplasmic particles scored as well as centriole-associated particles. In the presence of GM-CSF the number of cytoplasmic particles remained the same as untreated controls, however, there was an increase in centriole-associated particles comparable to that observed with TPA. If one assumes that particle maturation involved movement from the centriole to the cytoplasm to the cell membrane, it is tempting to speculate that we are observing alterations in the particle maturation induced by these growth factors; i.e., TPA might be more effective than GM-CSF and induce increases in particles in both the centriolar-region and progression to cytoplasm, where as GM-CSF might act at earlier stages and stimulate increases in centriole-associated particles. Perhaps the signals for this type of maturation remain intact in the CHO cell IAP genome. Studies are in progress to examine the effects of additional exogenous cellular growth factors on IAP particles, and to use the related Syrian Hamster IAP clones to screen for the CHO cell IAP genome, both in treated and untreated cells to assess whether treatment with growth factors affects levels of IAP RNA.

#40 CLINICAL USE OF BIOLOGICALS PRODUCED IN CONTINUOUS CELL LINES

Thomas Hoffman

Center for Biologics Evaluation and Research
8800 Rockville Pike
Bethesda, MD

Recent advances in biotechnology, namely DNA cloning and hybridoma formation, have greatly increased the availability of biologicals for clinical use. cDNA directed proteins and monoclonal antibodies may now be produced in sufficient quantities for wide-ranging experimental uses and, in some cases, licensed applications. Furthermore, novel uses of biological substances produced in minute amounts for normal homeostasis may now be broached using "pharmacological" doses.

Biologicals produced in continuous cell lines have found application in wide-ranging fields. These include: hormone replacement, vaccines, immunosuppression and immunotherapy, and control of homeostasis. Initial efforts have been focused in oncology and infectious disease, including HIV-infection. Currently, therapies for a wide variety of conditions including thrombo-hemorrhagic syndromes, inborn errors of metabolism, cataracts, and arthritis are being developed. Conceivably, virtually every aspect of medicine, including diagnosis and surgery, will be affected by this revolutionary approach.