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Research Article

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Efficacy of Car T-Cell Therapy in Head and Neck Cancers- A Meta-Analysis

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Abstract

Background: *Cancer, defined by the World Health Organization (WHO) is "a large group of diseases characterized by the growth of abnormal cells beyond their usual boundaries that can then invade adjoining parts of the body and/or spread to other organs". From extensive surgical excisions, radiotherapy, laser therapy to immunotherapies, various treatment strategies have been proposed and implemented so far but unfortunately none could improve the five year survival rate of the patients globally. Immunotherapy, being one amongst them, is a type of cancer treatment that boosts the body's natural defenses to fight against cancer. The current concept of immunotherapy involves Chimeric antigen receptor or the CAR T-Cell therapy which involves alterations and modifications of T cells to fight cancer cells better. Until recently, the use of CAR T-cell therapy has been restricted to small clinical trials, largely in patients with advanced blood cancers and has also shown a promising window of hope in head and neck (especially oral) cancers as well. But these treatments have nevertheless captured the attention of the people because of the remarkable responses they have produced in some patients for whom all other treatments had stopped working. The current concept of immunotherapy involves the cancer vaccines making use of CAR T-cells which are the most powerful antigen presenting cells for the induction of antigen specific T cell response. This evidence based study therefore aims to highlight the clinical perspective of CAR T-Cell based immunotherapy in oral and other head and neck cancers.*

Aim: *Assessment of efficacy of Car T-Cell Therapy in head and neck Malignancies.*

Research Question: *Is Car T-Cell Therapy actually effective in treating head and neck cancers?*

Materials and Methods: *Study sample included review of 70 research articles, based on scientific data bases from the English literature based COCHRANE collaboration having a definite RCT (Randomized Control Trial). The literature was studied, analyzed and assessed; comparison was made on their p (probability) values between various techniques in terms of their sensitivity and specificity. The articles were scrutinised based on the criterion for meta-analysis and finally 11 study articles were chosen for the study.*

Result and Conclusion: *Due to its unique individual characteristics, it helps combat against the cancer cells at its very inception. Promises a complete and permanent cure for malignancies at the grass root level. It has a sensitivity and specificity of greater than 80-90% and enhances recovery rate from 40-50% to more than 90%. So on a bulls eye view, we can say that as the already existing techniques of oral cancer treatment are very superficial and not very significant, Car T-Cell therapy tends to bring about a paradigm shift in oral cancer treatment thereby giving a new ray of hope to cancer ailing patients.*

Keywords: Car T-Cell Therapy, Antigen presenting cells, Cancer, Immunotherapy

Introduction

Cancer, the six letter word agonizing and annihilating, obliterating and destroying innocent lives since the beginning of time is a silent killer within people. Cancer, according to the World Health Organization (WHO) is defined as 'A large group of diseases characterized by the growth of abnormal cells beyond their usual boundaries that can then invade adjoining parts of the body and/or spread to other organs'. Various conventional treatment methods have been tried so far and have had its failures and successes like

surgical excision, radiotherapy and immunotherapy. However, these conventional techniques have started to fade giving rise to the current concept of "Immunotherapy". Immunotherapy is a type of cancer treatment that boosts the body's natural defenses to fight against cancer. The current concept of immunotherapy involves Chimeric antigen receptor or the CAR T-Cell therapy which involves alterations and modifications of T cells to fight cancer cells better. Until recently, the use of CAR T-cell therapy has been restricted to small clinical trials, largely in patients with advanced blood cancers and has also shown a promising window of hope in head and neck (especially oral) cancers as well. But these treatments have nevertheless captured the attention of the people because of the remarkable responses they have produced in some patients for whom all other treatments had stopped working. The current concept of immunotherapy involves the cancer vaccines making use of CAR T-cells which are the most powerful antigen presenting cells for the induction of antigen specific T cell response. This evidence based study therefore aims to highlight the clinical perspective of CAR T-Cell based immunotherapy in oral and other head and neck cancers.

Materials and Methods

Study sample included review of 70 research articles, based on scientific data bases from the English literature based COCHRANE collaboration having a definite RCT (Randomized Control Trial). The literature was studied, analyzed and assessed; comparison was made on their p (probability) values between various techniques in terms of their sensitivity and specificity. The articles were scrutinised based on the criterion for meta-analysis and finally 11 study articles were chosen for the study.

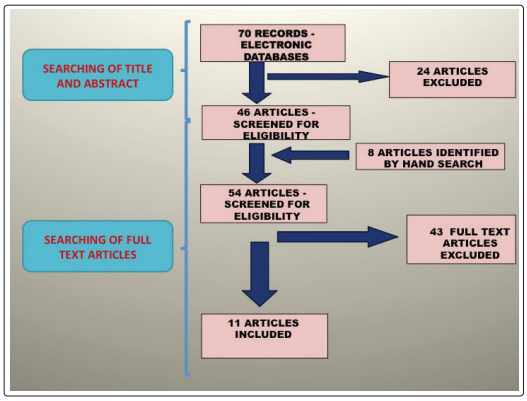

Figure 1: Schematic representation of the selection criterion for the studies in this meta-analysis

Result and Conclusion

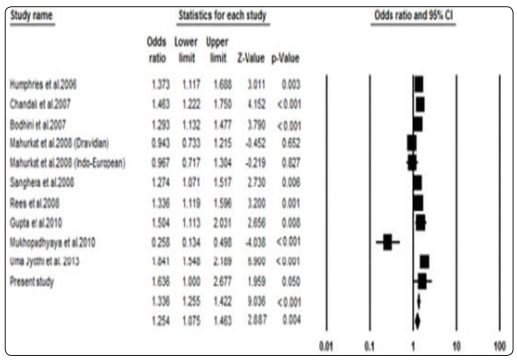

Figure 2: Forest plot or meta-analysis graph of our study

- So when the P values or probability values of all the 11 articles were subjected to meta-analysis, we obtained a forest plot as seen in Figure 2.
- To interpret this forest plot, to the extreme left of the graph are the 11 articles chosen for meta-analysis and the middle of the graph represents the odds ratio and standard deviation values along with the p values of all the 11 articles chosen.
- Towards the extreme right, moving onto the graph proper there is a central line called the median line or the null hypothesis line. The median line has boxes on the right and left of the line signifying the sample sizes of each study that was chosen.
- The boxes on the left of the graph proper and the ones touching the median line states that those particular studies are not significant but the ones on the right side of the median line and not touching the line are considered to be significant.
- Finally, the diamond shaped box on the bottom of the graph (which is a summation of all the probability values) is neither touching the median line and is on the right side of the line proving that our study is significant.

This meta-analysis conducted in order to document the significance of the Car T-Cell therapy although in the early era of acceptance has been found to be highly significant and a potent future weapon of fight against oral cancers with high sensitivity and specificity.

Discussion

Oral cancer is one of the 10 most common cancers in the world, with a delayed clinical detection, poor prognosis, without specific biomarkers for the disease and expensive therapeutic alternatives.

The genetic engineering of T cells through the introduction of a chimeric antigen receptor (CAR) allows for generation of tumor-targeted T cells. Once expressed by T cells, CARs combine antigen-specificity with T cell activation in a single fusion molecule. Most CARs are comprised of an antigen-binding domain, an extracellular spacer/hinge region, a trans-membrane domain and an intracellular signaling domain resulting in T cell activation after antigen binding.

Tumor associated antigens can be divided into several groups including antigens that

1. Contain novel peptide sequences due to gene mutation.
2. Are expressed in a tissue/lineage specific fashion.
3. Are normally expressed during fetal development or at immunoprivileged sites.
4. Are expressed at higher than normal levels on tumor cells compared to non-malignant host cells.

Mutated Antigens

Ideally, the targeted antigen should contain a novel peptide sequence, limiting its expression to tumor cells. One example is a splice variant of the epidermal growth factor receptor (EGFRvIII). Other antigens that may provide some target exclusivity include those with altered post-translational modifications or those that present conformational epitopes unique to the tumor microenvironment.

Tissue/Lineage Antigens

Tissue/Lineage restricted antigens by definition show restrictive expression patterns and may seem particularly attractive. While tissue expendability is acceptable or correctable in certain conditions like B-cell aplasia with CD19 CART cells, the function of most solid organs cannot be readily replaced.

Developmental Antigens

Antigens expressed during fetal development or at immunoprivileged sites such MAGE family members or NY-ESO-1 is actively being targeted with αβ TCRs. However most of these antigens are in cytoplasm, making them inaccessible to scFv-based CARs that recognize antigens expressed on the cell surface.

Overexpressed Antigens

The majority of targeted antigens are only overexpressed in comparison to normal tissues, raising concerns about 'on target/ off tumor' side effects, which in most cases cannot be adequately assessed in murine models.

Manufacturing and Preparation of Car T-Cells

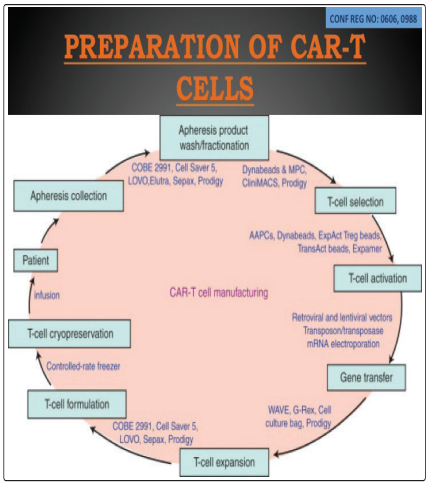

Figure 3: Schematic representation of manufacturing of Car T-Cells

Steps Include

1. Patient blood collection at the hospitals and clinics are carried out and sent in vials of preserving mediums like Leukopacks that improve the viability of the cells.
2. The CAR-T cell-manufacturing processing starts from the collection of peripheral blood mononuclear cell from the patient, commonly achieved by a leukapheresis process. Consenting physicians choose the appropriate window for collection based on treatment regimens to ensure the presence of sufficient numbers of T lymphocytes.
3. Collected apheresis products can be processed in various ways depending on the downstream procedures. Devices such as Haemonetics Cell Saver 5+, COBE2991, and Fresenius Kabi LOVO have the ability to remove gross red blood cells and platelet contaminants.
4. Terumo Elutra and Biosafe Sepax systems provide size-based cell fractionation for the depletion of monocytesand the isolation of lymphocytes.
5. Instruments such as CliniMACS Plus and Prodigy systems allow the enrichment of specific subsets of T cells, such as CD4+, CD8+, CD25+, or CD62L+ T cells using Miltenyi beads post-cell washing as shown in Figure 2.
6. The processed T-cell source material can either be used directly for downstream procedure or cryopreserved for future use. There are pros and cons for either practice. Nevertheless, cryopreserving the processed T cells allows time for product release testing and more flexibility for downstream process planning.
7. T-cell activation: The ex vivo expansion of T cells requires sustained and adequate activation. T-cell activation needs a primary specific signal via the T-cell receptor (Signal 1) and costimulatory signals such as CD28, 4-1BB, or OX40 (Signal 2). T-cell activation is also required for the transduction of the CAR cDNA via retroviral vectors. T-Cell activation can be done via :

- **Cell-Based T-Cell Activation**
  Antigen-presenting cells, such as dendritic cells (DCs), are the endogenous activators of T-cell responses. While therapeutic applications of DCs continue to be investigated, DC potency varies from patient to patient. Such limitation hampers the usage of DCs as a reliable source for T-cell activation.

- **Beads-Based T-Cell Activation**
  Several biotech companies have generated off-the-shelf clinical-grade T-cell activation reagents including the Invitrogen CTS Dynabeads CD3/28, the Miltenyi MACS GMP ExpAct Treg beads, Miltenyi MACS GMP TransAct CD3/28 beads, and the Juno Stage Expamer technology.

- **Antibody-Coated Magnetic Beads**
  Dynabeads CD3/28 is uniform super-paramagnetic beads covalently coupled to CD3 and CD28 antibodies. The added value of this reagent is that it enables the selection and activation of T cells in a single step when used in conjunction with the Dynal ClinExVivo MPC magnet.

- **Antibody-Coated Nano Beads**
  Miltenyi MACS GMP TransAct CD3/28 beads are polymeric nanomatrix conjugated to CD3 or to CD28 monoclonal antibodies. The advantage of the TransAct CD3/CD28 beads is that they are biodegradable, and therefore do not require removal prior to formulation, although upstream T-cell purification is needed prior to activation.

- **Expamer Technology**
  The most recent development in T-cell activation reagent is the Expamer from Juno Therapeutics. Its unique core Streptamer technology has been used to isolate viral-specific lymphocytes. It has been reported recently that as a soluble and dissociable T-cell stimulation reagent, Expamer efficiently induces T cell receptor (TCR) signaling and efficiently activates T cells to support retroviral transduction and expansion.

8. Genetic modification of T cells: Current CAR-T cell therapies largely rely on stable CAR expression upon delivery by viral and nonviral gene transfer systems. There are three major types of stable gene expression vectors used for clinical applications: σ-retroviral vectors, lentiviral vectors, and the transposon/ transposase system. Messenger RNA transfer-mediated gene expression is another method to introduce CARs into cells while avoiding long-term expression.

Both retroviral and lentiviral vectors are complicated complex biological reagents that require intensive and expensive biosafety testing. A relatively new plasmid based expression system, the transposon/transposase system, has been used to introduce anti-CD19 CAR into T cells by electroporation. The advantages of this system are its simple manufacturing procedure, relatively low cost, and straightforward release testing. Integration is random, posing a potential oncogenic risk secondary to mutagenesis.

9. Expansion of CAR-T cells: Depending on the CAR-T cell modification strategy, there are several expansion platforms readily available to generate therapeutic doses of CAR-T cells.

- Expansion of CAR-T cells using GE bioreactors
- Expansion of CAR-T cells using G-Rex bioreactors
- Expansion of CAR-T cells using Prodigy
- Expansion of CAR-T cells through recursive AAPC stimulation

10. Finally, it is formulated and cryopreserved and sent to the clinic where it is infused back into the patient intravenously.

Mechanism of Action in the Human Body

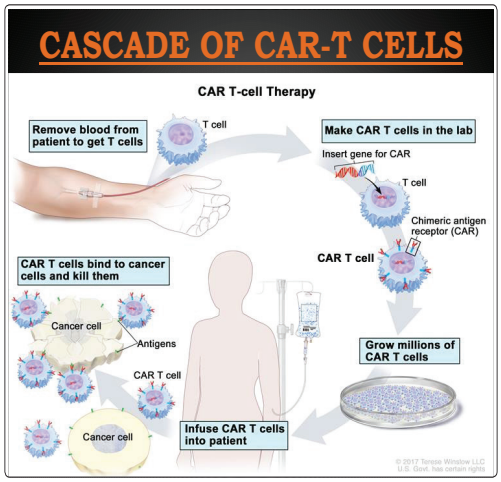

Figure 4: Schematic representation of how Car T-cells act on the tumour cells in the body

Mechanism of Action of Car T-Cells in the Human Body

- First, the tumour cells are identified.
- Then the patient's blood is collected and as given in FIG-2 CAR T cells are produced.
- Then sent to the clinics where it is intravenously infused into the patient.
- Where these CAR T cells travel through the blood circulation and activate to attack the tumour cells thereby reducing its size.
- It is usually done in two to three sessions in the hospital (Figure3).

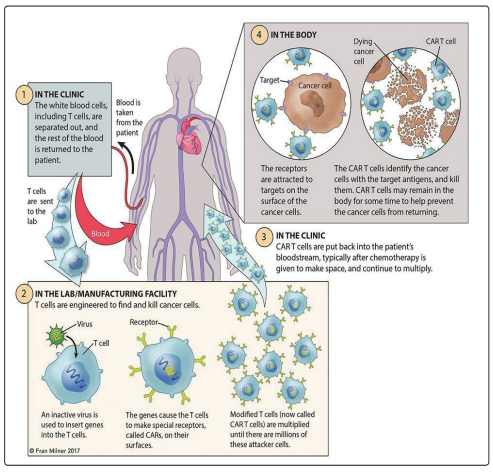

Figure 5: Summarising the production and action of Car T-Cells

Some Toxic Effects Car T-Cell Therapy Includes Cytokine Release Syndrome (CRS)

This potentially serious side effect is frequently associated with CAR T-cell therapy. Cytokines (chemical messengers that help the T cells carry out their functions) are produced when the CAR T cells multiple in the body and kill the cancer cells. CRS symptoms can range from mild flulike symptoms that include nausea, fatigue, headache, chills and fever to more serious symptoms, such as a low blood pressure, tachycardia (abnormally rapid heart rate), capillary leakage (fluid and proteins leak out of tiny blood vessels and flow into surrounding tissues, resulting in dangerously low blood pressure), cardiac arrest, cardiac arrhythmias, cardiac failure.

B-Cell Aplasia

CAR T-cell therapy targeting antigens found on the surface of B cells not only destroys cancerous B cells but also normal B cells. Therefore, B cell aplasia (low numbers of B cells or absent B cells) is an expected result of successful CD19-specific CAR T-cell treatment and has served as a useful indicator of ongoing CAR T-cell activity. This effect results in less ability to make the antibodies that protect against infection. Intravenous or subcutaneous immunoglobulin replacement therapy may be given with the aim of preventing infection.

Tumor Lysis Syndrome (TLS)

Another known side effect of CAR T-cell therapy is tumor lysis syndrome (TLS), a group of metabolic complications that can occur due to the breakdown of dying cells-usually at the onset of toxic cancer treatments. However, TLS can be delayed and may occur one month or more after CAR T-cell therapy. TLS can cause organ damage and can be a life-threatening complication of any treatment that causes breakdown of cancer cells, including CAR T cells. The complication has been managed by standard supportive therapy.

Conclusion

The actual need for this meta-analysis was laid on the foundation of following conclusions which were drawn from the final 11 studies that were finally selected after having undergone the inclusion and exclusion criterion, via;

- Due to its unique individual characteristics, it helps combat against the cancer cells at its very inception.
- Promises a complete and permanent cure for malignancies at the grass root level.
- It has a sensitivity and specificity of greater than 80-90% and enhances recovery rate from 40-50% to more than 90%.

So on a bulls eye view, we can say that as the already existing techniques of oral cancer treatment are very superficial and not very significant, Car T-Cell therapy tends to bring about a paradigm shift in oral cancer treatment thereby giving a new ray of hope to cancer ailing patients.

References

1. Curran KJ, Pegram HJ, Brentjens RJ (2012) Chimeric antigen receptors for T cell immunotherapy: current understanding and future directions. J Gene Med 14: 405-415.
2. Hartmann J, Schüßler-Lenz M, Bondanza A, Buchholz CJ (2017) Clinical development of CAR T cells-challenges and opportunities in translating innovative treatment concepts. EMBO Mol Med 9: 1183-1197.
3. Xiuyan Wang, Isabelle Rivière (2016) Clinical manufacturing

of CAR T cells: Foundation of a promising therapy. Mol Ther Oncolytics 3: 16015.
4. Shivani Srivastava, Stanley R Ridell (2015) Trends in Immunology.
5. Kakarla S, Gottschalk S (2014) CAR T cells for solid tumors: armed and ready to go? Cancer J 20: 151-155.
6. Kochenderfer JN, Rosenberg SA (2013) Treating B-cell cancer with T cells expressing anti-CD19 chimeric antigen receptors. Nat Rev Clin Oncol 10: 267-276.
7. Almåsbak H, Aarvak T, Vemuri MC (2016) CAR T Cell Therapy: A Game Changer in Cancer Treatment. J Immunol Res 2016: 5474602.
8. Bruce L Levine, James Miskin, Keith Wonnacott, Christopher Keir (2017) Global Manufacturing of CAR T Cell Therapy 4: 92-101.
9. Maude SL, Teachey DT, Porter DL, Grupp SA (2015) CD19-targeted chimeric antigen receptor T-cell therapy for acute lymphoblastic leukemia. Blood 125: 4017-4023.
10. Maude SL, Barrett D, Teachey DT, Grupp SA (2014) Managing cytokine release syndrome associated with novel T cell-engaging therapies. Cancer J 20: 119-122.