A Summary of the studies conducted with CV247

Introduction

Several epidemiological studies have shown that people deficient in trace elements such as selenium, copper, manganese, and vitamins, mainly C,D and E, have a higher risk of developing cancer. Retrospective analyses have also found an association with use of salicylates and a lower incidence of cancer.

CV247 was developed as a consequence of these observations. CV247 is a patented combination of 4 well characterised components: Vitamin C, Manganese Gluconate, Copper Gluconate and Sodium Salicylate, the rationale for their inclusion being:

- **Manganese**

Manganese is an essential trace element for normal brain functioning and for many enzymatic reactions, including hexokinase, superoxide dismutase and xanthine oxidase. Superoxide dismutases are part of the defence mechanism against reactive oxygen species, and altered amounts have been implicated in multistage carcinogenesis in both rodents and man

- **Copper**

Copper can adopt distinct states allowing it to play a pivotal role in free radical scavenging. The importance of copper can also be attributed to its role as a co-factor in a number of enzymes that are involved in the defence against oxidative stress, and a deficiency increases cell susceptibility to oxidative DNA damage.

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- **Vitamin C**

Vitamin C (ascorbic acid) is regarded as the most important anti-oxidant in mammalian cells. As a strong reducing agent, vitamin C forms a part of the body's antioxidant defences against reactive oxygen species and free radicals. Ascorbic acid is the single nutrient supplement most commonly used by cancer patients.

- **Sodium salicylate**

Sodium salicylate is non-steroidal anti-inflammatory drug .There is much current interest in these compounds as possible chemotherapeutic agents. The salicylates, in general, are known to be able to exert an anti-inflammatory effect, by direct blocking of prostaglandin synthesis, and by virtue of their inhibition of the cyclooxygenase enzymes, which are induced in response to cell activation by tumour promoters.

In vitro studies investigating the properties of CV247

A preliminary in vitro viability investigation

In this study using the cancer cell line DJD 15/11 and control cell lines, it was found, after CV247 was added to the incubation mix, that it had a direct anticancer activity on the cancer cells.

The effect on the expression rate of genes involved in cancer protection

The expression rate of certain genes that maybe related to cancer cell death (apoptosis)was also investigated using CV247 from which it was found that in particular expression is increased for several important genes involved in the control of cancer devlopment, namely the tumour protein P53 which is important as a tumour suppressor, the cyclin dependent kinase inhibitor P21 which acts as a regulator of the early interphase of the cell cycle and is itself controlled by P53, the protein kinase family (Akt) involved in cell signaling and the protein complex (NFkB) that controls DNA transcription. CV247 did not appear to act via direct induction of the caspase apoptosis pathway and hence suggests that CV247 activated another mechanism that lead to cell death. This preliminary work encouraged further investigation of CV247.

Activity in common cancer cell cultures

The project focused on efforts to demonstrate in vitro that CV247 has anticancer activities using a range of common cancer cell cultures, and to confirm the location of the mechanism of action.

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Four types of malignancies, breast, colon, prostate and lung carcinoma were selected for testing CV247, with three cell lines for each type. The viability and cell toxicity of the cultures was measured by several methods to determine necrotic, apoptotic and viable cells before and after CV247 treatment. The combination of these techniques provided solid scientific evidence because the advantage of the one technique overlapped the disadvantage of the others, and hence the results were reliable. The results clearly supported the benefit of the combination compared with the individual components, and suggested cancer specificity for CV247, notably colon and breast cancers. The reasons for this are not clear but it seems likely that the properties of the combination therapy are unusual. Whereas traditional cytotoxic drugs have a rapid onset, they rarely kill tumour stem cells and often only inhibit cultured tumour cells for short periods. Inhibition for longer periods is preferred and the fact that CV247 maintains tumour cells in one phase of the cell cycle and thus delays or prevents progression into mitosis, is in itself an unusual property when compared to traditional cytotoxic drugs, and suggests prolonged inhibition is likely.

CV247 is now routinely used as part of a chemosensitivity screen at The Research Genetic Cancer Centre which designed to ascertain possible susceptibility of individual patients to different cancer treatments. More than 1000 patient samples have analysed, 13% of whom showed an increase in caspase and cytochrome C, both markers of a positive response, between 25 and 55% in response to CV247

In vivo studies investigating the properties of CV247

Effect on induced lymphoma in mice

This preliminary study, though not able to demonstrate whether any of the proposed mechanisms of action of CV247 are activated, was designed to show whether CV247 possessed any anti-tumourogenic properties on the growth of transplantable thymoma tumours in inbred mice, without the side effects commonly attributed to anti-cancer drugs.

There was no significant difference between experimental and control animals during the early time points, but there was a statistically significant difference in the size of the tumours and also for the weight of the excised tumours, measured later in the study. In addition 4 tumours were too small to measure in the animals treated with CV247, compared with only 1 in the control group. In a number of mice more than 1 tumour grew along the injection needle tract, but this was considerably more frequent in the control group compared with the experimental group. In 3 control mice, tumours could not be excised because they were infiltrating deeper tissues.

At the doses used in this controlled study, CV247 demonstrated a convincing measurable effect on the growth rate of thymoma in mice. CV 247 did not cure the cancer as tumours grew progressively in both the control and treatment groups, albeit more slowly in the latter. There were no attributable side effects suggesting that CV247 is safe.

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Effect on induced lung carcinoma in mice

This study assessed the anti-tumour activity of CV247 and combinations of the formulation's constituents against the highly metastatic and drug resistant Lewis Lung Carcinoma induced in mice. The effect of CV 247 was compared to untreated control animals bearing tumours and positive control animals treated with Gemcitabine.

Macroscopic examination of tumours on excision revealed differences in tumour structure between treatment groups. Gemcitabine treated tumours were slightly smaller and did not appear to contain much fluid. CV247 without copper appeared to have some effect in reducing tumour volume (growth), but significant differences in tumour weights between CV247, Gemcitabine and the untreated controls were observed. This reduction was most marked for Gemcitabine, and though not significant, was more apparent for CV247 than its components, notably sodium salicylate alone. Microscopic examination was less conclusive. All treatments, when compared to the untreated mice, suggested a similar positive, though insignificant, benefit with regard to intra-tumoural necrosis, but not intra-tumoural inflammation.

This study demonstrated potential for CV247 to be an anti-tumour agent. The decrease in final tumour weight indicates a mechanism for tumour reduction that is not necessarily related to tumour volume. Speculatively it may be suggested that an immunological process is initiated that results in break down the tumour core, therefore reducing the weight (but not size) of the Lewis lung carcinoma, a cell line that is particularly aggressive, and which may explain why Gemcitabine did not demonstrate significant efficacy in this model.

Examination of the anti-oxidative effects in rats

In a recent rat study at the Semmelweiss University in Budapest the antioxidant effects of CV 247 has been studied. The toxic effects of heavy metals are largely induced by oxidative mechanisms, and the widely used chemotherapeutic agent cisplatin is known to cause tissue damage, particularly the kidney, as a consequence of the oxidative effects of its main component, platinum. In this study renal damage in the rat was induced by a single injected heavy dose of cisplatin and a blinded biochemical and histological evaluation undertaken comparing the effects of CV247 with a control administered orally for 14 days after cisplatin. Cisplatin induced marked oxidative effects which CV247, but not the control, was found to be able to reduce, by scavenging the cisplatin induced free radicals. In addition metal analysis found that there was a significant decrease in the concentration of the toxic heavy metals, lead, platinum and tin in the kidney following treatment with CV247. The inclusion of copper in CV247 is particularly beneficial as cisplatin treatment expresses the renal transporter mechanism for copper in the kidney, which is down regulated by CV247 partly inhibiting platinum uptake. These resultant decreased concentrations of heavy metals reduced the oxidative stress in the kidney.

Cisplatin also increases the concentration of iron in the liver, which may induce free radical reactions. Treatment with CV247 was observed to decrease these concentrations and hence reduce free radical levels. A non-significant improvement in the overall reducing power of cisplatin treated liver samples after CV 247 follow-up treatment was also seen. The severely reduced diene content of the cisplatin treated livers was significantly restored after CV 247 follow-up treatment apparently due the prevention of lipid peroxidation.

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Use in veterinary Practice

An open clinical assessment of CV247 for the treatment of cancer in dogs

This open study was undertaken in a single veterinary practice. All cases were independently monitored by the named Veterinary Surgeon of the Imperial Cancer Research Fund. A total of 51 formally diagnosed dogs were monitored in the study. The duration of treatment ranged between 0.5 and 25 months. No adverse effects were reported.

During the study a total of 17 tumours were observed to have regressed. In addition of the 51 dogs that form this study 38 (74.5%) exhibited an increase in their life expectancy beyond that which would have been a "reasonable expectation" at the time the prognosis was made.

Case notes illustrated rapid improvement in the well being of the majority of animals following treatment with CV 247. Of the 51 dogs treated, 2 had inadequate records, but only 4 dogs showed no improvement in quality of life scores, leaving 45 (88%) that showed an improvement. A quality of life rating of 8 and above (good or better) is considered the normal life style for the majority of animals whilst a rating of 10 is considered excellent and provides a life style not previously seen in the animal. 34 of the 51 dogs recruited had a total score of 8 or above, and of those, 6 had a quality of life score of 10.

The Veterinary panel met as a body and considered each case in the study to assess the overall effectiveness of treatment. They concluded that CV247 treatment was successful in 19 dogs and was a qualified success in a further 18, that is over 72% of the dogs treated.

A Clinical Study of CV247 for the Treatment of Cancer in Dogs

The study was designed as an open label evaluation of the effectiveness and tolerability of CV247 in dogs with uncontrolled progressive malignant disease over a 6 month period though the dogs could remain on the medication indefinitely, if benefit continued.

A total of 53 dogs were recruited into the study, the primary objective of which was to evaluate any quality of life changes in the recruited animals, all of whom had a guarded or poor prognosis on entry, and for whom, euthanasia would often have been the recommended course of action, so as to avoid unnecessary suffering. A comparison of the scores between entry and study completion suggests that CV247 promoted an improvement in quality of life in over 70% of the dogs recruited into the study, sometimes significantly so, even if only for a brief period before the animal succumbed to their disease. For many dogs, however, this improvement in well being was apparent for many months, and though estimates of life extension was not undertaken on entry into the study, it appeared that lives were extended in a few animals. Of more significance with regard to the study objectives, was an evaluation of disease stabilisation. This was assessed by each investigator, taking into account any changes in quality of life and tumour regression. The latter was determined at intervals, either by physical measurement, tumour palpation, and/or clinical symptoms. The assessment of disease stabilisation was subsequently independently evaluated by a veterinary expert, the outcome of which, showed that disease progressed in 15 (28%) of the dogs recruited, was stabilised in 25 (47%), and was found to regress in 13 (25%) of the animals treated with CV247. There did not appear to be a trend with regard to dog type, gender, age, disease type or prognosis. During the course of the study, CV247 was found to be a safe drug with no reported adverse effects.

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Use in Clinical Practice

A Preliminary phase II study of CV247 in patients with malignant disease.

This pilot study evaluated the tumour static properties of the formulation CV247 in 37 patients with progressive malignancy at trial entry. Patients had been extensively pre-treated or had diseases in which the benefits of starting conventional treatment were uncertain. 14 had prostate cancer, 2 ovary, 12 colorectal, 2 breast, 7 miscellaneous. The treatment was well tolerated, with enthusiastic dietary compliance.

Of the 2 patients with rapidly progressing ovary cancer, 1 stabilised for 8, the other for 14 months at the time the study was terminated. Of the 7 patients with progressive early prostate cancer, 6 had PSA stabilisation and improved quality of life. These impressive results justify a planned randomised double blind trial comparing CV247 & diet versus salicylates alone. 28 patients had no prolonged worthwhile response but all had been heavily pre-treated with either or both radio- and chemotherapy. There were no drug related serious events. Some patients complained of mild abdominal discomfort, poor appetite, and bad taste. Serum copper/manganese levels remained normal.

A study to compare the safety and efficacy of CV247 and Sodium salicylate for the treatment of early stage progressive prostate cancer

All patients recruited into the study had evidence of progressive cancer, which would normally be managed by a “watch and wait” programme and would be patients who would usually expect to demonstrate continuing and often accelerating disease progression, with few spontaneous remissions. The design of the study was to determine the period of disease stabilisation that patients would enjoy when treated with one or other of the 2 test medications assigned randomly. The duration of the study was set at 12 months though patients who were adequately stabilised at the end of this period were entitled to continue with the randomised medication indefinitely. A total of 110 patients were recruited into the study at the end of which 38 patients (34.5%) had been stabilised for between 12 and 34 months in the double blind phase. 21 (55%) of these patients had been randomised to sodium salicylate and 17 (45%) to CV247. A further 10 patients were stabilised for 10 months (6 on CV247 and 4 on sodium salicylate). At the end of the study 10 patients were still being treated in the double blind phase. A total of 13 of the 38 patients stabilised for 12 months or more had an overall decrease in PSA levels during the period of stabilisation (8 on sodium salicylate and 5 on CV247). Examination of the treatment “failures” found that 42 patients (38%) were withdrawn after only 4 months or less in the study. However the majority of these (78.6%) had been pre-treated at an earlier stage following diagnosis of prostate cancer including radiotherapy and chemotherapy. Further examination of the difference in periods of stabilisation when comparing pre-treated with non pre-treated patients revealed that the mean treatment period for all patients on CV247 was 7.4 and 11.3 months respectively, whilst the figures were 6.4 and 12.9 months for those patients randomised to sodium salicylate. The adverse event (AE) profile was similar for both treatments. Dyspepsia and nausea were the most common AEs for both treatments. Increased manganese levels were recorded for 6 patients on CV247 and for 9 on sodium salicylate, the reason for the increase whilst on sodium salicylate is not clear.

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It is not known whether the success rate of the 2 treatments would have been greater had the patients recruited all been previous treatment naïve, nor whether the trend would have been similar when comparing the 2 treatments. Nonetheless the evidence would strongly suggest that benefit was derived by a significant number of patients with early stage prostate cancer, and that within the limitations of this study design, that similar benefit was derived from both treatment options. Subsequent in vitro studies at RGCC have suggested that prostate cancer may not be the most responsive cancer to CV247.

A study to ascertain if CV247 influences quality of life and malignant progression in patients with cancer who have completed all available conventional treatment

A total of 36 patients were recruited into this 6 month open prospective study, during which patients undertook monthly quality of life assessments. All patients had a documented history of late stage progressive cancer, which in several cases, notably breast and prostate cancers had metastasized to involve typically the brain or bone. The range of cancers presenting was varied: breast (7), prostate (7), mesothelioma (5), ovarian (3), lung (3), rectal (2) and 1 each of cervical, Non-Hodgkins lymphoma, thymoma, fallopian, bladder, colonic, myeloma, pancreatic, and basal cell.

A total of 12 (33%) patients completed 6 months treatment with CV247, 3 of whom presented with breast cancer, 5 with prostate, and 1 each of the patients with mesothelioma, NH lymphoma, ovarian and thymoma. Seven of the 12 patients have continued treatment for more than 12 months. Withdrawals from treatment were usually after the first assessment (11 patients) and were often because the patient decided for a variety of reasons that treatment with CV247 was not their preferred treatment option. The primary end-point for efficacy was Quality of Life based upon the utilization of a self scored, validated (EORTC) questionnaire. Of the 25 patients who continued after the initial assessment, 12 had no change (+/- 1) in their combined total global health and quality of life scores, 3 had decreased scores and 10 (40%) had an improvement. For 11 of the 12 patients who completed the 6 month study, the mean combined score improved, albeit marginally possibly because the type of highly motivated patient typically presenting, gave a false impression of their true health status, particularly on study entry.

There were no serious adverse events. One patient withdrew due to a “feeling of bloatedness”, one due to constipation, 3 because of indigestion and one reported “feeling drowsy”.

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A Summary of Compassionate Use of CV247 over Five Year Period

109 patients with confirmed cancer have been treated with CV247 on a compassionate basis, under the medical supervision of Dr R Taylor. No formal rating scales were used during assessment, just objective reports from the patients about improvement in symptoms. No blood tests were done, but any recently reported blood tests performed by other Health Care Professionals were recorded and documented where possible. The patient cohort had an age range from 14 to 85. There were a huge range of different tumours and patients were seen from shortly after diagnosis to some who were referred in the terminal stages. A simple 10 point assessment grading system was developed.

31 patients were only seen once or twice, with a treatment duration of less than 3 months. No global assessment has been done on this group. In this group most people did not progress with treatment as they were too far advanced in their disease or could not cope with the journey. Of the remaining 78 patients, 48 (61%) were on CV247 between 3 months and 1 year, 13 patients (17 %) were on CV247 for 1 – 2 years and 17 patients (22 %) have been taking the CV247 for 2 years or more. 31 of the 48 patients showed a significant symptom improvement or disease stabilisation, beyond what was expected and which could often not be accounted for by other modes of treatment. Four of this group showed signs of remission. All the patients treated for more than 1 year improved, with 17 patients showing signs of remission several of whom had never had any conventional treatment with chemotherapy or radiotherapy. In the absence of sequential objective testing it is difficult to definitely conclude that symptom reduction was indicative of disease stabilisation, but in a significant proportion of patients on CV247 for longer than 3 months, there was an improvement in symptom burden, general quality of life and in several patients with lung cancer, significant life extension beyond normal prognostic projections. Adverse events were minimal. The only people who found the medication really hard to tolerate were those who were already in the very terminal phase with difficulty eating and drinking generally.

CV247 CV THERAPY