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Serum Selenium Levels in Patients with Malignant Melanoma

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The incidence of malignant melanoma of the skin has increased rapidly among white people during the last decade. Although the pathogenesis of malignant melanoma is not clear, epidemiologic data and experimental work indicates that ultraviolet (UV) radiation plays a critical role in tumorigenesis. Recent data implicate peroxidative reactions in UV-carcinogenesis and clearly demonstrate that selenium (Se) confers protection, in part by inducing cellular-free radical scavenging systems and by enhancing peroxide breakdown, thus enhancing the capacity of the cell to cope with oxidant stress. With this in mind, we investigated the Se status of 101 patients with malignant melanoma. Our study revealed significantly lower Se levels in the sera of melanoma patients than of controls. Although patients in all clinical stages had lower Se levels than the controls, patients in stage III (disseminated melanoma) had the lowest levels. Separate analysis of histogenetic subtypes of melanoma revealed that lentigo maligna melanoma (LMM) and superficial spreading melanoma (SSM) had the strongest association with depressed Se serum levels. Our results, showing for the first time that malignant melanoma is associated with low Se concentrations, are consistent with results of epidemiologic studies showing an inverse correlation between serum Se levels and certain cancers. ***Key words: Antioxidative systems; UV-radiation.***

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Selenium (Se) is an essential constituent of the diet of man and other animals. It plays a key role in the activity of the enzyme glutathione peroxidase (GSH-Px) which protects cellular membranes and lipid-containing organelles from peroxidative damage by endogenous peroxides (1). A number of recent studies have demonstrated that Se supplementation can inhibit chemical carcinogen-induced tumorigenesis in skin, liver, colon, and mammary gland, as well as viral-induced tumorigenesis in mouse mammary gland (2–4). The cause of the inhibiting effect is incompletely understood, but it is likely that Se acts through several different mechanisms. Alleviation of carcinogen-induced oxidative damage by induction of GSH-Px and regulation of lipid peroxidation, alterations in carcinogen metabolism and selective toxicity to rapidly dividing cells, are proposed mechanisms of Se-mediated anticarcinogenic effects. In addition, it has been shown that nutritional intake of Se affects the function of the immune system (5).

In humans, epidemiologic studies have indicated a lower death rate from cancer in areas of high vis-à-vis low food Se content and revealed an inverse correlation between cancer occurrence and blood Se levels (6). Several case-control studies in which blood Se measurements were made in specimens obtained after diagnosis have shown lower Se levels in cancer patients (7, 8). This was especially notable for patients with non-melanoma skin cancers, breast cancer, head and neck and gastrointestinal cancers (9). Others have not found similar differences in their subjects, which raises the question whether Se protects only against specific types of cancer (10, 11).

Increasing incidence rates of malignant melanoma in most western countries have been associated with increased sun exposure and experimental work indicate that ultraviolet (UV) radiation plays a critical role in tumorigenesis (12). Production of activated oxygen species in the skin by UV radiation is well documented and evidence suggests that reactions induced by free radicals are involved in the process of skin tumour initiation and promotion (13). In animal models, dose-dependent Se-mediated protection against UV-induced skin carcinogenesis has been demonstrated (14).

In this context, the present study was undertaken to investigate a possible relationship between serum selenium levels and cancer in patients with malignant melanoma.

Table I. *Serum selenium concentration within subsets of the patient group and controls*

	n	Serum selenium (µg/l) Mean ± standard error
Controls	80	81.2 ± 2.0
Sex		
Male	42	79.6 ± 2.2
Female	38	82.6 ± 3.6
Patients	101	69.6 ± 1.2*
Sex		
Male	49	70.4 ± 1.7*
Female	52	68.5 ± 1.8*
Clinical stage		
Stage I	77	71.2 ± 1.4**
Stage II	10	70.0 ± 4.1
Stage III	14	61.1 ± 3.8*

* $p<0.001$; ** $p<0.01$.

MATERIALS AND METHODS

Patients and controls

101 patients, diagnosed with malignant melanoma, were selected for study. Their mean age was 53 (range 20–83) years; 52 men and 49 women. Diagnosis of malignant melanoma was confirmed by histologic examination of tissue obtained by surgery or biopsy. Histopathological staging was performed using the histologic classification system developed by Clark et al. (15). Of the 101 subjects, 10 had lentigo maligna melanoma (LMM), 55 superficial spreading melanoma (SSM) and 36 nodular melanoma (NM). Using the staging system of the International Union Against Cancer (UICC) three different patient groups were defined (stage I: primary tumour; stage II: regional lymph node involvement; stage III: disseminated melanoma). Of the 101 patients selected for this study, 77 (76%) were in stage I, 10 (10%) in stage II and 14 (14%) in stage III.

The randomly selected control group consisted of 80 age and sex matched apparently healthy volunteers. The mean age of controls was 51 (range, 19–83) years; 42 men and 38 women. Patients and controls specifically denied intake of selenium supplements. Because people from different areas may differ in their serum Se concentration, patients and controls were selected from the same part of the Federal Republic of Germany (region of Bonn).

Selenium determinations

The Se determinations were performed at the Laboratorium für spektralanalytische und biologische Untersuchungen, Stuttgart, FRG. Serum Se concentrations were determined by hydride generation and atomic absorption spectrophotometry using a Perkin Elmer 3030 atomic absorption spectrophotometer equipped with a hydride generation system and a selenium electrodeless discharge lamp. Calibration was done against standards with known content of Se (Merck, Darmstadt, FRG).

Data analysis

The differences between datasets were assessed using the Mann-Whitney Test (U-Test). Data are expressed as means ± standard error (S.E.). For correlation analysis the Spearman correlation coefficient was used.

RESULTS

Results of the comparison of serum Se levels in patients with malignant melanoma and controls are given in Table I. Serum Se levels in the cancer patient group ranged between 40 and 91 µg/l (mean 69.6 ± 1.2 µg/l). Se levels of controls were in the range 60–128 µg/l (mean 81.2 ± 2.0 µg/l). The difference of 14.2% was statistically significant ($p<0.001$). The mean se-

Table II. *Mean serum selenium concentration within subsets of the melanoma patient group*

	LMM		SSM		NM	
	n	Se (µg/l)	*n*	Se (µg/l)	*n*	Se (µg/l)
All	10	65.8*	55	68.4*	36	72.6***
Male	4	69.7	28	68.9*	20	73.2
Female	6	61.0*	27	66.7*	16	71.7
Stage I	9	66.3**	43	70.6*	25	73.5
Stage II	1	61.0	4	65.0***	5	75.8
Stage III	–	–	8	55.1*	6	65.6***
Clark						
Level I	–	–	2	62.0***	–	–
Level II	6	65.1***	11	69.8**	6	79.0
Level III	2	66.5	29	68.1*	16	72.4
Level IV	2	65.4	11	70.2**	12	66.1***
Level V	–	–	2	62.0***	2	76.0

* $p<0.001$, ** $p<0.01$, *** $p<0.05$.

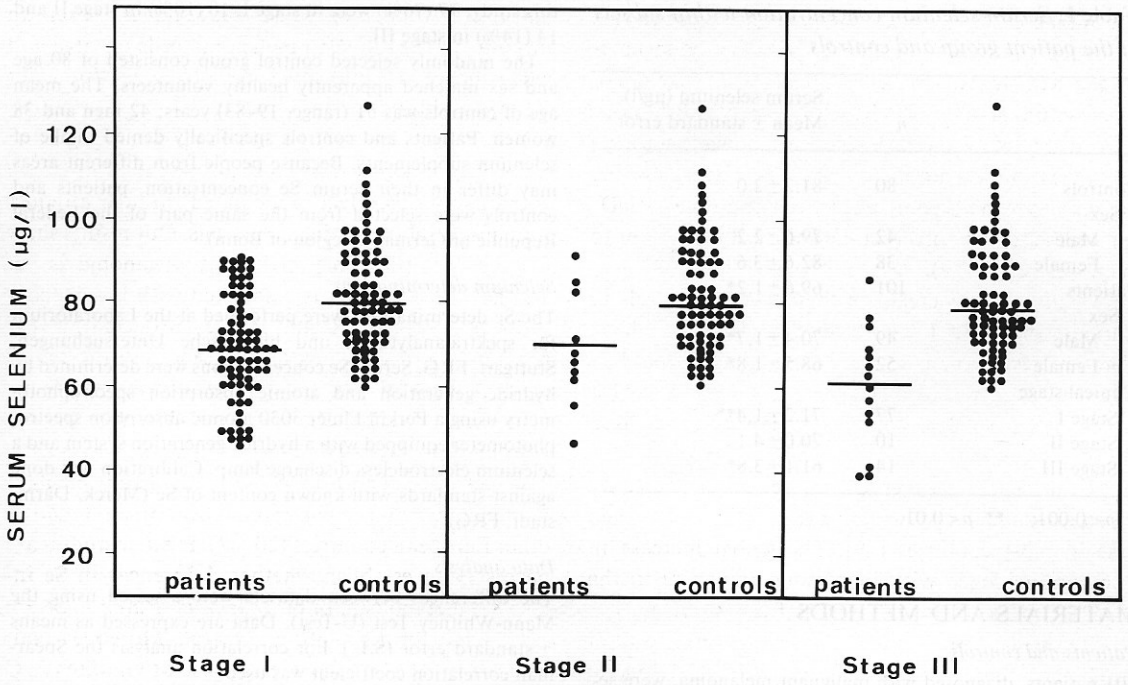

Fig. 1. Comparison of serum selenium concentrations in patients with different clinical stages and controls ($n=80$).

rum Se concentration was 10.4% lower among patients than among controls in men and 18.1% lower in women. The difference in average Se between men and women was 4.8% in controls and 2.7% in patients.

Fig. 1 shows the serum Se concentrations in the sera of melanoma patients who were grouped on the basis of clinical stage, compared with control values. A clear correlation between serum Se concentration and clinical stage was observed. Although patients in all clinical stages had showed lower Se levels than had controls, patients in stage III had the lowest levels. Table II shows the effects of sex on serum Se concentration in different clinical stages. Se levels were 13.4% lower in women than in men in stage III. Sex had little effect, with a spread of $<3\%$ between the means of males and females, in stages I and II. Table II shows the relationship between different types of melanoma examined in categories of sex, clinical stage and histopathologic progression and serum Se concentration. Patients with LMM showed the lowest Se levels (mean 65.8 ± 2.3 µg/l), followed by patients with SSM (mean 68.4 ± 1.7 µg/l) and with NM (mean 72.6 ± 2.1). This difference was greater in females than in males. Analysis on the basis of different types of melanoma showed a clear correlation between serum Se concentration and clinical stage. As the disease progresses, the concentration of Se in the serum decreases. No significant relationship was observed between serum Se concentration and local tumour progression when analysed according to Clark's classification.

DISCUSSION

Our results show that despite the relatively low Se levels of the analysed normal healthy donors from the Federal Republic of Germany, compared with other parts of the world, patients with malignant melanoma in this geographical area manifested a stage-dependent decrease in their serum Se levels. The data suggest that there is a negative correlation between the degree of advancement of malignant melanoma and serum Se levels. Consequently, the decreased serum Se level in patients with stage III might be a consequence of their illness rather than of cancer *per se,* since tumours are known to concentrate Se (16). Furthermore, patients with advanced cancer are known to develop metabolic abnormalities that interfere with

the intake or assimilation of nutrients. Despite this evidence, the selective influence of Se on pathogenetic mechanisms of metastasis cannot be excluded, since serum samples for Se analysis prior to tumour spread are not available. It has been shown that the Se status may modulate the thromboxane/prostacycline ratio in vivo and thereby influence tumour-cell–platelet interactions involved in the ability of neoplasms to metastasize (17, 18).

In contrast, a decrease in their serum Se levels in patients in stages I and II seems unlikely to be a consequence of their illness. Separate analysis of histogenetic subtypes of melanoma revealed that LMM and SSM had the strongest association with decreased Se serum levels.

During recent decades the incidence and mortality rates from malignant melanoma have shown a steep rise in most countries (19). The global increase in tumour incidence with increasing proximity to the Equator speaks for a causal role of UV light.

Interestingly, in northern Europe, mortality rates are higher than in southern Europe (20). The Federal Republic of Germany holds an intermediate position. In addition, several studies have indicated a positive association between melanoma risk and exposure to fluorescent and other lighting sources in indoor workers (21). As well as these possible causative agents, several host factors leading to enhanced susceptibility to UV radiation-induced damage have been associated with the occurrence of malignant melanoma (22).

Recent data implicate peroxidative reactions in UV-carcinogenesis and clearly demonstrate that selected antioxidants can inhibit such events (23, 24). In the 2-stage mouse skin model it has been shown that peroxides, which are capable of generating free radicals, can enhance tumour progression (4). Moreover, dose-dependent Se-mediated protection against UV-induced skin carcinogenesis has been demonstrated (14, 25). It has been shown that UV radiation can deplete skin glutathione (26).

Serum Se levels are correlated with measurements of erythrocyte or platelet GSH-Px activity and Se deficiency results in a decrease in GSH-Px activity and GSH-Px protein concentration (27). Se confers protection against UV radiation damage, partly by enhancing peroxide breakdown. Vitamins E and C also appear to confer protection (28). It has been shown that vitamin E reduces the oxidative damage seen in Se deficiency and a recent epidemiologic study has suggested that Se affords stronger protection to subjects with low serum levels of vitamins A and E (29). Moreover, Se has been shown to exert an inhibitory effect on B16 melanoma cells in vitro and when inoculated into mice in vivo, which indicates selective antitumour activity (30).

In areas where the Se intake is relatively high, the incidence of some forms of cancer tends to be low. Presumably this effect is mediated through a reduction in several free-radical reactions. Our results show for the first time that malignant melanoma is yet another cancer which is associated with low Se concentration. Epidemiologic studies investigating the relationship of Se status and melanoma in different parts of the world are not available at present. However, dietary intake of Se varies in different countries, with known low European areas in Scandinavia (31). This correlates with the fact that malignant melanoma tends to be more common in Scandinavia than in other European countries (20, 32). Further studies are necessary to establish whether differences in Se intake, Se metabolism, or other factors related to Se are responsible for the relation observed. Moreover, concentrations of other substances, such as vitamins E, C and A, with antioxidative and anticarcinogenic properties must be considered. In patients with dysplastic naevi with a very substantial risk of melanoma, especially if they also have a family history of melanoma, attention should be focused on the capacity of antioxidative systems in these subjects.

Results such as those presented in this study imply the need for more careful monitoring of Se levels and that prophylactic dietary Se supplementation may be desirable preventive measures for certain cancers such as malignant melanoma.

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