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Suppression of squamous cell carcinoma in hairless mice by dietary nutrient variation

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Abstract

In experiments involving the induction of squamous cell carcinoma in 1846 hairless mice that were maintained on a wide variety of diets, it was found that those diets with the least optimum balance of nutrients had the greatest inhibitory effect on growth of cancer. Rate of onset and severity of tumors was caused to vary over a 20-fold range by means of dietary balance alone. These experiments suggest that dietary variation in general and intentional malnutrition in particular should be given special attention in the control of existing cancer in humans.

Keywords: Carcinoma; Diet; Vitamin C

1. Introduction

Research concerning the minimization of incidence and severity of cancer in humans usually involves experiments directed toward four objectives: (1) reduction of initial production of cancer cells by metabolic or exogenous carcinogens; (2) increase in systemic resistance to the establishment of initial cancer cells as growing tissues; (3) reduction in rate of growth of cancer tissues once they have been established; and (4) techniques for selective destruction of cancer tissues.

During recent decades there has been increasing recognition that the relative and absolute amounts of ordinary nutrients in the diet can have important effects on all four of these objectives. Nutritional experiments are, however, laborious, time-

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consuming, empirical, and often practical only in animal systems, so progress has been slow. This will likely continue to be the case until techniques are developed for the quantitative measurement of human health [1].

There has also developed a general belief in both the research and lay community that there is some optimum balance of nutrients that is ideal for human health [2]. While it is recognized that this balance is subject to biochemical individuality [3], individual differences in nutrient needs are even more difficult to determine than are species optima.

Diet design depends upon specific needs. For example, optimum diets for longevity, intellectual performance, reproductive performance, or athletic performance are probably different and age dependent [4–6].

With respect to cancer, there has been a tendency to believe that there is some 'good' diet that confers optimum resistance to cancer. Advocates of individual nutrients widely argue that their individual recommendations minimize cancer mortality as in, for example, the statement "My present estimate is that the incidence and mortality from cancer could be decreased by 75% by the proper use of vitamin C alone, starting out with taking vitamin C prophylactically" [7].

It is likely, however, that dietary optima for the four anticancer objectives listed above may be quite different. It is possible, for example, that diets which increase systemic resistance to the production and establishment of cancer cells as growing cancer tissue may be counterproductive for the inhibition of growth of established cancer tissue.

Three sets of experiments on nutrition and cancer in hairless mice were designed, directed and evaluated by one of us (ABR) with substantial ideas and help contributed by another (AH). A fourth set of these experiments was carried out by one of us (FCW) in collaboration with the other two (ABR and AH). The four experiments comprised 38 different diets with a total of 1846 mice.

The experimental results are remarkable for three reasons. First, the rate of growth of cancer varied over a 20-fold range as a function of the amounts of ordinary nutrients. Second, the most cancer retarding diets were those for which anecdotal accounts exist for similar retardation of human cancer [8]. Third, the overall results suggest that those diets that would conventionally be considered least suitable for ordinary mouse and human nutrition caused the greatest inhibition of cancer growth.

2. Materials and methods

2.1. Induction of squamous cell carcinoma

Hairless, non-athymic female mice SKh-hr strain were obtained (Experiment sets 1–3) from Temple University Health Sciences Center, Philadelphia, PA at ages 40–60 days. For the fourth experiment, similar mice (HRS/J strain) were obtained from Jackson Laboratory.

These mice were equilibrated for 15 days on specific diets. They were then subjected to five daily suberytheral doses of UV radiation each week for 15 weeks according to the procedure of Black and Chan [9]. Total radiation was about 135

joules/cm. Squamous cell carcinoma lesions began to appear at the end of this period. At 14-day intervals during Experiments 1–3, these lesions were counted and scored on a five-level severity scale where 1, 2, 3, 4, and 5 designated lesion sizes 0–1.9 mm, 2.0–3.9 mm, 4.0–5.9 mm, 6.0–9.9 mm, and 10 mm and larger, respectively. In the fourth set of experiments, overall lesions on each mouse were graded on an increasing scale of severity from 1 to 40.

Care was taken to rotate the cage position of the mice during radiation so that uniformity of UV exposure was achieved. For the first three experiments, a semiautomatic apparatus for quantitative UV exposure was constructed and used. For the fourth set of experiments, a fully automated and improved version of this apparatus was constructed and used. The mice were housed five to each cage in self-washing stainless steel cages. These cages were alternated and rotated in the cage racks, so that all environmental variables in the laboratory were averaged over all groups equally. A surplus of food and water was offered to all mice at all times.

It is important to note that this procedure resulted in substantial numbers of squamous cell carcinoma lesions in mice receiving each of the 38 different diets and that no significant decrease of number or size of cancer lesions over time within the individual experimental groups was ever observed. Such lesions seldom metastasize. The lesions were restricted to the UV-exposed backs of the animals. The rate of cancer growth was substantially different with various diets, so there were times during these experiments when it could have been incorrectly concluded that some diets were markedly preventing the initiation of cancer. At certain times, mice with some diets were afflicted with severe and wide-spread lesions, whereas those receiving other diets appeared almost lesion-free. Eventually, however, nearly all of the mice developed squamous cell carcinoma. For these reasons, it is likely that our results relate primarily to the rate of growth of cancer lesions rather than to the prevention or destruction of cancer. At the end of each experiment there were some mice without visible lesions especially in the low cancer growth groups. Therefore, these studies may have some relevance to cancer prevention.

2.2. Experiment 1 diets

Fifty control irradiated mice received Wayne Lab-Blox mouse chow (WLB) with 24% protein, 4% fat, and 4% fiber primarily from corn, wheat, soybeans, fish, whey, yeast, and vitamin and mineral supplements. 50 Black mice, in a repeat of the Black and Chan experiment [9], received WLB supplemented with 0.5 g vitamin E (DL-α-tocopherol acetate), 12 g vitamin C, 5 g butylated hydroxytoluene, and 1 g glutathione per kg of food. (WLB contains 0.035 g/kg of vitamin E.) Fifty mice received WLB supplemented with 12 g ascorbic acid per kg of food. Fifty mice received WLB supplemented with 0.5 g vitamin E per kg of food. Fifty mice received WLB and a water supply consisting of 1 part sea water to 4 parts fresh water. Fifty mice received WLB supplemented with 9.9 g nutrient oil per kg of food. The nutrient oil was manufactured by R. P. Scherer Corp. and contained 1917 IU vitamin A, 230 IU vitamin D, 3.8 mg thiamine, 1.36 mg riboflavin, 413 mg vitamin C, 13.5 mg niacinamide, 4.0 mg vitamin B-6 HCl, 13.34 mg aminobenzoic acid, 33.1 mg calcium pantothenate, 0.34 mg folic acid, 174 μg vitamin B-12, 35 IU vitamin E, 7 μg vitamin

K-1, 0.27 mg chromium, 0.020 mg copper, 1.0 mg manganese, 0.0033 mg molybdenum, 2.67 mg zinc, 73.1 mg choline bitartrate, 66.7 mg inositol, 25.3 mg biotin, 13.3 mg rutin, 66.7 mg calcium gluconate, 0.94 mg ferrous fumarate, 26.7 mg magnesium oxide, 53.3 mg monobasic potassium phosphate, 0.1 mg potassium iodide, 1.34 μg selenium dioxide, 3.34 mg sulfur, 4.37 mg beta carotene 24%, and 292 mg linoleic acid per unit of manufacture. These diets were fed as powdered mixtures of WLB meal.

2.3. Experiment 2 diets

Sixty control irradiated mice received WLB. Forty-five Miquel mice received WLB supplemented with 1 g *p*-chlorophenoxyacetate tocopherol and 1 g magnesium thiazolidine carboxylate per kg of food. Five groups of 45 mice each received WLB supplemented with 3.00 g, 6.00 g, 12.0 g, 24.0 g, and 48.0 g ascorbic acid, respectively per kg of food. Five groups of 45 mice each received WLB supplemented with 0.125 g, 0.250 g, 0.500 g, 1.00 g, and 2.00 g of vitamin E, respectively per kg of food. These mixtures were pelleted before feeding as were the WLB diets for Experiment 3. Forty-five (FaV) mice [8] received a rotating diet of raw fruits, vegetables, seeds, and grass as follows: Monday, a mush of 15 g wheat grass juice, 100 g water, and 1000 g peeled bananas (BWG); Tuesday, washed and cored apples and pears; Wednesday, BWG; Thursday, washed and cored apples and pears; Friday–Sunday, washed and cut carrots and tomatoes with shelled sunflower seeds. All foods for this group had been grown without use of industrial fertilizers, herbicides, or pesticides. We do not suggest that this method of growth is beneficial. It was included in order to duplicate published recommendations for this diet [8].

2.4. Experiment 3 diets

Sixty control irradiated mice received WLB and UV radiation. Eleven separate control mice received WLB but no UV radiation. Three groups of 45 mice each received WLB supplemented with 48.0 g, 96.0 g, and 192 g ascorbic acid, respectively per kg of food. Forty-five mice received WLB supplemented with 192 g sucrose per kg of food. Forty-five mice received FAV supplemented with a blend of 25 g equivalent vitamin C (47% as ascorbic acid and 53% as sodium ascorbate) per kg FaV as above. (A supplement of 50 g equivalent vitamin C per kg FaV was tried, but this diet was fatal for the mice.) Forty-five mice received diet FaV as in Experiment 2. Forty-five mice received WLB supplemented with 0.05 g abscisic acid per kg of food. Forty-five mice received the FaV (fruit and vegetable) diet with 30 g wheat grass juice instead of 15 g as above in order to increase wheat grass consumption. Forty-five mice received the FaV diet without wheat grass. Forty-five mice received the FaV diet without tomatoes. Forty-five mice received the FaV diet without carrots. Forty-five mice received the FaV diet without apples and pears (these received tomatoes, carrots, and sunflower seeds on Tuesday and Thursday). Forty-five mice received the FaV diet supplemented with 200 g of high protein powder per kg FaV. The high protein powder was made by grinding 20 g soya protein powder (Fearn Soya Protein, 91% protein), 85 g sesame seeds, 200 g raw peanuts, and 700 g

sunflower seeds into a powder with a blender. Forty-five mice received an enriched raw foods diet as follows: Monday and Friday, a blend of 150 g spinach, 25 g parsley, 50 g green pepper, 150 g bean sprouts, 150 g alfalfa sprouts, 300 g cabbage, 15 g wheat grass, 250 g sunflower seeds, and 100 g sesame seeds; Tuesday, a blend of equal amounts of bananas and grapes; Wednesday, a blend of 600 g carrots, 15 g wheat grass, 100 g sesame seeds, and 250 g sunflower seeds; Thursday, same as Tuesday plus apples and pears; Saturday and Sunday, carrots, tomatoes, and sunflower seeds.

2.5. Experiment 4 diets

A basal purified pelleted mouse chow was obtained from Ralston Purina Company which contained 43.65% dextrin, 21% casein, 15% sucrose, 5% corn oil, 5.0% lard, 5.0% RP mineral mix #11, 3.0% solka floc, 2.0% RP vitamin mix, 0.2% choline chloride, and 0.15% DL-methionine. Also obtained from Ralston Purina were modifications of this pelleted basal diet which contained 21% casein (C) and 10% ascorbic acid (AA); 5% C and 5% AA; 9% C and 5% AA; 13% C and 5% AA; 17% C and 5% AA; 21% C and 5% AA; 30% C and 5% A; 40% C and 5% AA; and 60% C and 5% AA. The varied weight % of components in these modified diets was adjusted to 100% by Ralston Purina by appropriate changes in weight % of dextrin.

3. Results

3.1. Experiment 1

This experiment reproduced the cancer suppression effect of Black and Chan [9] with a mixture of glutathione, butylated hydroxytoluene, vitamin E, and vitamin C; measured the separate effects of vitamins E and C in the Black mixture; measured the effect of a sea water supplement; and measured the effect of a nutrient mixture of the sort that is popular among vitamin advocates. Table 1 summarizes these results for total lesions and 'severe lesions' which were those of grades 3, 4, or 5 as described.

Table 1
Squamous cell carcinoma with Experiment 1 diets, 2 months after end of UV irradiation

Diet	Number of mice		Total lesions[b]
	Total	Severe[a]	
Control	46	10	133
Black mixture	43	2	77
12 g/kg Vitamin C	44	11	134
0.535 g/kg Vitamin E	47	13	167
20% Sea water in water	48	14	157
Meganutrient mix	48	13	195

[a]The number of mice with at least one lesion of severity 3 or greater.
[b]The sum of all lesions of severity 1–5.

The measurements in Table 1 were the final set taken during Experiment 1. These were taken 12 weeks after the first onset of lesions which was about 28 weeks after the beginning of the 100-day radiation period. The mice were sacrificed at this point to provide space for Experiment 2 mice. Regardless of severity of lesions selected and time of measurement, the condition of these animals was, throughout the 12-week post-onset lesion period, qualitatively and quantitatively similar to the measurements shown in Table 1.

The Black mixture clearly suppressed cancer, 12 g/kg vitamin C was indistinguishable from control, 0.535 g/kg vitamin E and 20% sea water were a little higher than control, and the meganutrient mix definitely enhanced cancer growth as compared to control. Therefore, unless an unusual synergism is present, the suppressive ingredients in the Black mixture were not vitamin C or vitamin E.

3.2. Experiments 2 and 3

Table 2 gives values for the fifth biweekly lesion counts about 2 months after the end of radiation for both Experiments 2 and 3. Note the extraordinary reproducibility of these results as indicated by the control, 48 g/kg vitamin C, and FaV mice which were replicated between these two experiments performed about 9 months apart. These actual lesion counts have not been adjusted in any way between the two experiments. Table 3 gives values for the ninth biweekly lesion counts about 4 months after the end of radiation for Experiment 3. Some of these values are shown in Figs. 1 and 2.

Vitamin C at low doses was found to accelerate the growth of cancer, while at high doses it suppressed cancer growth. Maximum cancer growth rate was achieved at 3 g/kg vitamin C, while minimum growth rate occurred at 96 g/kg.

Two-month values are more reliable for the higher growth rates and 4-month values for the lower. These values allow substantial cancer growth and yet not such extensive growth as to be debilitating to the mice. Using these values, the growth rate of cancer at 3 g/kg vitamin C was about double that of the control diet, while the growth rate of cancer at 192 g/kg vitamin C was about one-fourth that of the control diet.

Raw fruits and vegetables markedly reduced cancer growth. This effect was found to be independent of the carrot, tomato, and wheat grass components, but was partially dependent upon the inclusion of apples and pears. The suppression of cancer was entirely lost when a high protein (and higher fat) mixture of seeds and nuts was added to this diet. When 165 g/kg dry weight vitamin C was added, the raw fruit and vegetable repression of cancer was enhanced. A mixture with 330 g/kg dry weight vitamin C in raw fruits and vegetables was also tried, but this proved rapidly fatal to the mice. Abscisic acid was tested, because some investigators had focused attention upon this ingredient of wheat grass, but no cancer suppression was observed. The Miquel mixture showed little effect. Overall, suppression of cancer by raw fruits and vegetables was about the same as that of 96–192 g/kg vitamin C and did not require subjection of the mice to this near lethal dose of vitamin C.

Since the average American diet contains about 20% sucrose, a corresponding diet

Table 2
Squamous cell carcinoma with Experiment 2 and 3 diets, 2 months after end of UV irradiation

Diet	No. of mice	Lesions in severity ranges 2–5 and 3–5			
		Total lesions		Per 50 mice	
		2–5	3–5	2–5	3–5
Control — Experiment 2	58	54	14	47	12
Control — Experiment 3	60	54	13	45	11
Control — no UV irradiation	11	0	0	0	0
3 g/kg Vitamin C	45	85	28	94	31
6 g/kg Vitamin C	42	65	16	77	19
12 g/kg Vitamin C	45	52	14	58	16
24 g/kg Vitamin C	44	47	7	53	8
48 g/kg Vitamin C — Experiment 2	45	22	3	24	3
48 g/kg Vitamin C — Experiment 3	44	18	2	21	2
96 g/kg Vitamin C	45	5	0	6	0
192 g/kg Vitamin C	45	7	0	8	0
192 g/kg Sucrose	45	33	9	37	10
0.16 g/kg Vitamin E	42	59	11	70	13
0.285 g/kg Vitamin E	45	67	21	74	23
0.535 g/kg Vitamin E	43	49	12	57	14
1.035 g/kg Vitamin E	45	27	11	30	12
2.035 g/kg Vitamin E	44	60	19	68	22
Fruits and vegetables — Experiment 2	44	11	4	12	4
Fruits and vegetables — Experiment 3	40	11	1	14	1
FaV[a] + 165 g/kg vitamin C[b]	34	4	0	6	0
FaV + seeds and nuts	43	74	21	86	24
FaV w/o wheat grass	39	13	3	17	4
FaV + wheat grass	42	11	3	13	4
FaV w/o tomatoes	44	14	2	16	2
FaV w/o carrots	43	12	2	14	2
FaV w/o apples and pears	41	23	5	28	6
Enriched FaV	45	45	13	50	14
Miquel mixture	42	50	14	60	17
0.005% Abscisic acid	44	61	14	69	16

[a]FaV, fruits and vegetables.
[b]Since this mixture of fruits and vegetables averaged 850 g/kg water, the 25 g/kg fruit and vegetable dose of vitamin C was about 165 g/kg equivalent dry weight.

Table 3
Squamous cell carcinoma with Experiment 3 diets, 4 months after end of UV irradiation

Diet	No. of mice	Lesions in severity ranges 2–5 and 3–5			
		Total lesions		Per 50 mice	
		2–5	3–5	2–5	3–5
Control — Experiment 3	56	218	95	195	85
48 g/kg Vitamin C — Experiment 3	43	115	33	134	38
96 g/kg Vitamin C	45	56	13	62	14
192 g/kg Vitamin C	37	68	24	92	32
192 g/kg Sucrose	42	133	47	158	60
Fruits and vegetables — Experiment 3	37	61	24	82	32
FaV[a] + 25 g/kg vitamin C	34	24	1	35	1
FaV + seeds and nuts	37	175	72	236	101
FaV w/o wheat grass	36	71	17	99	24
FaV + wheat grass	42	74	27	88	32
FaV w/o tomatoes	42	81	23	96	27
FaV w/o carrots	40	62	25	77	31
FaV w/o apples and pears	38	130	42	171	55
Enriched RPF	39	136	60	174	77
0.005% Abscisic acid	43	209	74	243	86

[a]FaV, fruit and vegetables.

of 192 g/kg sucrose was studied. This diet mildly suppressed cancer growth rate to about 80% that of the control diet without sucrose.

The vitamin E supplemented diets showed enhancement of cancer growth at low doses and suppression at high doses similar to, but not so pronounced as, that observed with vitamin C. Maximum cancer growth occurred at 0.285 g/kg vitamin E, while minimum growth was at 1 g/kg vitamin E.

Table 3 shows that these effects continued in a qualitatively and quantitatively similar manner as extensive squamous cell carcinoma developed during the third and fourth months after irradiation. The 4-month values are of use in distinguishing differences between diets showing low cancer growth. The 4-month values are, however, biased somewhat in the high cancer growth rate diets by the general health consequences of extensive tumor development.

Four-month values show that the greatest cancer suppression occurred with the raw fruits and vegetables plus 165 g/kg dry weight equivalent of vitamin C. Cancer growth in these mice was about 10-fold less than control and about 20-fold less than that observed in the highest cancer growth diet which was mouse chow plus 3 g/kg vitamin C.

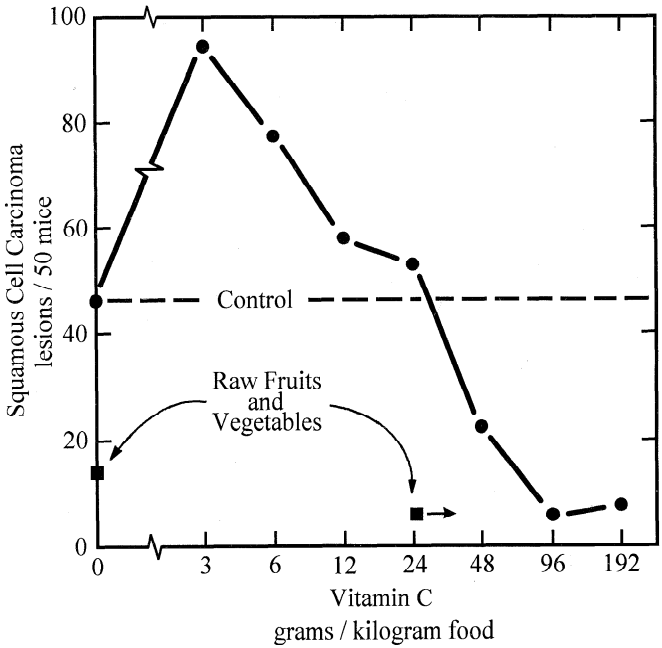

Fig. 1. Sum of severity 2–5 squamous cell carcinoma lesions 2 months after UV irradiation and after normalization to 50 mice for ●, dry Wayne Lab Blox mouse chow plus vitamin C; and for ■, raw fruit and vegetable diet plus vitamin C. Since the mixture of fruits and vegetables was 850 g/kg water, the dry food comparative value of the raw fruit and vegetable diet plus 25 g/kg vitamin C value is about 165 g/kg vitamin C. The maximum cancer growth for the dry mouse chow is at 3 g/kg vitamin C.

3.3. Experiment 4

Table 4 and Fig. 3 summarize these results. Maximum cancer growth occurred at 170 g/kg casein. Cancer growth was about threefold slower at both the extremes of low and high protein, 50 g/kg casein and 600 g/kg casein.

This result is in agreement with the observations in Experiment 3 wherein addition of high protein foods removed the cancer suppressive effect of the raw fruit and vegetable diet as did subtraction of the low protein components of apples and pears. As expected from Experiments 2 and 3, at 210 g/kg protein, cancer growth was reduced by addition of 50 and 100 g/kg vitamin C.

4. Discussion

4.1. General evaluation

These experiments demonstrate, with 38 different diets of widely varying composition, that the rate of development of squamous cell carcinoma in hairless mice is strongly dependent upon diet. Regardless of the experimental system used and the specific diets under study, it is clear that dietary balance should have a high priority in research on the treatment of cancer.

In order to discuss these results in greater detail and with possible significance to

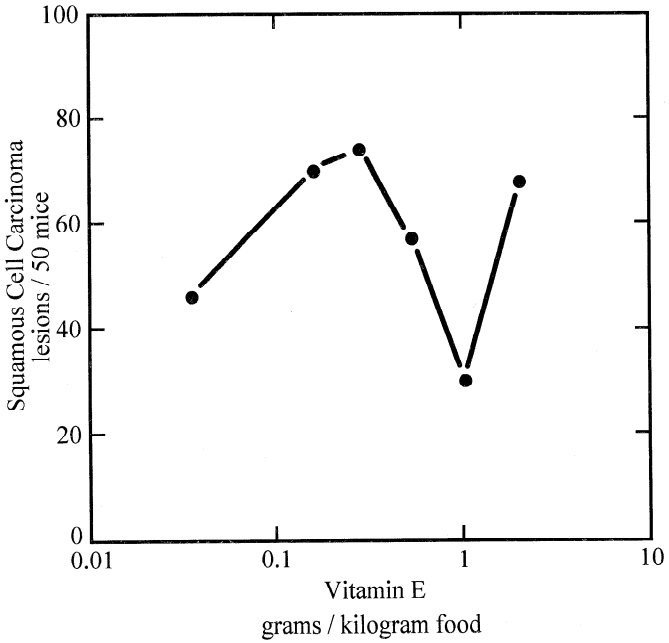

Fig. 2. Sum of severity 2–5 squamous cell carcinoma lesions 2 months after UV irradiation and after normalization to 50 mice for dry Wayne Lab Blox mouse chow plus vitamin E, ●. The vitamin E values include 0.035 g/kg vitamin E in the Wayne Lab Blox mouse chow plus added vitamin E. The maximum cancer growth is at 285 g/kg vitamin E.

human cancer, we must extrapolate these diet compositions from mice to humans. Mice eat several-fold more food per body weight than do humans and have body weights more than three orders of magnitude less. Extrapolations based on absolute amounts of nutrients consumed and functions of body weight are, therefore, uncertain.

Table 4
Squamous cell carcinoma with Experiment 4 diets, 1 month after end of UV irradiation

Diet	No. of mice	Average severity
50 g/kg Protein + 50 g/kg vitamin C	9	11
90 g/kg Protein + 50 g/kg vitamin C	15	20
130 g/kg Protein + 50 g/kg vitamin C	15	26
170 g/kg Protein + 50 g/kg vitamin C	17	31
210 g/kg Protein + 50 g/kg vitamin C	18	21
300 g/kg Protein + 50 g/kg vitamin C	14	19
400 g/kg Protein + 50 g/kg vitamin C	18	18
600 g/kg Protein + 50 g/kg vitamin C	18	12
210 g/kg Protein + 100 g/kg vitamin C	11	19
210 g/kg Protein	14	31

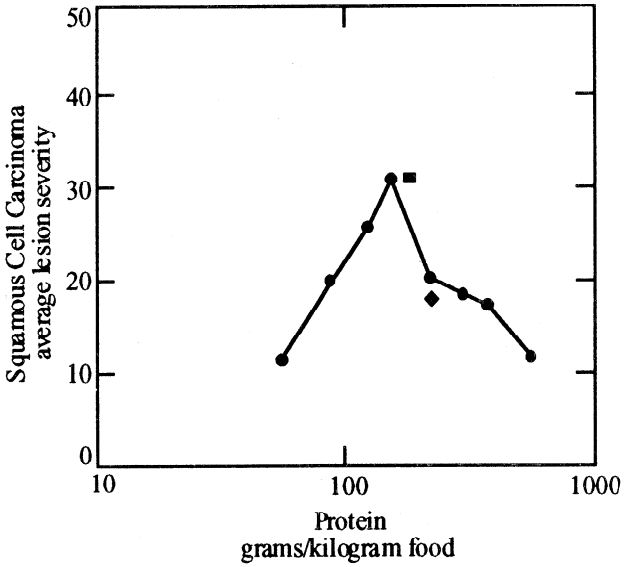

Fig. 3. Average severity of squamous cell carcinoma lesions 1 month after UV irradiation for dry Ralston Purina mouse chow plus casein protein and ●, 50 g/kg vitamin C; ◆, 100 g/kg vitamin C; and ■, no vitamin C. The maximum cancer growth of the 50 g/kg vitamin C values is at 170 g/kg casein protein.

A moderately reliable extrapolation based upon proportions of dietary nutrients can, however, be made. Extensive research on human survival under moderate stress has led to a suggested civilian daily survival ration containing 2600 calories per 748 g from wheat, beans, dried milk, vegetable oil, and sucrose [10]. This diet has 160 g/kg protein. Soldiers under stress require about 3500 calories or about 1 kg/day of this diet.

We suggest, therefore, that the daily human equivalent doses of the nutrients studied here be taken as the numerical amounts in the mouse diets as expressed in g/kg. This assumption is used below. Thereby, 3 g/kg vitamin C extrapolates to an active adult human intake of 3 g/day. For a very sedentary human, this value would be about half or 1.5 g/day.

4.2. Vitamins C and E

There is no doubt that moderate vitamin C supplements accelerate the growth of squamous cell carcinoma in these mice. The increase of cancer growth rate at 3 and 6 g of vitamin C/day in Experiment 2 is consistent with increased cancer growth from the meganutrient mix in Experiment 1 which also included about 3 g/kg vitamin C.

On the other hand, as vitamin C levels approach the lethal dose (we found 300 g/day to be lethal but did not determine an LD50), marked suppression of cancer becomes evident, especially at 100–200 g/day. When the equivalent of 165 g/day vitamin C was added to the raw fruit and vegetable diet, a further suppression of cancer was also seen.

While the reason for this effect is unknown, it is well known that vitamin C oxidation products react destructively with proteins, DNA, and other essential molecules [11,12]. These high levels of vitamin C and its concomitant oxidation products may be depressing the bioavailability of amino acids or other essential nutrients, and they may also have direct cytotoxic effects.

To the extent that these results are relevant to humans, they suggest that cancer therapeutic effects of vitamin C may be limited to very large doses for which no clinical tests have yet been carried out. Low doses of vitamin C may well be counterproductive, although our low dose values should be considered in the light of the fact that mice synthesize their own vitamin C and synthesis may have been suppressed by dietary supplementation. Vitamin E also exhibits values of maximum cancer growth with diminished values at higher and lower doses.

4.3. Protein

The maximum value of cancer growth at 170 g/day of protein (or 85 g/day for a sedentary individual) is consistent with conventional wisdom about protein nutrition. Presumably the tumors found 85–170 g of protein quite adequate for their needs. At 25–50 g, the tumors may have been starved for protein. Levels of 300–600 g of protein were evidently too high.

These protein experiments were undertaken to examine the effect of protein in view of our earlier results with raw fruits and vegetables which suppressed cancer until protein and fat were added. The fact that apples and pears which average only 23 g/kg protein by dry weight enhanced cancer suppression while carrots and tomatoes which average about 120 g/kg protein by dry weight did not is also consistent with this. The apple and pear result is independent of fat. Protein reduction is evidently a major factor in these experiments.

Apparently, low protein in the raw fruit and vegetable diet is a significant factor in its reduction of cancer growth rate.

4.4. Raw fruits and vegetables

This unusual diet was tested as a result of widespread reports that it is effective in reducing cancer growth in humans [8]. The remarkable cancer suppression observed in Experiment 2 led to extensive trials during Experiment 3 in which the original observation was confirmed and variations were tested.

Raw fruits and vegetables seem to suppress cancer primarily as a result of nutrients they lack (or restrict due to dietary bulk) rather than because of unique substances which they contain. The complete reversal of cancer suppression when seeds and nuts are added to the raw fruit and vegetable diet is as spectacular as is the very remarkable cancer suppressive effect of the diet itself.

It must be kept in mind that, when this diet is used in humans, it can only be rigorously utilized for a few weeks or months. A strict raw fruit and vegetable diet without addition of staple seeds, nuts, grains, beans or other foods will not support human life over the long term. When minimal amounts of staples are included [8], the resulting long-term maintenance diet has proved very healthful for individual humans who have been entirely restricted to it for at least 25 years.

4.5. Malnutrition

The results of these experiments may easily be related to numerous hypotheses concerning cancer growth. Obviously much more must be done before this subject is reliably understood. We believe, however, that the very simple hypothesis of ordinary malnutrition should be considered.

Cancer growth was most rapid at the human equivalent daily doses of 1–3 g of vitamin C, 150–300 mg (about 150–300 International Units) of vitamin E, and 80–170 g of protein. Cancer growth was accelerated by a multinutrient mixture typical of human 'megavitamin' supplements and by a rich mixture of seeds and nuts.

Conversely, cancer growth was suppressed by near lethal daily doses of 100–200 g of vitamin C, by 200 g of sucrose, by protein deprivation and protein overdoses, by a raw plant food diet so restrictive that will not support long-term human life, and by large doses of butylated hydroxytoluene and glutathione.

Thus a daily intake of ordinary supplements of vitamin C, vitamin E, and multivitamins, a 'well balanced' amount of fruit, vegetables, seeds, and nuts, and minimal amounts of candy and other sweets — a diet considered healthy for most Americans — would appear to be a harmful diet for a cancer victim; whereas insufficient protein and fat, high 'empty calories' from sucrose, and near lethal amounts of vitamin C would appear good for a cancer victim. In addition, it has long been known [4–6] that rat lifespan is maximized by a nutrient-rich diet early in life followed by nutrient restriction later in life.

Perhaps nutrition during cancer therapy should be viewed as the provision of fuel for a race between rapidly growing young tissues and mature older tissues wherein nutrient restriction or malnutrition may favor the older tissues. The prevailing attitude that 'good' nutrition is the same for everyone may be especially dangerous for cancer victims.

4.6. Conclusion

In experiments involving 38 different diets and 1846 non-athymic hairless mice exposed to UV radiation, we have found that the rate of growth of squamous cell carcinoma varies over a 20-fold range as a function of nutritional balance. This suggests that nutrition should be carefully researched as a useful component of cancer therapy. This conclusion is warranted by the magnitude of differences observed herein and is probably independent of the particular cancer system we have studied or the specific diets we have used.

In addition, these results suggest that, in the case of therapy for already established cancer, special attention should be given to diets that are not necessarily ideal for ordinary good health.

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We dedicate this publication to the memories of Edie Mae Hunsberger and Laurelee R. Robinson. Without their efforts these experiments would neither have been attempted nor completed.

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