Repurposing Insulin in Oncology: A Literature Review

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Introduction

The committed efforts of people in healthcare and science have led to laudably decreased mortality rates in the last 26 years for the four most common types of cancer, namely lung, colon, breast, and prostate. By some estimates, approximately 2.9 million cancer-related deaths have been averted between 1991 and 2017 (Siegel, 2020). Primary prevention, early detection, and key developments in therapeutic advances are to be applauded. Yet with reductions in death slowing for breast and colon cancers, and at a standstill for prostate between 2008 and 2017 (Siegel, 2020), there is still a great need to nurture hope for therapeutic improvements for the 608,570 individuals who are projected to die of the disease in 2021 (American Cancer Society, 2021).

An American Cancer Society analysis of treatment patterns revealed a rather large unmet need for acceptable management options in people diagnosed with stage IV disease. 26% of breast cancer, 20% of colon cancer, 17% of rectal cancer, 14% of diffuse large B cell, 32% of stage IV lung cancer and 19% of stage III, 13% bladder cancer, and 13% of uterine cancer patients living with stage IV disease opted not to pursue any conventional forms of management, specifically radiotherapy, treatment-directed surgery, and/or chemotherapy (American Cancer Society, 2019). These figures have likely changed to a certain extent with the introduction of checkpoint inhibitors, however, these medications have not been shown to be ubiquitously beneficial across all tumor types. Aside from palliative care and hospice, what can the oncologic and healthcare community at large offer these patients to nurture hope? Hope of relief from pain

and discomfort? Hope for another birthday, holiday, graduation, or grandchild? Hope for some form of control over the progression of disease while optimizing quality of life?

Hope as highlighted in the field of medical oncology has historically been framed within the context of new drugs and new therapies. However, the cost of bringing a new cancer drug to market is estimated at two to three billion USD, and these new drugs potentially carry with them a risk of significant toxicities and nebulous survival benefits. To address this problem, a growing number of researchers have proposed "drug repurposing" (Aggarwal et al., 2021; Benni and Patil, 2016; Fong et al., 2019; Nowak-Sliwinska et al., 2019; Pantziarka et al., 2019; Parvathaneni et al., 2019; Zhang et al., 2020) as an answer to the question of how to fulfill the hope of advancing cancer therapy while addressing the treatment needs of stage IV patients who otherwise forego standard, NCCN-informed management.

Drug repurposing is defined as a means of developing novel cancer therapies from medications already in clinical use for other disease states (Pantziarka et al., 2021). One agent that shows promise as figuring importantly in the quest to manage metastatic cancers with an aim at mitigating treatment-related toxicities is insulin. Insulin is applied routinely in the management of diabetes, however, there is a decades long history of publications and research in the application of this agent within the clinical practice of oncology. In the case of the repurposing of insulin, clinician and family physician Steven G. Ayre coined the term "Insulin Potentiation Therapy" or "IPT" to describe the application of insulin as a biologic response modifier in conjunction with fractionated low dose chemotherapy (Ayre et al., 1986). A Turkish oncology team practicing out of Istanbul have published clinical results on their use of insulin combined with chemotherapy under the term, "Metabolically Supported Chemotherapy" or

"MSCT" (Iyikesici et al., 2016, 2017, 2019, 2020) The purpose of this paper is to perform a review of the literature to map out available publications on in vitro and clinical repurposing of insulin in conjunction with chemotherapy with an aim at informing practice, shaping policy, and prioritizing future research to perhaps fulfill the unmet needs of the significant percentage of stage IV patients who otherwise do not pursue any form of standard management.

Literature Review

The repurposing of insulin within the context of cancer therapy has differed greatly from the research path taken to bring a new cancer drug to market, therefore the available research also looks different. Furthermore, there are clinician and patient stakeholders who are supportive of the applications of these therapies, and there are practicing clinical oncologists who might have no awareness of these modalities, and either have no opinion on the practice of them, or be in disagreement with the practices due to lack of evidence and fear of patient harm. This paper attempts to develop a platform upon which a possible synthesis of the practice of IPT within the larger practice of oncology might occur by identifying gaps in research that would standardize and optimize the practice of IPT to safely, and effectively meet the currently unmet therapeutic needs of stage IV patients living with various cancer diagnoses.

Repurposing Insulin in the Treatment of Malignancy: Background

The collaborative efforts of a Canadian research team through the University of Toronto led to the introduction of a purified form of insulin for human use in 1921. Children with Type I diabetes who otherwise did not survive much more than one - two years beyond their

diagnosis were granted reprieve from a difficult and abbreviated existence. The discovery and worldwide dissemination of insulin was considered a triumph of modern medicine, relieving vulnerable children from what was a certain death. Frederick Banting and John Macleod shared a 1923 Nobel Prize with fellow researchers Charles Best and and James Collip. 100 years later, insulin has improved millions of lives and the story of its development has sparked a "culture of ingenuity and collaboration that continues to change the world" (University of Toronto, 2021).

By 1932, insulin had made its way to Mexico City where Donato Perez Garcia Senor, MD, a surgeon lieutenant in the Mexican military, treated himself for a longstanding, emaciating gastrointestinal illness of unknown pathology. In addition to diabetes mellitus, insulin at the time was indicated for non-diabetic malnutrition. Dr. Perez thought a course of insulin might benefit him, and he began injecting himself before meals. He found that his weight, body mass, and energy levels improved. He hypothesized that if insulin could facilitate the assimilation of nutrients, then it might have a similar effect on the uptake of medications in the body. He conducted an animal study with dogs wherein he concluded that insulin combined with Salvarsan facilitated the uptake of the early antimicrobial across the blood brain barrier, and into the tissues of the brain where it could be most effective in eradicating the bacteria harbored there (Ayre, 2000). Salvarsan, also known as arsphenamine, was an inorganic arsenic antimicrobial, and an accepted treatment at the time for neurosyphilis before the introduction of penicillin (Vernon, 2019). Following this animal study, he administered treatments on populations of people with tertiary neurosyphilis at St. Elizabeth's Hospital in Washington D.C., Austin State Hospital in Austin, Texas, and the San Diego Naval Hospital in 1944. This 1944 visit to the United States resulted in the article "Insulin for Everything" in the April 10, 1944 issue of Time

Magazine (Time, Inc, 1944). The first application of insulin in conjunction with chemotherapy was applied in 1947 by Dr. Garcia in a patient with squamous cell carcinoma of the tongue, who purportedly went on to live another thirty years (Ayre, 2000). Dr. Garcia and his work with the therapy he termed, "Cellular Therapy to Change the Biophysical, Biochemical Constants of the Blood," was succeeded by his son, Donato Perez Garcia y Bellon MD, and eventually his grandson, Donato Perez Garcia MD.

At the urging of a mutual patient who was pleased with the results she achieved with the Cellular Therapy, Canadian physician Steven Ayre MD travelled to Mexico City in November 1975 to observe and learn more about the practice of Dr. Garcia y Bellon (son of the developer). Thus began a life-long correspondence and collaboration between Dr. Ayre, Dr. Perez-Garcia y Bellon and in turn, Dr. Perez Garcia's son, also named Donato Perez Garcia and also a physician. Dr. Ayre saw his role as that of "scientific liason," working to scientifically validate the empirical, and reportedly successful results being achieved in Mexico with insulin, chemotherapy, and cancer.

Ayre worked tenaciously from the time of his 1975 visit to Mexico City until his death in 2013 to advance the science and clinical practice of Insulin Potentiation Therapy or IPT. This was a term he coined in a 1986 publication in *Medical Hypotheses.* In the early eighties, a research collaborator working with Dr. Ayre came across a 1981 publication that serendipitously reported on the effects of combining insulin and chemotherapy, some forty years after Dr. Garcia applied the combination in a clinical setting in a time before the conception of the randomized control trial and Institutional Review Boards. Alabaster et al. reported that insulin enhanced the cytotoxic effects of methotrexate in MCF-& breast cancer cells by a factor of 10,000 (Albaster et

al., 1981). This study established the groundwork upon which Ayre based his call to the medical community for more in-depth studies, in the hopes that such work would one day lead to a more sophisticated method of applying chemotherapy, and hence substantial improvement in the lives and survival of people living with cancer. He travelled to present his findings at medical conferences in both the United States and abroad. He corresponded over several decades with employees of the National Cancer Institute in an attempt to obtain clinical trial authorization and funding through an IND (Investigational New Drug) application. This was all in addition to his day-to-day work as a community-based Family Practitioner. Through his persistent and ebulliently enthusiastic endorsement of the need to study IPT, there has resulted in a body of both in vitro and clinical publications related to the repurposing of insulin to enhance the cytotoxic effects of chemotherapy.

Methods

As this literature review is specific to the already existing practice of repurposing of insulin in conjunction with chemotherapy in the treatment of cancer, and not an exhaustive analysis on insulin and cancer, article selection involved cross referencing initial publications on the practice by Ayre et al. to map out existing theoretical, in vitro, animal, and clinical publications that have followed.

Google Scholar identified 20 studies that referenced the 1986 Ayre et al. publication, “Insulin potentiation therapy: a new concept in the management of chronic degenerative disease.” 39 studies referenced the later publication, “Insulin, chemotherapy, and the mechanisms of malignancy: the design and demise of cancer” (Ayre et al., 2000) Of these 59

studies, five in vitro studies were selected, one in vitro which bridged to an animal study, one animal only study, and ten clinical studies, which consisted of one randomized, controlled trial, five retrospective cohort studies, and four case study summaries.

Theoretical Publications

In 1986, Ayre et al. published, "IPT: A New Concept in the Management of Chronic Degenerative Disease," coalescing what evidence was available in the medical literature at the time to substantiate the clinical results reported by the Perez Garcia family of physicians. Likewise, the article, "Insulin, chemotherapy, and the mechanisms of malignancy: the design and demise of cancer" enumerates the many discoveries related to the relationship between cancer, insulin, and how a chemo-hormonal approach to treating cancer with fractionated doses of chemotherapy and insulin could be a safe and efficacious method of treatment (Ayre et al., 2000).

Another theoretical application for repurposing insulin proposed by Ayre et al. was to enhance the treatment of HIV with antiretrovirals, using insulin to more efficaciously facilitate passage of medications across the rigorously restrictive blood brain barrier (1989). He proposed that such an action could have implications as well for treating primary and metastatic malignancies in the brain.

In a personal essay appearing on the website of the private practice he founded in 1999, Ayre explained:

> In my class on Medical Ethics back in second year at the University of Ottawa Medical School, I remember reading in my little pamphlet on the subject that were a physician to discover something of value while working to help his patients, it would be his/her ethical responsibility to communicate such findings to his/her medical community (Ayre, 2000).

These early theoretical publications in the journal *Medical Hypotheses* were Ayre's efforts to fulfill what he felt was his ethical responsibility to the profession of medicine by communicating a finding of potential clinical benefit to patients.

In a 2019 missive appearing in the Lancet, Sissung et al. dispute the theoretical basis for IPT, citing numerous biochemical reasons for why insulin would not enhance membrane permeability of cancer cells, and therefore not result in an increased membrane permeability of chemotherapy drugs via this method. Ayre based much of his early theoretical work on a 1981 publication by Alabaster et al. wherein these researchers reported that insulin enhanced the intracellular uptake of methotrexate by a factor of 10,000 in MCF 17 breast cancer cell line. Sissung et al. acknowledge the results of this in vitro study, however, attribute the results to pharmacologic and biochemical characteristics specific to methotrexate and MCF-7 breast cancer cells specifically and do not agree that such results would be expected across cancer cell types and chemotherapeutic agents (2019). The authors of this missive bring up important points of contention. However, there have been a number of in vitro studies on the effects of combining insulin and various chemotherapeutics on a number of cell lines subsequent to the 1981 Alabaster et al. publication, and these all bring up the possibility that other biochemical pathways might be at play to account for an apparent sensitizing effect of insulin on cancer cells to chemotherapeutics.

In Vitro and Animal Publications

Two in vitro studies, both published in 1981, served as a starting point for the theoretical versus empirical explanation for what the Perez Garcia physicians were reporting. Alabaster et

al. found that insulin increased the intracellular uptake of methotrexate in MCF-7 breast cancer cells by a factor of 10,000, and Oster et al. reported that preincubation with insulin in MDA-MB-231 human breast cancer cells enhanced the uptake of ellipticine, a plant alkaloid chemotherapeutic (1981). In subsequent years, there have been a number of additional in vitro studies directly referencing Ayre et al. as the basis for their research, or referencing a work which arose from that line of theoretical query.

The theoretical mechanisms proposed for this phenomenon by these in vitro researchers as derived from their laboratory work varies somewhat, but all conclude that insulin potentially has both a membrane altering effect, and a metabolic altering effect on cancer cells, which could lead to enhanced clinical application of chemotherapeutics. The membrane effect is proposed to result in an increased intracellular concentration of the drug. Agrawal et al. specifically references insulin-receptor mediated endocytosis (2017). The metabolic effect involves recruitment of a greater proportion of cells into the phases of cellular division, notably S phase and G_2, at which point cells are most sensitive to the damaging effects of cell cycle phase specific chemotherapies (i.e. plant alkaloids, antimetabolites, antitumor antibiotics, and epipodophyllotoxins).

In 2003, Jiao et al. assessed the effects of etoposide, 5 fluorouracil, and cisplatin on human esophageal and lung cancer cells in cultures with and without the addition of insulin. They made the observation that cells with high differentiation and decreased metabolic rate tended to be in the G_0 phase of cell activity more often than not, and are therefore not highly responsive to cytotoxic chemotherapeutics. Poorly differentiated cells which are actively proliferating and have a higher metabolic rate are much more susceptible to chemotherapeutic

drugs. This propensity for proliferation and increased metabolic rate is of course a hallmark of cancer in general, and the rationale for applying chemotherapeutic agents with their ability to inflict irreversible damage to cells in the phases of growth and division. When assessing the effects of insulin alone on the cancer cell lines, the authors found that insulin increased the number of cells in S phase. After eight hours this effect waned and was similar to the activity observed in the control group of cells which were left untreated by insulin. The authors ascertained that this effect was therefore reversible. Overall, Jiao et al. reported that insulin enhanced the S phase blocking abilities of etoposide, cisplatin, and 5FU by 80% and the blocking of G_2/M phase by 5FU by 90% in the GEC lung cancer cell line in vitro. The respective metabolic blocking effects of etoposide in the G_2/M phase was found to be enhanced by 10 – 30% in the GLC esophageal cell line. Insulin enhanced the effects of 5FU on the G_2/M phase of the esophageal cells by 20%. The authors report that no measurable difference was observed with insulin combined with cisplatin in the NEC esophageal cell line.

The results of the 2003 Jiao et al. in vitro study was referenced in a following in vitro study assessing the effects of combining insulin and chemotherapy in cancer cell cultures. Zou et al. tested the effects of pretreating human esophageal (Eca 109) and colon (Ls-174-t) cancer cells with insulin prior to addition of 5FU (2007). These authors findings were similar to those reported by Jiao et al. in that pretreating cancer cells with insulin resulted in increased uptake and increased cytotoxic activity in the cell lines tested. As in the Jiao et al. study, the human colon and esophageal cells were pre-treated with insulin prior to administration of the chemotherapy medication, in this instance 5FU. Results were assessed at 24 hours. The authors concluded that the enhanced cytotoxicity observed and measured was a result of a metabolic

effect. Similar to the conclusion drawn by Jiao et al., insulin increases the number of cells in the S phase at any given time, where they are most sensitive to the cytotoxic effects of 5FU (Zou, 2007) They also found that insulin increased the binding of thymidylate synthase (TS) when compared to the cells treated with 5FU alone. As the inhibition of the enzymatic action of thymidylate synthase is an integral means by which 5FU induces cellular apoptosis, this finding was determined to be of additional significance in how insulin can enhance the cytotoxic effects of 5FU (Zou et al., 2007). Obeid et al. conducted a similar in vitro study with 5FU, insulin, and the human colon cancer cell line SW-480. Their aim was to measure the difference in cellular uptake of 5FU between insulin treated, and non-insulin treated cells, and they used the concentration of 5FU in the culture medium to indirectly measure the cellular consumption of 5FU, and avoid measuring changes related to drug metabolism and drug efflux. Their findings support the theory of a membrane altering effect that insulin can confer in cancer chemotherapeutics, in this case resulting in an enhanced cellular uptake of 5FU in the insulin pre-treated cells (2014).

The in vitro findings of the Jiao et al. (2003), Zou et al. (2007), and Obeid et al. (2014) studies were further tested in vivo in a mouse model (Wang et al., 2008). S180 sarcoma, H22 liver, and Eca-109 esophageal cells were transplanted into athymic nude mice. Tumor weight was significantly reduced in all instances of tumor treatment (5FU alone, and FU + low, medium, and high dose insulin combinations), but all were most reduced in the high dose insulin + 5FU combination group. The authors speculated that once again, a metabolic effect was at play. These authors also added a finding of interest in that in the group of mice treated with the combination of 5FU and insulin, circulating levels of IGF-I were decreased compared to the control and 5FU

group alone in the mice transplanted with the sarcoma and liver carcinoma cells. Levels were not significantly lower however in the esophageal transplant group of mice (Wang et al., 2008). This finding is of interest as higher levels of IGF-I are associated with tumorigenesis and treatment resistance. The authors concluded that additional research in this area could yield clinically beneficial understanding on how to modulate circulating levels of IGF-I with the introduction of exogenous insulin to therapeutic benefit, as this could be a result in a downregulation of cancer cell paracrine self-stimulation by IGF-I.

In more recent years, Agrawal et al. (2017, 2019) conducted and published a series of cell studies that likewise observed insulin having a potentiating or sensitizing effect on the cytotoxicity of chemotherapy drugs in vitro. These studies bring several new insights up for consideration. Namely, these authors tested MCF-7 breast cancer cells with the previously studied antimetabolite 5FU, and expanded testing to the alkylating agent cyclophosphamide (2017). They also tested insulin pretreated human colon cancer cell lines Caco-2 and SW480 to a variety of chemotherapeutics: 5FU, oxaliplatin, irinotecan, and docetaxel (2019).

The authors also expand on the theory of how insulin can have a metabolically potentiating effect on cancer cells. In addition to induction of a greater number of cells into the S phase where they are most susceptible to 5FU, Agrawal et al. explore how insulin's chemotherapy enhancing effects on colon cancer cell lines could be a result of the downregulation of the genomic variant phosphatidylinositol-4,5 bisphosphate 3 kinase catalytic subunit alpha (PIK3CA), which plays a role in cell signaling, and growth factor receptor-bound protein 2 (GRB2), which plays a regulatory role in cell proliferation and cell differentiation. This downregulation was measured as a decrease in mRNA concentration for the aforementioned

substrates. This is a more in-depth proposition than previously understood as to possible mechanisms of metabolic effect of insulin on the chemotherapeutic effect of various anti-cancer drugs (Agrawal et al., 2019). The authors conclude that increased intracellular concentration of drug inside cancer cells in the presence of insulin is owed to the membrane altering effects of insulin-mediated endocytosis wherein cells take up glucose, amino acids, electrolytes, hormones, growth factors, enzymes, and plasma proteins through the hormonal actions of insulin (Agrawal et al., 2017). The authors concur with Ayre at al. that the potentiating effects that insulin can have in cancer chemotherapeutics seem to be a combination of a membrane effect and metabolic effect (1986, 2000).

The authors Agrawal et al. included an animal study as an extension of their in vitro work with Caco-2 and SW480 human colon cancer cell lines in a mouse allograft model (2019). The authors concluded that a statistically significant decrease in tumor weight at five weeks was discerned in the group that was pretreated with insulin and then 5FU. Also of note, a marked difference was noted in the number of Circulating Tumor Cells (CTC) between the control group and the groups that were treated with both 5FU and insulin. “Group 4” in the animal study was composed of ten mice treated with 5FU and insulin at the same time (median CTC 100 versus 1400 in control group). “Group 5” was composed of 10 mice who were treated with insulin sixty minutes prior to the administration of 5FU (median CTC 250 versus 1400 in control group). Tissue from the liver and pancreas of the experimentally treated mice (combination of insulin and 5FU) were compared to controls and no morphologically discernible differences were observed (Agrawal et al., 2019).

Agrawal et al. also tested insulin in conjunction with novel thiogylcosides (sulfur containing phytochemicals found in cruciferous vegetables) to assess the outcome and found there to be a suppressive effect (Agrawal et al., 2017).

There is evidence however, that the interaction between cancer, insulin, and chemotherapy is a complicated one. Miglietta et al. performed in vitro studies exposing MCF-7 breast cancer cells to insulin and paclitaxel and found that insulin administered simultaneously with paclitaxel decreased the cytotoxic effects of the paclitaxel. However, paclitaxel exposure *followed* [emphasis added] by insulin resulted in an increased cytotoxicity when compared to the control and was similar to the cell kill of paclitaxel alone (2004).

Volkova et al. arranged an in vitro experiment in an attempt to simulate the tumor microenvironment in obese colon cancer patients receiving chemotherapy, and found that IGF-1 exposure increased resistance of primary adenocarcinoma WiDr human colon cancer cells to chemotherapy. The authors observed mixed results with cell exposure to both insulin and chemotherapy. Insulin sensitized the WiDr cells to both 5FU and oxaliplatin, and increased sensitivity to 5FU in SW620 metastatic colon cancer cell line, but increased resistance in oxaliplatin and irinotecan treated cells. The authors concluded that in vitro, exposure to excess amounts of obesity-related growth factors such as insulin, IGF-1, and glucose resulted in confounding data in respect to chemotherapy sensitivity in different human colon cancer cell lines.

Similarly modelling tumor microenvironmental conditions in obesity and insulin resistance Krajnak et al. exposed a number of different breast cancer cell lines to insulin and low doses of vinorelbine and mafosfamide, such as would be physiologically measured with

treatment via oral metronomic chemotherapy (2021). The cells were treated for 24 hours with insulin prior to administration of the chemotherapeutics and a low dose of insulin (0.01 μg/ml) or a high dose of insulin (10 μg/ml). Chemotherapy response was measured at 72 hours. In all hormone receptor positive cell lines, the addition of insulin increased cell viability as compared to the control when treated with low doses of vinorelbine, which would indicate that insulin conferred a protective property to the cancer cells, or possible chemoresistance. The opposite seemed to occur in the triple-negative breast cancer cell lines, where the authors observed that insulin tended to enhance the cytotoxic effects of low dose mafosfamide. Krainek et al. discuss the somewhat contradictory results they obtained as compared to those reported by Agrawal et al. who reported that insulin had a potentiating effect when administered with cyclophosphamide and 5FU in MCF-7 breast cancer cells (a hormone positive cell line) (2017). Krainek et al. explain that their model was set up to assess the effects of physiological levels of insulin such as might be measured in healthy individuals (0.01 μg/ml) and insulin resistant individuals (10 μg/ml) respectively. They also assessed greatly reduced concentrations of chemotherapeutics as compared to those used by Agrawal et al. as they wished to study the physiological conditions such as would be measured in individuals receiving low dose oral metronomic chemotherapy. They also point out that Agrawal et al. used cyclophosphamide in their study, which must be hepatically converted to its active form in vivo, and they felt that the use of mafosfamide was more suitable to an in vitro study (2021).

Overall, this series of in vitro studies raise some interesting considerations when a model for in vivo application is considered. Factors at play that would determine whether the addition of insulin administered in conjunction with chemotherapy in vivo would either potentiate the

cytotoxic effects or possibly hinder them include dosage and exposure time to insulin, dosage of chemotherapeutics, specific chemotherapy agents used, and histological tumor type.

Clinical Publications

If research around Insulin Potentiation Therapy had proceeded according to the pharmaceutical industry standard for development of new drugs, then more in depth in vitro and animal studies would be performed before proceeding to any human trials. However, as the practice of IPT has proceeded in reverse of these standards (and was implemented in clinical practice for some decades before these standards became well-established) published clinical studies already appear in the medical literature. These studies consist of one prospective randomized control trial, five retrospective cohort studies, and four publications featuring case studies.

In February 1991, Ayre et al. made an oral presentation at the Third International Congress on Neoadjuvant Chemotherapy, Paris, France, February 6-9, 1991 titled, "Insulin plus low dose CMF as neoadjuvant chemohormonal therapy for breast carcinomas." As a result of this report to the oncologic community at large, Lasalvia-Prisco et al. (2004) conducted a randomized control trial based on the premise of Alabaster et al. that insulin enhanced methotrexate uptake in vitro in MCF-7 breast cancer cells. Thirty hormone refractory, estrogen receptor positive breast cancer patients previously treated with fluorouracil, adriamycin, and cyclophosphamide (FAC) were recruited and divided into three groups of 10 subjects. One group received only insulin, one group received only methotrexate, and one group received insulin + methotrexate at 2.5 mg/m^2. Rarely would a control group of cancer patients receive no treatment of known efficacy, however, the institutional ethics committee that approved this study design concluded that

second and third lines of chemotherapy and definitive improvement times in respect to survival was difficult to conclusively discern, and are administered at the cost of harm. Therefore, they considered quality of life and patient preference as relevant determining factors in the approval process. The committee also approved the study based on previous publications allaying fears that the administration of exogenous insulin could possibly stimulate the cancerous process further. In a cohort of 2,720 individuals, Kath et al. determined that the administration of pharmacologic insulin in Type I and Type II diabetes was not found to be a causative agent in the development of malignancy (2000). In another cohort study, serum levels of insulin alone were found not to be a causative factor in breast cancer incidence in a cohort of 7,894 women (Mink et al., 2002).

Lasalvia-Prisco et al. derived a number of conclusions from their study. They used RECIST criteria to evaluate tumor response in the three cohorts, and concluded that tumor growth was inhibited by a statistically significant amount in the insulin + methotrexate group when compared to the methotrexate-only and insulin-only groups. No patients discontinued the study, and no participants were observed to have any detrimental effects as a result of hypoglycemia. The authors also concluded that insulin alone did not promote tumor growth. Compared to the methotrexate- alone group, toxicities were somewhat mitigated in the insulin + methotrexate group with no incidence of anemia, leucopenia, nor thrombocytopenia observed, however 2 individuals in the insulin + methotrexate group compared to three in the methotrexate-alone group experienced mucositis. WHO criteria for toxicities were used. Long-term survival was not an endpoint assessed in this study, nor any other clinical endpoints beyond the duration of the eight week trial. The authors proposed studying the effects of insulin in conjunction with

other chemotherapeutics in other tumor types, as the potential membrane and metabolic alterating effects of insulin on cancer cell activity could be applicable to other malignancies (Lasalvia-Prisco et al., 2004).

Between 2009 and 2012, Damyanov et al. published three articles on the clinical results of their application of Insulin Potentiation Therapy (IPT) as provided at their out-patient clinic in Sofia, Bulgaria. The authors cite their aim in the use of IPT as the mitigation of chemotherapy toxicity while offering an improved quality of life through palliation of disease. One article is a retrospective study of all patients with various tumor types in whom Insulin Potentiation Therapy was administered (2012), a second article is a retrospective cohort study of hormone refractory metastatic prostate cancer patients (2012), and a third article (the first published) is a summary of three case studies wherein patients with severely debilitating metastatic disease experienced qualitative and functional improvement in their performance status, attributed to IPT (2009).

In summary of their three years of applying IPT in 196 patients, Damyanov et al. reported that none of the patients experienced any serious chemotherapy-related side-effects and 85 of 106 (80%) with advanced metastatic disease reported subject improvement in their quality of life. If such results could indeed be duplicated in the 13 -32% of those diagnosed with metastatic disease who do not pursue any form of standard therapy as reported by the American Cancer Society (2019), then such an intervention would be a major advance in the care of those living with advanced cancers.

The authors report that all subjects had pathologically confirmed diagnoses of various solid tumors, and the demographics, clinical characteristics, and location of tumor of origin were all reported. All patients had baseline lab and imaging performed prior to initiating therapy and

tumor response as measured by a variety of quantitative (tumor markers, imaging) and qualitative (subjective improvement in pain and/or functionality) measures after six treatments over three weeks. The most commonly reported side effect was lethargy and fatigue after receiving treatment. Four of the 148 patients who received more than six treatments reported nausea and vomiting the day of therapy, however, this was reported as resolved in all cases by the following day. A reduction in tumor marker levels was measured in 53% of patients who completed exactly six treatments. 3 of 106 patients with advanced metastatic disease experienced radiological remission for 24 months or more (Damyanov et al., 2012). Overall, the practitioner/ authors were very pleased with the results they were able to produce in a patient population with few other viable treatment options. They recommended further study of the therapy through prospective clinical trials (Damyanov et al., 2012).

Later in 2012, Damyanov et al. issued an article summarizing treatment with IPT in a small, 16 person cohort of hormone refractory, metastatic prostate cancer patients. After three weeks and six treatments, eight were categorized as exhibiting a partial response as measured by repeat PSA testing, four were categorized as having stable disease, and four were categorized as having progressive disease. The range in survival was 3 – 30 months, which was calculated as a median of 11.6 months. Symptomatic improvement of greater than 50 % as measured by the Beretta Symptom Inventory was 10 of 16 participants or 62.5%. Despite the small number of participants, the authors were encouraged enough by the outcomes to encourage more in depth research into the potential clinical benefit of IPT (Damyanov et al., 2012).

Sissung et al. references this Damyanov et al. (2012) publication on the prostate cancer cohort bringing up several points of contention as to its justification in promoting IPT as a

defensible practice. The authors point out that that the median survival reported was significantly less (11.6 months) than that reported through clinical trials with standard dose docetaxel (18.9 months) (2019). Sissung et al. also call into question the safety of the therapy received, pointing out that the authors report five of the 16 patients in the cohort received blood transfusions, indicative of grade 3 or higher anemia as defined by the Common Terminology Criteria for Adverse Events (2019). Also, treatment response as defined by Damyanov et al. was not assessed according to the standards of the Prostate Cancer Working Group 2 guidelines, who used nuclear bone scans and decrease in PSA to define response.

All of these points of contention are valid, and highlight the difficulties with a retrospective study reporting on the results of a small, community-based practice, and not a prospective clinical trial organized and executed by a research institution. Clinical trials have criteria for inclusion, whereas community-based practices will accept a patient for treatment if the possible benefits outweigh the harm. Damyanov et al. report baseline alkaline phosphatase levels of between 246 and 5,264 with median levels of 1673 and 1120 in Groups A and B respectively, as measured in mg/ml. These levels are possibly indicative of severe bone and/or liver damage, likely a result of extensive bony metastases, which independent of treatment-related anemia might necessitate blood transfusions due to marrow involvement and anemia of chronic disease. Such severely afflicted patients would also affect the median survival should they initiate therapy at such an advanced stage in the course of the disease. Damyanov et al. highlight the improvement in quality of life experienced by their patients, with few to no serious treatment-related side-effects. The authors concede that larger, prospective trials would have to be conducted to draw any additional conclusions as to outcomes.

Professor and oncologist Bulent Berkarda MD learned of the practice of IPT in a search to provide better outcomes in his stage IV patients at his out-patient, community-based practice in Istanbul, Turkey. He also expanded the scope of his oncology practice to include diet, nutraceuticals, and hyperthermia (Chemothermia, 2018). Between 2016 and 2020, his team of practitioners published three retrospective cohort studies and two case studies. Two of the cohort studies report outcomes in stage III and stage IV pancreatic cancer (2016, 2020) and one reports on outcomes in metastatic non-small cell lung cancer (2019).

Of the case studies, one was a complete response in a 29 YO woman diagnosed with triple negative breast cancer of the left breast, metastatic to the left axilla, liver, and abdomen. Following therapy with Metabolically Supported Chemotherapy [MSCT – otherwise known as IPT], ketogenic diet, hyperthermia, and hyperbaric oxygen therapy (HBOT) from October 2016 to April 2017, a follow-up PET revealed complete radiological response. The patient went on to undergo a left mastectomy, which identified a "3 cm fibro-hyalinized lesion with no evidence of live cells," indicative of a pathological response. The plan was to continue with MSCT, hyperthermia, ketogenic diet, and hyperbaric oxygen for one year to address potential microscopic disease. Long term follow-up has not been reported (İyikesici, 2017).

The second case report highlights the potential utility of Metabolic Supported Chemotherapy (MSCT or IPT) in the elderly, and in a case where surgery would have had life-long detriment to quality of life. An 81 year old woman presented with bloody stools and was diagnosed with locally advanced stage III rectal cancer. The patient refused an abdominoperineal resection, and underwent a course of weekly MSCT with FOLFOX6 for three months, concomitant with external beam radiation for a total dose of 50.6 Gy in 22 fractions. The

radiotherapy was scheduled five days a week for one month. Additionally, the patient received hyperthermia and hyperbaric oxygen therapy. A PET/CT at the conclusion of the three months of MSCT with FOLFOX6 demonstrated no evidence of disease, and at 27 months follow-up, the patient still remained free of any clinical evidence of recurrence. Though this case study represents results in only one patient, the authors posited that further research was warranted towards the standardization of MSCT, augmented by radiotherapy, hyperthermia, and hyperbaric oxygen therapy. If proven successful, the standardization of such an approach would spare other patients with similar presentations the permanently life-altering effects of an abdominoperineal resection that still carries with it a significant risk of local recurrence (İyikesici, 2016).

İyikesici et al. similarly challenge the current status quo of therapy for unresectable pancreatic cancer, proposing that MSCT may prove to significantly enhance overall survival and quality of life in this patient population, that faces significant morbidity and mortality. In a 2016 retrospective analysis of a patient database, 82.5% of patients with stage III and stage IV who had received MSCT [also known as IPT] were alive at one year. Median survival was 19.5 months and 18 of the 33 patients included in the analysis remained healthy and free of evidence of disease at the time of publication. This study was followed by a report on long-term survival of 25 stage IV pancreatic cancer patients who received either gemcitabine or FOLFIRINOX via the MSCT method [also known as IPT]. The median survival was 15.8 months overall, contrasted with a large, prospective RCT wherein the median survival of the FOLFIRINOX group was 11.1 months and 6.8 months in the gemcitabine group (İyikesici, 2019). Once again, the authors concluded that further research into MSCT [also known as IPT], ketogenic diet, hyperthermia, and hyperbaric oxygen therapy are warranted.

And most recently, İyikesici et al. published the results of a retrospective study of 44 stage IV NSCLC patients who received MSCT [also known as IPT] with paclitaxel and carboplatin, ketogenic diet, hyperthermia, and hyperbaric oxygen therapy. Those with brain metastases were offered radiotherapy. Patients with ALK fusion received appropriate targeted therapy as second-line therapy upon progression following chemotherapy. Median overall survival was reported as 42.9 months. Hematologic toxicities were observed at dosing approaching that of standard chemotherapy dosing levels (paclitaxel of 75 mg/m^2 and carboplatin AUC 2 weekly) with the most common being grade 3 anemia requiring RBC transfusion (10 patients). One patient experienced grade 5 neutropenia which resulted in death. The most common non-hematologic toxicity was grade three fatigue (n=5) and diarrhea (n=8). Only one patient developed grade 3 neuropathy. 4 of the 15 deaths observed during the study period were a result of pneumonia and resultant pulmonary insufficiency. No adverse effects were observed as a result of fasting, hypoglycemia, hyperthermia, or hyperbaric oxygen therapy. In a cohort with impaired ECOG performance status (≥2) and a 40% incidence of brain metastases, the fact that there were no treatment discontinuations and relatively few treatment-related toxicities when compared to standard dose chemotherapy administration, the authors found the outcomes encouraging and propose comparative clinical trials to standardize the MSCT [also known as IPT] method to maximum benefit (İyikesici et al., 2020).

The publications by İyikesici et al. represent an interesting model for assessing safety and efficacy in diverse modalities aimed at palliating disease while maintaining quality of life in stage IV patients. Results are reported within the context of a multi-modality approach (in this case ketogenic diet, hyperthermia, and hyperbaric oxygen therapy), which more closely duplicate

conditions of clinical practice. The treatment parameters such as participants and researchers adhere to during clinical trials almost never reflect the realities of day-to-day clinical practice. There work could potentially serve as a model for assessing safety and outcomes in therapies wherein randomized control trials are not feasible.

Finally, a case study of a patient with adenocarcinoma of the lung metastastic to the brain and bones demonstrates how IPT simultaneously palliates several problems that almost ubiquitously arise in stage IV cancers, namely pain, weight loss, and disease-related symptoms. Liu et al. report on a 64YO male who presented to Changzhou Hospital in Changzhou, China with whole body bone pain, weight loss, fatigue, and a persistent cough. He received one round of standard dose cisplatin and pemetrexed, but experienced no relief from his pain, and a marked increase in his CEA levels. The patient discontinued this course of therapy and initiated insulin-induced hypoglycemia followed by reduced doses of chemotherapy [also known as IPT]. After 3 sessions of IPT, the patient experienced relief from his pain. After two months, his coughing was markedly improved. At approximately four months, the patient was noted to have gained five kilograms as compared to admission. Aside from one incidence of grade 1 anemia, the authors report that the patient experienced no adverse effects related to the chemotherapy (cisplatin 10 mg, navelbine 10 mg, 5FU 250 mg biweekly) during his course of 56 treatments in total. PET/CT scans at three and eight months of treatment demonstrated continuously reduced F-FDG uptake, and an MRI of the brain at four months of treatment with IPT demonstrated reduction in the volume of the multiple brain metastases. After approximately 2 months of cessation of treatment with IPT, the patient demonstrated emotional changes, and was noted to have widespread soft meningeal metastases via MRI. He received whole brain radiotherapy followed

by a tyrosine kinase inhibiting medication (gefitinib) for approximately 8 months. Progression-free survival following “glucose inhibition with chemotherapy” [also known as IPT] was 11 months and overall survival was 25 months. This study demonstrates how IPT or “glucose inhibition with chemotherapy” can be of significant palliative value in the management of stage IV cancers, either alone or in conjunction with select standard therapies to improve overall survival with an aim at maintaining safety and quality of life (Liu et al., 2017).

Discussion

Randomized double blind, placebo controlled studies with IPT might never be forthcoming. Blinding the experience of hypoglycemia is not safe, therefore a trial would have to be open label. And an insulin-only group such as in the Lasalvia-Prisco et al. (2004) study is not ethical in today’s treatment landscape for metastatic breast cancer. How can study proceed on the safety and efficacy of this therapy to further refine and maximize the potential for benefit? Additional in vitro studies are a consideration, systematically testing different cell lines with different chemotherapeutics from every class of drug. And prospective, phase II study design wherein insulin and chemotherapy dosing is standardized is essential for a broader application of the therapy.

Though IPT is somewhat unique, the problems of drug repurposing still apply. There is no investment financial incentive to launch large-scale prospective trias and Ayre tried for years to secure government funding and failed. Not necessarily because the therapy did not have merit, but because the machinations of government funding are inscrutable. Though this body of

publications is incomplete in respect to definitively answering the question of the safety and efficacy of IPT in treating malignant neoplastic disease, it is a still growing body.

Conclusion

De facto study of a clinical practice already in use requires a different approach than the rigorously systematic path taken to bring a new agent to market. IPT uses no new drugs or agents, and the agents used have been studied extensively in the standard endocrinology and oncology setting respectively. What remains to unfold is an expansion of in vitro and clinical publications all performed with an aim at enhancing efficacy and maintaining the safety of stage IV patients who wish to forego standard available therapies, but nevertheless consent to treat their disease if their quality of life can be satisfactorily maintained. It is very possible that IPT can meet these criteria to fulfill the rather significant gap in palliating metastatic disease to patient satisfaction today. After eighty some years of clinical use, there is still much to be learned and discovered in respect to Insulin Potentiation Therapy or IPT.

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