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Clinical Study

Serum cholesterol in cerebral malignancies

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Summary

Reduced blood cholesterol levels were reported in patients with a variety of malignant peripheral tumors. This fact is likely related to increased cholesterol demand by proliferating tumor cells. The question arises whether this 'tumor-associated hypocholesterolemia' occurs also in patients with brain tumors, and – if it does not – whether its absence can be related to the location of the tumors. We have compared fasting serum total cholesterol levels among three groups of patients: 52 patients with gliomas, 56 patients with symptomatic metastatic brain tumors, and 50 patients harboring malignant tumors of peripheral location but showing no clinical signs of brain metastases. Patients in the last group, despite being – on an average – more age-advanced, had lower total serum cholesterol levels than either the patients with gliomas, or the patients with brain metastases. No difference in the cholesterol levels was found between the two latter groups, and a majority of these patients had borderline or elevated cholesterol levels. This apparent absence of 'tumor-associated hypocholesterolemia' in brain tumor patients may be related to either brain tumors' ability to synthesize cholesterol *de novo* and their reduced dependence on peripheral cholesterol supply, the existence of brain tumor–blood barrier, effect of medications used to counteract brain edema and seizures, or a combination of these factors.

Introduction

Patients with various peripheral malignancies (with notable exception of breast cancer) show decreased total and low density lipoprotein (LDL) blood cholesterol levels [1–4]. This 'tumor-associated hypocholesterolemia' may result from increased cholesterol uptake by proliferating tumor cells. Indeed, increased LDL uptake has been found in cells of many tumors *in vitro* and *in vivo* [5–8], and enhanced LDL receptor mRNA expression compared to normal tissue has been reported in human colonic cancer [9] and leukemic cells [8].

Blood–brain barrier (BBB) is impermeable to macromolecules (e.g., albumin), and its ability to deliver LDL to the brain has not been demonstrated [10]. Brain tumors are hidden behind the blood–tumor barrier (BTB) which is more permeable than BBB, but retains many of its properties [11] and may also limit LDL access to the brain. Noticeably, LDL binding in human brain tumors, while higher than that in the surrounding brain tissue, is low [10,12]. Therefore, the 'tumor-associated hypocholesterolemia' may not occur in brain tumor patients.

In the present study, we have addressed this problem by comparing fasting serum total cholesterol levels among three groups of cancer patients: patients with gliomas, patients with brain metastases from a variety of malignant tumors of peripheral location, and patients with gastrointestinal tract malignancies who presented no clinical signs of brain metastases.

Material and methods

Patients One hundred and fifty-eight consecutive patients admitted to our clinics because of either brain

tumor or malignant tumor of the gastrointestinal tract were included in the study. All patients were hospitalized for at least one week prior to the cholesterol assessment and received standard hospital diet. None of the patients was known to be ever treated for hypercholesterolemia. Fasting serum total cholesterol assay was part of blood chemistry test performed the day before surgery. Cobas Mira S (Roche) analyzer and Cholesterol Enzymatique PAP reagent kit (bioMerieux, Charbonnieres les Bains, France) were used for the assay.

Patients were categorized into three main groups: (i) those with primary peripheral malignancies but no clinical signs of brain involvement, the P group, (ii) those presenting with metastatic brain tumors, the M group, and (iii) those with gliomas, the G group. Brain metastases and gliomas were identified on the basis of post-surgical histology. Gliomas were classified according to the WHO international histological classification of tumors of the central nervous system [13]. The G group included 25 patients with glioblastoma, 11 patients with astrocytoma, 10 patients with anaplastic astrocytoma, 5 patients with oligodendroglioma and 1 patient with pilocytic astrocytoma. The P group consisted of 23 patients with adenocarcinoma of the stomach, 19 patients with adenocarcinoma of the colon/rectum, 7 patients with carcinoma planoepitheliale of the esophagus, and 1 patient with adenocarcinoma of the biliary duct. The M group included 16 patients with lung cancer, 5 patients with melanoma, 4 patients with kidney cancer, 3 patients with breast cancer, 1 patient each with cancer of the stomach, testis, prostate, bone marrow (myeloma) and urinary bladder, and 23 patients with metastases of unknown origin. These latter patients were scheduled for brain surgery because of severe neurological symptoms, yet the location of their primary tumor remained undisclosed. The M group patients were included in the study provided they received no chemo-, radio-, or hormonotherapy (except for short-term glucocorticoid treatment, see below), while some of them had their primary tumor surgically removed prior to the admission to the neurosurgery clinic. No attempt was made to exclude the presence of brain metastases (e.g. by brain CT or MRI scan) in patients who did not show symptoms of brain involvement. In both the G and M group, approximately 50% of patients were administered i.v. dexamethasone, 4–16 mg/day for seven days, to reduce brain edema, and about 20% of the patients were treated with anticonvulsants (mostly phenytoin) for up to 2 weeks prior to the cholesterol assessment.

Statistics Preliminary statistical analysis using Kruskal–Wallis one-way ANOVA by ranks showed no difference in age and serum total cholesterol levels among either the M group patients with gliomas of different histological types ($p = 0.17$ and $p = 0.72$, respectively), or the P group patients with tumors of different localizations ($p = 0.38$ and $p = 0.24$, respectively), or the M group patients with peripheral tumors of different localizations ($p = 0.52$ and $p = 0.10$, respectively). The data corresponding to G and P tumor subgroups represented by single cases (one each in the G and P group) were omitted and those corresponding to M tumor subgroups represented by single cases were combined for this preliminary analysis; data from M group patients with unknown primary cancer site were included as a single additional category. Following the preliminary analysis, the data representing different subgroups were combined within each of the three main groups for further statistical testing. The Kruskal–Wallis one-way ANOVA by ranks was used for verification of the null hypothesis that total serum cholesterol levels are identical among the three main groups of patients. After the null hypothesis was rejected, the method of multiple comparisons [14] was used to determine which groups differ significantly. In both cases, p value ≤ 0.05 was considered significant.

Results

Individual total serum cholesterol levels are displayed in Figure 1, and a summary of these data is included in Table 1. Incidence of lower-than-normal cholesterol levels was markedly lower in G group than in either of the remaining two groups. While the average patient age was significantly lower in G group than in either P or M group, approximately half of G group patients had borderline or elevated cholesterol levels (Table 1). A similar proportion of relatively high total cholesterol levels was found in M group. In contrast, no patient in P group had clearly elevated total serum cholesterol, the highest level in this group being similar to the average values in the remaining two groups. The Kruskal–Wallis one-way ANOVA by ranks revealed that total serum cholesterol levels differed among the three groups ($H[2, N = 158] = 23.92$, $p < 0.0001$). Multiple comparison analysis showed no difference in the cholesterol level between G and M groups (average rank difference: 7.7, critical value: 21.1), while P group

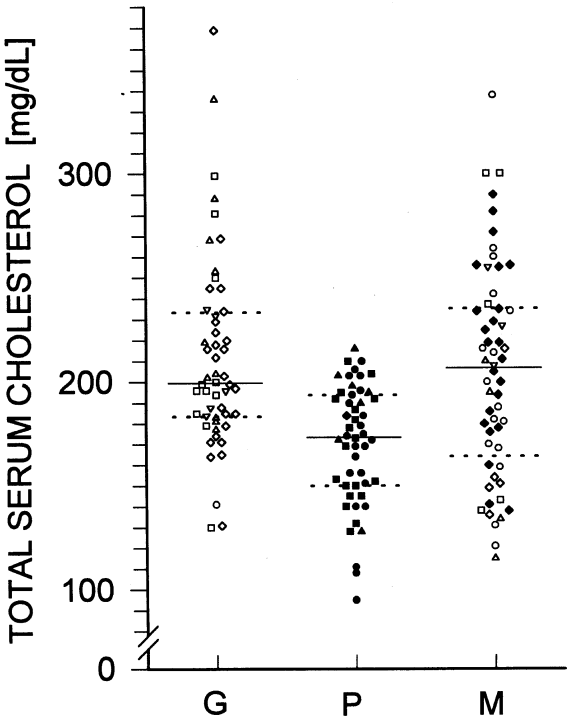

Figure 1. Fasting serum total cholesterol levels in patients with gliomas (group G: pilocytic astrocytoma – open circles, astrocytoma – open squares, anaplastic astrocytoma – open triangles, oligodendroglioma – open reversed triangles, glioblastoma – open diamonds), patients with untreated peripheral tumors showing no clinical signs of metastases to the brain (group P: gastric cancer – closed circles, colonic/rectal cancer – closed squares, esophageal cancer – closed triangles, biliary duct cancer – closed triangles), and patients with brain metastases from tumors of peripheral location (group M: lung cancer – open circles, melanoma – open squares, kidney cancer – open triangles, breast cancer – open reversed triangles, other known locations – open diamonds, unknown locations – closed diamonds). Median values are shown with continuous lines, and percentile P25 and P75 values are shown with dotted lines.

differed significantly from both G (average rank difference: 41.2, critical value: 21.8) and M group (average rank difference: 33.5, critical value: 21.4).

Discussion

There is a large body of evidence showing that cholesterogenesis plays a critical role in the growth of eucaryotic cells including those which have undergone malignant transformation [16–20]. As mentioned before, cells of a variety of malignant peripheral tumors display enhanced cholesterol uptake. Moreover, a rise in blood cholesterol level was reported in colonic cancer patients following curative but not after non-curative surgery [9], and in acute myelogenous leukemia patients during remission [3]. These facts probably reflect increased cholesterol requirement of proliferating malignant cells and indicate that the tumor-associated hypocholesterolemia is a consequence of the malignancy.

Average serum total cholesterol levels (which usually reflect plasma LDL cholesterol) differed among the three patient groups included in this study, being the highest in the glioma patients. Moreover, the apparent distribution of individual cholesterol values differed among these groups. Both the gliomas and cerebral metastases were associated with relatively high incidence of elevated total serum cholesterol, and higher average serum total cholesterol level (and even more so in the context of average age of the patients) than that in patients with peripheral tumors showing no symptoms of brain metastases. Similar incidence of low cholesterol levels in P and M patient groups was most likely related to the presence of the primary tumor and/or metastase(s) of peripheral location in some of M group patients. In general, results of this study suggest that, unlike a number of peripheral malignancies, brain tumors do not cause or are associated with hypocholesterolemia.

There are several possible reasons for this dissimilarity. First, human gliomas, unlike normal adult brain tissue, can synthesize cholesterol *de novo* [21,22]. It had also been postulated that brain tumors sequester cholesterol from necrotic or damaged brain tissue, or synthesize it from precursors entering sterol biosynthesis beyond the early steps [22]. Thus, although these tumors express LDL receptors [12,23] which suggest a potential to utilize cholesterol from external sources, their growth may not result in a meaningful depletion of serum cholesterol. The report of Maltese [22] shows also the presence of the key enzyme of cholesterol biosynthesis, 3-hydroxy-3-methylglutaryl coenzyme A reductase (HMG-CoA reductase), in malignant peripheral tumors. This suggests that these tumors may synthesize cholesterol *de novo*, but the availability of circulating cholesterol does not render them critically dependent on this metabolic pathway. Interestingly, in that report HMG-CoA reductase activity was found in all four metastatic brain tumors studied, two of which showed the highest HMG-CoA activity

Table 1. Fasting serum total cholesterol levels in patients with gliomas (G), patients with peripheral tumors with no clinical signs of brain metastase(s) (P), and patients with symptomatic metastatic brain tumors (M)

	Patient group		
	G	P	M
Age [years]:			
mean ± S.D.	47.0 ± 15.6[a,b]	62.8 ± 12.4	54.8 ± 11.8[a]
range	18–78	25–88	23–80
Cholesterol level [mg/dL]:			
mean ± S.D.	212 ± 47[a]	170 ± 29	205 ± 51[a]
range	130–369	95–217	115–338
Distribution of cholesterol levels[c]:			
low (<140 mg/dL)	2/52 (3.8%)	6/50 (12.0%)	7/56 (12.5%)
normal (140–199 mg/dL)	23/52 (44.2%)	36/50 (72.0%)	18/56 (32.1%)
borderline (200–230 mg/dL)	12/52 (23.1%)	8/50 (16.0%)	14/56 (25.0%)
high (>230 mg/dL)	15/52 (28.9%)	0/50 (0%)	17/56 (30.4%)

[a] $p < 0.05$ *vs*. P group.
[b] $p < 0.05$ *vs*. M group (Kruskall–Wallis one-way ANOVA by ranks followed by the method of multiple comparisons).
[c] lower limit of normal serum total cholesterol range was taken from the cholesterol assay kit instructions; borderline and high limits were taken from Ref. [15].

reductase per mg DNA among all tumors [22]. This latter finding suggests that the ability for sterol synthesis may favor formation of active brain metastases and reduce their dependence on peripheral cholesterol, and thus preclude the occurrence of 'tumor-associated cholesterolemia'. Second, while primary and metastatic brain tumors express LDL receptors at higher levels than normal adult brain tissue [12], the absence of 'tumor-associated hypocholesterolemia' in brain tumor patients may be related to the BTB-related restrictions in the transfer of LDLs – which are the main blood cholesterol carrier – into the brain [24]. Intravenously injected ^{99m}Tc-labelled autologous LDLs accumulate poorly in human gliomas, showing tumor to normal brain tissue ratio similar to that of ^{99m}Tc-labelled serum albumin. However, certain differences in intracerebral distribution and rate of accumulation of these substances indicate that both non-specific and LDL receptor-mediated processes may play a role in LDL uptake by human gliomas *in vivo* [10]. On the other hand, there is a report showing that cholesterol esters are greatly elevated in most human gliomas and surrounding brain areas compared to normal adult brain tissue, and originate mainly from the blood [25]. These findings suggest that plasma LDLs may contribute to glioma cholesterol supply and that the accumulation of ^{99m}Tc-labelled LDLs in human gliomas [10] was hampered by already increased LDL content in these tumors.

The apparent absence of 'tumor-associated cholesterolemia' in patients with primary or metastatic brain tumors could also be related to the medications given to counteract neurological consequences of the brain tumor. Anticonvulsants (mainly phenytoin) are known to increase total serum cholesterol level in children and adolescents [26–29]. However, the data on the effect of anticonvulsants on total serum cholesterol in adults are ambiguous [cf. 30–36]. Moreover, these data relate exclusively to drug administration for periods exceeding many times those in our patients. Similarly, while chronic glucocorticoid treatment elevates total serum cholesterol [37–39], it is not clear whether this effect occurred in our patients who were subject to dexamethasone treatment for no longer than one week.

If, as discussed above, blood cholesterol contributes to the growth of brain tumor, then the potential for elevating serum cholesterol should be given appropriate consideration during the choice of treatment aimed at alleviating consequences of the brain tumor presence, i.e., brain edema and/or seizures.

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